Navigating the Cary Eclipse Online Help

The Cary Eclipse Help system is designed for easy navigation. The idea is that you can find what you are after quickly and efficiently.

So how is this achieved?

Table of Contents Tab

1. Contents tab (Table of Contents)
   Use the Table of Contents to view the titles of the help topics. Topics are grouped together into categories to make navigation simpler.

2. Search tab
   The 'Search' and 'Search within Search' features enable you to locate very specific information. You can refine a search further by using the 'Search within Search' feature. This feature enables you to search only within the results of your initial search - e.g. If you search for 'Cuvettes', you can then select 'Search within Search' and search only in the results displayed in the initial 'Cuvettes' search.

3. Favorites tab
   If you are having repeated problems performing a particular task, you can store the link to that topic in the
Favorites section. This means that you can quickly go to the Favorites list next time you want to use that section of the Help, rather than browsing through the table of contents or using the Search mode.

4. Plus / Minus sign

Click on the plus signs to expand a branch. Click on the minus signs to collapse a branch.

5. Contents pane

When you click on a topic, the contents of that file will appear in the Contents pane.

See Also
Multimedia Help

The Multimedia Help system provides animated help.

Video Clips

Sound and vision reinforce the text based information contained within the help system. Look for the video icon that appears throughout the help system.

Video icon

Whenever you see a video icon such as the one adjacent, it indicates that a video is available. You can click on these icons to run videos.

These video and animation sequences aid in:

- highlighting problem areas associated with installing particular accessories
- providing information about the location of important accessory and instrument components
- make the installation of accessories a quicker and more efficient process

Trouble shooting

If you are having troubles then check the following:

- The CD-ROM labeled "Help & Videos" is in your CD drive.
- You installed all of the video drivers and codecs when you installed Windows.

See Also

Help on the Varian Video Player application
For optimum viewing of the Cary Eclipse software and help your monitor resolution should be set to 800 x 600 pixels (or greater) and High colour (16 bit) (or greater). 16 or 256 color setting may not display the video and multimedia correctly.

To set the monitor resolution:

Step 1
Click the Windows Start button, point to Settings and then select Control Panel. The Control Panel window is displayed.

Step 2
Double-click on the Display icon to access the Display Properties dialog.

Step 3
From the Display Properties dialog, select the Settings tab.

Step 4
In the Desktop area section, drag the sliding bar so that the pixel amount is set to 800 x 600 pixels (or higher).

Step 5
In the Color palette section select High color (16 bit).

Step 6
Select OK. At the prompt, which informs you that your settings will be adjusted, select OK again. When asked if you wish to keep the setting, select Yes.

Step 7
You can then exit the Display Properties dialog and the Control Panel window.
Help is available for all of the Cary Eclipse Software applications. You can press F1 from within an application at any time to display the online help system for that application. Alternatively, click the Help button if available or select Help from the main menu.

Multimedia Video Tour

For a multimedia tour of all the Cary Eclipse Software applications and what they can do for you, click on the button below.

Further Reading

- What is the purpose of each Cary Eclipse application?

Each application that comes with the Cary Eclipse software has a section of the online help system devoted to it. If you would like to go to the help section of a specific application, click on the name of that application below.

<table>
<thead>
<tr>
<th>ADL Shell</th>
<th>Lifetimes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Advanced Reads</td>
<td>Scan</td>
</tr>
<tr>
<td>Align</td>
<td>Simple Reads</td>
</tr>
<tr>
<td>Concentration</td>
<td>System Information</td>
</tr>
<tr>
<td>GLP Administration</td>
<td>Validate</td>
</tr>
<tr>
<td>Kinetics</td>
<td></td>
</tr>
</tbody>
</table>

The application help guides you through the user interface and explains how different fields control different instrument, accessory and software parameters.

Note

By going to a section of the software you are having problems with and pressing F1 — you can get help related specifically to that part of the software.
Help System Icons

Look out for 'Tips', 'Notes' and 'See also' icons throughout the help system. The 'Tips', 'Notes' and 'See also(s)' provide you with or link you to useful information that will allow you to complete various tasks with less effort, more efficiency and less chance of you damaging your equipment but more importantly — ensuring that things are done safely.

These are the icons that represent 'Tips', 'Notes' and 'See also':

- Tips
- Notes
- See also

By following 'Tips' and 'Notes' and reading the 'See also' links, you will become a better Cary Eclipse user.
Printed

<< Back | Back to Start >>

Single-page print (right-click for the options menu)

Right-click on the page you wish to print. The following menu is displayed.

Select **Print**... from the menu. The **Print** options window is displayed.
Select the printer and print options and click **OK** to print.

**Multi-page print (right-click on the Table of Contents)**

Right-click on the Table of Contents. The following window is displayed.

Select **Print...** from the menu. The following window is displayed.
You can print the selected topic or all the topics in the selected heading. What would you like to do?

- Print the selected topic
- Print the selected heading and all subtopics

Select **Print the selected topic** if you only wish to print the topic that is displayed. Select **Print the selected heading and all subtopics** if you wish to print all of the topics that come under the Table of Contents folder or topic that is currently open. Click on **OK**, to display the Print options window, as shown below.

Select the printer and print options and click **OK** to print the topics.
Send away to Varian for a printed manual

If you want a complete printed version of the Cary Eclipse Online Help system, e-mail us at fluorescence@varianinc.com.
About Safety

Your Varian Cary Eclipse instrument and accessories have been carefully designed so that when used properly you have an accurate, fast, flexible and safe analytical system.

If the equipment is used in a manner not specified by the manufacturer, the protection provided by the equipment may be impaired.

Information on safety practices appears throughout the documentation (both hard copy and online) provided with your instrument and accessories. Before using the instrument or accessories, you must thoroughly read these safety practices.

Observe all relevant safety practices at all times.
Lamp module

The lamp is enclosed in a self-contained module. This module contains components operating at high voltages. To avoid electric shock, NEVER disassemble the module.

When operating, the lamp module emits high intensity visible and UV radiation which can cause serious damage to eyes. To avoid eye damage, never operate the lamp outside the instrument.

See also:
Safety Home Page
Ultraviolet radiation

Hazardous UV radiation is emitted by the Xe flash lamp in the instrument. This radiation can cause serious damage to eyes. NEVER look directly at the UV source lamp. ALWAYS wear safety glasses manufactured to an approved standard and which are certified or otherwise warranted to protect the eyes from UV radiation when the lamp is operating and the sample compartment is open.

See also:
Safety Home Page
Panels, covers and modules

You are permitted to remove the top colored cover to:

- Replace the lamp module
- Adjust the lamp module

Any other panels or covers, which are retained by screws on the spectrophotometer and accessories, may be opened ONLY by Varian-trained, Varian-qualified, or Varian-approved service engineers. Consult the manuals or product labels supplied with your PC, monitor and printer/plotter to determine which parts are operator-accessible.

Operators and other unauthorized personnel are permitted access ONLY to the lamp module and the sample compartment of the Cary Eclipse. ALWAYS switch off the instrument and disconnect the mains cord before changing a lamp module.

See also:
Safety Home Page
Other precautions

The lamp module operates at a high temperature, and touching it may result in burns. Before replacing a lamp module that has been operating, switch off the Cary Eclipse and ensure that the lamp module has cooled, or protect your fingers from burns.

Do not block the ventilation grills on the spectrophotometer and accessories. Consult the manuals supplied with your PC, monitor and printer/plotter for their specific ventilation requirements.

Use of the Cary Eclipse system and accessories may involve materials, solvents and solutions, which are flammable, corrosive, toxic or otherwise hazardous.

Careless, improper, or unskilled use of such materials, solvents and solutions can create explosion hazards, fire hazards, toxicity and other hazards which can result in death, serious personal injury, and damage to equipment and property.

ALWAYS ensure that laboratory safety practices governing the use, handling and disposal of such materials are strictly observed. These safety practices should include the wearing of appropriate safety clothing and safety glasses.

See also:
Safety Home Page
Warnings and Cautions

Specific warnings and cautions appear in the online Help where appropriate, and these:

- detail the specific hazard;
- describe how to avoid it;
- specify the possible consequences of not heeding the warning or caution.

**Warning**  A ‘Warning’ message appears in the Help when failure to observe instructions or precautions could result in death or injury. Symbols depicting the nature of the specific hazard are also placed alongside warnings.

**Caution**  A ‘Caution’ message is used when failure to observe instructions could result in damage to equipment (Varian supplied and/or associated equipment).

A ‘Note’ is used to give advice or information.

Read all warnings and cautions carefully and observe them at all times.

See also:

[Safety Home Page](#)
A triangular symbol indicates a warning. The meanings of the symbols that may appear alongside warnings in the documentation are as follows:

- Electrical shock
- Eye hazard
- Noxious gases
- Hot surfaces and cryogenic temperatures
- Fire hazard
- Sharp edges
- Corrosive liquids
- Moving part
- Heavy weight (danger to feet)
- Heavy weight (danger to hands)
- Part can be ejected

The following symbol may be used on warning labels attached to the instrument. When you see this symbol you must refer to the relevant operation or service manual for the correct procedure referred to by that warning label.

See also:
Safety Home Page
Color coding

The indicator light appearing on the Cary Eclipse instrument has been color coded to represent the status of the instrument or accessory:

<table>
<thead>
<tr>
<th>Color</th>
<th>Status</th>
</tr>
</thead>
<tbody>
<tr>
<td>Steady green</td>
<td>normal / standby</td>
</tr>
<tr>
<td>Flashing green</td>
<td>scanning / reading</td>
</tr>
<tr>
<td>Steady orange</td>
<td>start up</td>
</tr>
<tr>
<td>Flashing orange</td>
<td>a problem, may require servicing</td>
</tr>
<tr>
<td>Steady yellow</td>
<td>firmware initialization</td>
</tr>
</tbody>
</table>

See also: [Safety Home Page](#)
The following symbols appear on the Cary Eclipse to provide you with additional information:

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Mains power on</td>
</tr>
<tr>
<td>0</td>
<td>Mains power off</td>
</tr>
<tr>
<td><img src="image" alt="Fuse" /></td>
<td>Fuse</td>
</tr>
<tr>
<td><img src="image" alt="Single phase alternating current" /></td>
<td>Single phase alternating current</td>
</tr>
<tr>
<td><img src="image" alt="CE" /></td>
<td>When attached to the rear of the product, indicates that the product complies with the requirements of one or more EU Directives</td>
</tr>
<tr>
<td><img src="image" alt="Focus" /></td>
<td>Focus</td>
</tr>
<tr>
<td><img src="image" alt="Horizontal movement" /></td>
<td>Horizontal movement</td>
</tr>
<tr>
<td><img src="image" alt="Vertical movement" /></td>
<td>Vertical movement</td>
</tr>
<tr>
<td><img src="image" alt="CSA" /></td>
<td>Attached to the rear of the product, and indicates that the product has been certified (evaluated) to CAN/CSA–C22.2 No 1010.1-92 and VL3101-1.</td>
</tr>
</tbody>
</table>

See also:
- [Safety Home Page](#)
FCC advisory

The following is a Federal Communications Commission advisory:

**Caution**
This equipment generates, uses, and can radiate radio frequency energy and if not installed and operated in accordance with the instruction manual, may cause interference to radio communications. It has been tested and found to comply with the limits for a Class A computing device pursuant to Subpart J of Part 15 of FCC Rules, which are designed to provide reasonable protection against such interference when operated in a commercial environment. Operation of this equipment in a residential area may cause interference in which case the user at his or her own expense will be required to take whatever measures may be required to correct the interference.

See also: Safety Home Page
Cary Eclipse instruments have been designed to comply with the requirements of the Electro-magnetic Compatibility (EMC) Directive and the Low Voltage (electrical safety) Directive (commonly referred to as the LVD) of the European Union.

Varian has confirmed that each product complies with the relevant Directives by testing a prototype against the prescribed EN (European Norm) standards.

Proof that a product complies with the Directives is indicated by:

- the CE Marking appearing on the rear of the product
- the documentation package that accompanies the product containing a copy of the Declaration of Conformity. This Declaration is the legal declaration by Varian that the product complies with the Directives, and also shows the EN standards to which the product was tested to demonstrate compliance.

It is also signed by Varian's Authorized Representative in the EU, and by the representative of the manufacturing plant.
About your Cary Eclipse Instrument

Overview

The Cary Eclipse fluorescence spectrophotometer is ideal for routine laboratory work and offers high performance and advanced design features. The instrument consists of two Czerny-Turner monochromators (excitation and emission), a Xenon light source, a range of fixed width selectable slits, selectable filters, attenuators and two photomultiplier tubes as detectors.

Other design features include:

- A large sample compartment
- Quartz overcoating—to protect the optics
- A small footprint—to save bench space
- Schwarzchild collection optics
- A horizontal beam—less than 0.5mL needed for measurement.

All of these features combine to provide you with an instrument that is fast and efficient.

Xenon flash lamp and Schwarzchild collection optics

![Cary Eclipse Optics](image)

The Schwarzchild collector mirrors collect the light energy from the Xenon Flash Lamp module. This light is then focused through a lens onto the excitation entry slit.

The Xenon flash lamp flashes at a rate of up to 80 flashes per second and has a pulse width of approximately 2 to 3 microseconds.

The Xenon lamp has a long lamp life which also means that you will have less downtime and greater productivity.

Cary Eclipse Optics - full cutaway of the Cary Eclipse.

Multimedia
See also:

- Cary Eclipse Specifications
- Cary Eclipse Optics
Diagram of the Cary Eclipse Optics

Scan fast – in fact the whole wavelength range in less than 3 seconds.

Built-in filters on both sides means that you don’t have to buy your own to exclude second-order stray light.

Excellent sensitivity is a result of using an intense Xenon flash lamp, coupled with Schwarzschild collection optics to send a huge amount of light through the sample.

Photosensitive samples are not exposed to continuous light as the Xenon flash lamp flashes only to acquire a data point.

Less than 0.5 mL is needed for a measurement. The horizontal beam profile ensures excellent sensitivity even with such low volumes.

See also:
Overview
Cary Eclipse Specifications
Specifications

- **Source lamp**
- **Beam arrangement**
- **Excitation monochromator**
- **Emission monochromator**
- **Sample compartment**

### Source lamp

<table>
<thead>
<tr>
<th>Lamp Full spectrum</th>
<th>15W Xenon pulse lamp</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pulse width</td>
<td>2-3us</td>
</tr>
<tr>
<td>Power</td>
<td>60-75kW</td>
</tr>
</tbody>
</table>

### Beam arrangement

<table>
<thead>
<tr>
<th>Beam Splitter</th>
<th>92% - 8%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Detectors</td>
<td>Photomultiplier (X2)</td>
</tr>
<tr>
<td>Reflective surfaces are designed for constant focus at all wavelengths and are quartz overcoated for high performance and corrosion resistance.</td>
<td></td>
</tr>
</tbody>
</table>

### Excitation monochromator

<table>
<thead>
<tr>
<th>Type</th>
<th>Czerny–Turner</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dispersing method</td>
<td>Grating, 30 x 35 mm, 1200 lines/mm</td>
</tr>
<tr>
<td>Blaze angle</td>
<td>370 nm</td>
</tr>
<tr>
<td>Spectral bandwidths</td>
<td>1.5 nm 2.5 nm 5.0 nm 10 nm 20 nm 10 nm round</td>
</tr>
<tr>
<td>Filters</td>
<td>Selectable</td>
</tr>
<tr>
<td></td>
<td>Open</td>
</tr>
<tr>
<td></td>
<td>Closed</td>
</tr>
<tr>
<td></td>
<td>250 - 395 nm</td>
</tr>
<tr>
<td></td>
<td>UG5</td>
</tr>
<tr>
<td></td>
<td>335 - 620 nm</td>
</tr>
<tr>
<td></td>
<td>BG38</td>
</tr>
<tr>
<td></td>
<td>550 - 1100 nm</td>
</tr>
<tr>
<td></td>
<td>OG550</td>
</tr>
<tr>
<td></td>
<td>695 - 1100 nm</td>
</tr>
<tr>
<td></td>
<td>RG695</td>
</tr>
<tr>
<td></td>
<td>Auto</td>
</tr>
<tr>
<td>Reciprocated Linear dispersion</td>
<td>3 mm/mm</td>
</tr>
<tr>
<td>Wavelength range</td>
<td>190 to 900 nm</td>
</tr>
<tr>
<td>Accuracy</td>
<td>±1.0 nm, (±0.5 nm at 541.92 nm)</td>
</tr>
<tr>
<td>Wavelength repeatability</td>
<td>±0.2 nm</td>
</tr>
<tr>
<td>Wavelength drive</td>
<td>Software controlled motor drive.</td>
</tr>
<tr>
<td></td>
<td>Non measurement phase stepping.</td>
</tr>
</tbody>
</table>

Diagram of the Cary Eclipse optics
### Emission monochromator

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Specification</th>
</tr>
</thead>
<tbody>
<tr>
<td>Type</td>
<td>Czerny–Turner</td>
</tr>
<tr>
<td>Dispersing method</td>
<td>Grating, 30 x 35 mm, 1200 lines/mm</td>
</tr>
<tr>
<td>Blaze angle</td>
<td>440 nm</td>
</tr>
<tr>
<td>Spectral bandwidths</td>
<td>Selectable fixed width at</td>
</tr>
<tr>
<td></td>
<td>1.5 nm</td>
</tr>
<tr>
<td></td>
<td>2.5 nm</td>
</tr>
<tr>
<td></td>
<td>5.0 nm</td>
</tr>
<tr>
<td></td>
<td>10 nm</td>
</tr>
<tr>
<td></td>
<td>20 nm and</td>
</tr>
<tr>
<td></td>
<td>10 nm round</td>
</tr>
<tr>
<td>Filters</td>
<td>Selectable</td>
</tr>
<tr>
<td></td>
<td>Open</td>
</tr>
<tr>
<td></td>
<td>Closed</td>
</tr>
<tr>
<td></td>
<td>250 - 395 nm UG5,</td>
</tr>
<tr>
<td></td>
<td>295 - 1100 nm WG295,</td>
</tr>
<tr>
<td></td>
<td>360 - 1100 nm 9086,</td>
</tr>
<tr>
<td></td>
<td>430 - 1100 nm GG435</td>
</tr>
<tr>
<td></td>
<td>550 - 1100 nm OG550</td>
</tr>
<tr>
<td></td>
<td>3% T attenuator</td>
</tr>
<tr>
<td></td>
<td>Mesh (1.5 Abs)</td>
</tr>
<tr>
<td>Reciprocated Linear dispersion</td>
<td>3 mm/mm</td>
</tr>
<tr>
<td>Wavelength range</td>
<td>190 to 900 nm</td>
</tr>
<tr>
<td>Accuracy</td>
<td>±1.0 nm, (±0.5 nm at 541.92 nm)</td>
</tr>
<tr>
<td>Wavelength repeatability</td>
<td>±0.2 nm</td>
</tr>
<tr>
<td>Wavelength drive</td>
<td>Software controlled motor drive,</td>
</tr>
<tr>
<td></td>
<td>Non measurement phase stepping.</td>
</tr>
<tr>
<td>Maximum Slew rate</td>
<td>24,000 nm/min</td>
</tr>
<tr>
<td>Maximum Scan rate</td>
<td>24,000 nm/min</td>
</tr>
</tbody>
</table>

### Sample compartment

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Specification</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dimensions</td>
<td>198 x 273 x 205 mm (WxDxH)</td>
</tr>
<tr>
<td>Access</td>
<td>Front and top</td>
</tr>
</tbody>
</table>

See also:  
- **Overview**  
- **Cary Eclipse Optics**
Software Applications Help

The Cary Eclipse software comprises modular software applications, which means that you can choose the application you need, depending upon what you want to do. For example, if you want to perform quantitative analysis, use the Concentration application. If you want to perform scanning, use the Scan application.

Click on the links below to learn more about the software application you're interested in.

<table>
<thead>
<tr>
<th>Software Application</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>ADL</td>
<td>ADL is pre-defined template for writing ADL (Applications Development Language) programs</td>
</tr>
<tr>
<td>Lifetimes</td>
<td>Use the Lifetimes Application to measure phosphorescence lifetimes</td>
</tr>
<tr>
<td>ADL Shell</td>
<td>ADL Shell provides pre-written commands which are already implemented. Use the ADL Shell to provide the basic functionality and build on that.</td>
</tr>
<tr>
<td>Ratios</td>
<td>Use the Ratio Application to obtain a concentration value for particular reactions.</td>
</tr>
<tr>
<td>Advanced Reads</td>
<td>Use the Advanced Reads Application to take Absorbance readings of multiple samples</td>
</tr>
<tr>
<td>Scan</td>
<td>Use the Scan Application to perform wavelength scanning</td>
</tr>
<tr>
<td>Align</td>
<td>Use the Align application to align the Cary Eclipse lamps and accessories</td>
</tr>
<tr>
<td>Simple Reads</td>
<td>Use the Simple Reads Application when you want to take absorbance readings at a single wavelength</td>
</tr>
<tr>
<td>Concentration</td>
<td>Use the Concentration Application when you want to perform quantitative measurements</td>
</tr>
<tr>
<td>System Information</td>
<td>Use the System Information Application for recording your company and instrument details for use in reports</td>
</tr>
<tr>
<td>GLP Administration</td>
<td>Use the GLP Administration Application for setting up file and method security and access control</td>
</tr>
<tr>
<td>Thermal</td>
<td>Use the Thermal Application to perform thermal analyses on DNA</td>
</tr>
<tr>
<td>Kinetics</td>
<td>Use the Kinetics Application to perform time-based single or multicell measurements</td>
</tr>
<tr>
<td>Validate</td>
<td>Use the Validate Application to test your instrument and ensure that it is working correctly</td>
</tr>
</tbody>
</table>

See also:
- Application How To List
- Glossary
- Cary Application Descriptions
## Cary Application Descriptions

<table>
<thead>
<tr>
<th>Application</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>ADL Shell</td>
<td>A pre-defined template for writing ADL programs.</td>
</tr>
<tr>
<td>Advanced Reads</td>
<td>For taking intensity readings of multiple samples.</td>
</tr>
<tr>
<td>Align</td>
<td>A utility application for aligning lamps and accessories.</td>
</tr>
<tr>
<td>Concentration</td>
<td>For quantitative measurements.</td>
</tr>
<tr>
<td>GLP Administration</td>
<td>For setting up file and method security and access control.</td>
</tr>
<tr>
<td>Kinetics</td>
<td>For time-based single or multicell measurements.</td>
</tr>
<tr>
<td>Ratios</td>
<td></td>
</tr>
<tr>
<td>Scan</td>
<td>For wavelength/wavenumber scanning.</td>
</tr>
<tr>
<td>Simple Reads</td>
<td>For taking intensity readings at a single wavelength.</td>
</tr>
<tr>
<td>System Information</td>
<td>For recording your company and instrument details for use in reports etc.</td>
</tr>
<tr>
<td>Thermal</td>
<td>For performing thermal analyses on DNA.</td>
</tr>
<tr>
<td>Validate</td>
<td>For testing your instrument to ensure that it is working correctly.</td>
</tr>
</tbody>
</table>
# Applications How To… List

To learn how to perform many of the everyday procedures you'll use your Cary Eclipse for, just follow these simple step-by-step examples.

Click on the links below to view How To's on the software application you're interested in.

<table>
<thead>
<tr>
<th>ADL</th>
<th>Ratios</th>
</tr>
</thead>
<tbody>
<tr>
<td>ADL is pre-defined template for writing ADL (Applications Development Language) programs</td>
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</tbody>
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<table>
<thead>
<tr>
<th>Advanced Reads</th>
<th>Scan</th>
</tr>
</thead>
<tbody>
<tr>
<td>Use the Advanced Reads Application to take Absorbance readings of multiple samples</td>
<td>Use the Scan Application to perform wavelength scanning</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Align</th>
<th>Simple Reads</th>
</tr>
</thead>
<tbody>
<tr>
<td>Use the Align application to align the Cary Eclipse lamps and accessories</td>
<td>Use the Simple Reads Application when you want to take absorbance readings at a single wavelength</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Concentration</th>
<th>System Information</th>
</tr>
</thead>
<tbody>
<tr>
<td>Use the Concentration Application when you want to perform quantitative measurements</td>
<td>Use the System Information Application for recording your company and instrument details for use in reports</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>GLP Administration</th>
<th>Thermal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Use the GLP Administration Application for setting up file and method security and access control</td>
<td>Use the Thermal Application to perform thermal analyses on DNA</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Kinetics</th>
<th>Validate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Use the Kinetics Application to perform time-based single or multicell measurements</td>
<td>Use the Validate Application to test your instrument and ensure that it is working correctly</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Lifetimes</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Use the Lifetimes Application to measure phosphorescence lifetimes</td>
<td></td>
</tr>
</tbody>
</table>

See also:
[Application Help List](#)
What is ADL?

The Applications Development Language (ADL) is a BASIC-like spectroscopy language which is built-into the Cary software. It uses simple, English words to perform common spectroscopic functions e.g. "collect" starts a data collection, "curvefit" fits a curve. The Cary version of ADL uses a commercial programming language—SAXBasic® to provide basic functionality with Cary-specific commands added.

Find out more about ADL and how to write your own controls for your instrument using this simple language.
<table>
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<tr>
<th>To find out about…</th>
<th>See the…</th>
<th>Show me…</th>
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<tr>
<td>ADL Shell</td>
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<tr>
<td>File management</td>
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<td>Save As</td>
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<tr>
<td></td>
<td>Filename extensions</td>
<td></td>
</tr>
<tr>
<td>Screen features</td>
<td>Graphics Area</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Stop Instrument button</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Commands menu</td>
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<td></td>
<td>Status Line</td>
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<td>Function keys</td>
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<tr>
<td>Viewing and editing</td>
<td>View menu</td>
<td>How to use advanced reporting options</td>
</tr>
<tr>
<td>features</td>
<td>Edit menu</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Print Preview</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Print</td>
<td></td>
</tr>
<tr>
<td></td>
<td>User Data Form</td>
<td></td>
</tr>
<tr>
<td>ADL</td>
<td>ADL Command Line</td>
<td>How to insert ADL commands using the Quick</td>
</tr>
<tr>
<td></td>
<td>ADL Hooks</td>
<td>Reference</td>
</tr>
<tr>
<td>Help and safety</td>
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<td></td>
</tr>
<tr>
<td>information</td>
<td>Safety Home Page</td>
<td></td>
</tr>
</tbody>
</table>
ADL Shell Application

Introduction

The ADL Shell gives you a pre-defined template for writing ADL programs. Rather than needing to write the code for basic functions such as graphing and filing, the ADL Shell has a number of these commands already implemented. In other words, you do not have to design your own interface—you can use the ADL Shell to provide the basic functionality and build on that.

For example, your company may perform a particular analysis and series of calculations repeatedly. You may like to use the ADL Shell to write a series of prompt dialogs that will take the user through each step in sequence.

Some frequently asked questions on the ADL Shell applications are listed below:

What is provided by the ADL Shell?

The ADL Shell pre-defined modules include:

- Stop button
- Connect button
- Abscissa and Ordinate display
- File handling functions
- Printer and print preview functions
- Graphics interface including the functions provided by the toolbar or graph menu
- Reports area and report editing and font functions
- Rapid Result
- Go To Wavelength
- Zero
- Function keys
- Align functions (i.e. fluorescence spectrophotometer is driven to white light)

What sort of items would I need to program?

Should you decide to use the ADL Shell to write your own applications, some of the items that you might like to program could include:

- Command buttons on the main window
- A Setup dialog as seen in other Cary Eclipse applications that includes instrument and accessory control and reporting and filing functions
- Instrument control via a Start button

How do I write an ADL program?

The easiest method is to use the ADL Editor. You can display the ADL Editor from the ADL menu item in the View menu. The Editor provides ready access to ADL commands and the syntax required for these commands as well extensive debugging tools. A further discussion on ADL commands is provided in the ADL Help. Click the button below to view this Help.

Software controls
The application comprises the following:

**Instrument Status Readings**

The Instrument Status Readings, which display the Y and X values of the graph axes are displayed in the top left and right corners of the ADL Shell Application window.

**Y value**
Displays the current Y value, usually intensity. This value is continually updated during a data collection.

**X value**
Displays the current X values, in nanometers, for both excitation and emission.

**Stop button**
The Stop button at the top of the ADL Shell Application window is used to stop instrument analyses. The button can also be accessed from the Commands menu.

**Graph area**
The Graphics area refers to the plotting area of the application. It can contain traces of collected data, labels and pictures. In the Cary Eclipse system, the term ‘graph’ refers to the plotting area and is represented by a set of axes. The rectangular area defined by the axes is referred to as a graph box.

To display or hide the Graphics area, select the Graphics command on the View menu. Double-clicking on the Graphics area enables you to toggle between half or full size display sizes. To manually size the Graphics area, place the mouse cursor over the border between the Graph and Report areas until the cursor changes to a double arrow (↵). Then hold down the mouse and drag it up or down to resize the areas.

**Tell me more about the Graph area**

**Report area**
Use the ADL Shell Report area to view the current report. The report may include the results of your data collection, the method parameters used to collect the results and any graphics you have included. You can select the information to include in your report on the Reports page of the Setup dialog.

You can copy the generated report onto the Windows clipboard. To do this, highlight the report (or the relevant section) and press CTRL+C. You can then paste the report into a word processor such as Microsoft® Word.

**Hot Tip**
You can edit the ADL Shell Report by pressing CTRL+R or selecting the Edit Report command from the Edit menu.

**Menus**
The ADL Shell Application contains the following menus:

File, Edit, View, Commands, Graph, Help

See also:

[Keyboard function keys](#)
[ADL Shell home page](#)
Menu line—ADL Shell

- File menu
- Edit menu
- View menu
- Commands menu
- Graph menu
- Help menu
File menu—ADL Shell

Use the File menu to open or save a method or data file, print or preview a report, or exit the Cary Eclipse application.

**Hot Tip**
Highlight a method or data file in Explorer and simply drag and drop it onto the application to open it. Any data will be displayed in the Graphics area.

The following commands can be accessed from the File menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>New</td>
<td>Select New to clear all method, report and graphics information and create a new file containing the default settings.</td>
</tr>
<tr>
<td>Open Method…</td>
<td>Select Open Method to display the Open dialog where you can search for and load a stored method.</td>
</tr>
<tr>
<td>Open…</td>
<td>Select Open to display the Open dialog where you can search for and load method, data, report, graph template and batch files, as well as other file types compatible with the Cary Eclipse software. When you load a batch file, the method which collected the data is loaded as well as the associated report and graphics layout.</td>
</tr>
<tr>
<td>Save Method As…</td>
<td>Select this option to display the Save As dialog and store the current method. You can use the Save As dialog to store the method, data, report, or graphics template separately or together as a Batch file. The data can also be saved as a comma delimited ASCII Spreadsheet file.</td>
</tr>
<tr>
<td>Save As…</td>
<td>Select this option to display the Save As dialog and store the collected data. You can use the Save As dialog to store the method, data, report, or graphics template separately or together as a Batch file. The data can also be saved as a comma delimited ASCII file.</td>
</tr>
<tr>
<td>Print Setup…</td>
<td>Select this option to display the Windows Print Setup dialog where you can select the printer, printer page size and orientation.</td>
</tr>
<tr>
<td>Print Preview…</td>
<td>Select this option to display the Print Preview dialog where you can view the current report as it will be printed.</td>
</tr>
<tr>
<td>Print…</td>
<td>Select this option to display the Windows Print dialog where you can choose the printer and pages to print as well as the number of copies you wish to print.</td>
</tr>
<tr>
<td>Explorer…</td>
<td>Select this option to run Windows Explorer. This may help you locate method and data files.</td>
</tr>
<tr>
<td>History List</td>
<td>This displays a list of the four most recently opened files.</td>
</tr>
<tr>
<td>Exit</td>
<td>Select this option to exit the ADL Shell application.</td>
</tr>
</tbody>
</table>

See also: [Open](#), [Save As](#), [Print Preview](#), [Filename Extensions](#)
Open—ADL Shell

Use the dialog to open a file previously saved by the application. To access the Open dialog, select Open or Open Method from the File menu.

**Look in**  
The Look In field lists the available folders and files. Click the down arrow on the right of this field to view the location of the current folder in your computer’s hierarchy. You can also select a new location simply by selecting a different folder. The folders and files in the selected folder are displayed in the list box below the Look In field.

*Note*
To open the folder in the level that is one higher than the current folder, select on the toolbar.

**Look in list box**  
The Look In List Box, which is directly below the Look In field, displays the folders and files in the selected location. To view the contents of a folder, double-click it.

- Click to create a new folder. Type the name of the new folder in the highlighted text entry field that appears and press Enter. Then double-click on the new folder to open it.

- Click to display the directory structure of the drive.

- Click to display the files as a list.

- Click to display files as a detailed list including the file size, file type and the date the file was last modified.

- Click to preview the selected trace before you load it into the application.

**File name**  
Enter the name of the file you want to open in this field. The easiest way to do this is to find the file in the Look in list box and then select the file. However, you can type the full path of a file and then select Open to open the file.

**Files of type**  
Click on the arrow on the right of this field to view a list of the different types of application files that can be opened in the list box.

- **Methods (*.FMKN)** Choose this file type to open a Method file. Only the method is loaded. No data is loaded.

- **Data (*.FDKN)** Choose this file type to open a Data file. Only the data is loaded. The method that was used to collect the data is not loaded. The last used method or default method will be displayed.

- **Report (*.FRKN)** Choose this file type to open a Report file. You can view the report in the Report area.

- **Graph Template (*.FGKN)** Choose this file to open a Graph Template file. The file will set up the layout of the Graphics area including any bitmaps as well as the range and style of the graph axes.

- **Batch (*.FBKN)** Choose this file type to open a Batch file. A Batch file includes data, the method used to collect the data, report and graphics setup.

- **Spreadsheet Ascii (*.CSV)** This will open ASCII files that contain single comma delimited files with two or more columns.

- **GRAMS (*.SPC)** Choose this file type to open a Cary Eclipse GRAMS data file. This allows you to use data files collected using a Cary Eclipse GRAMS system. The method that was used to collect the data is not loaded.

- **All files (*.*)** This enables you to view all files in the directory so that you can
open any compatible file.

Tell me more about Filename Extensions

**Overlay Data**  Select this check box to overlay individual data files in one graph box. If this is not checked the data from each file will appear in individual graph boxes. This option is only available if the Open dialog has been accessed by selecting 'Open…’ from the File menu.

**Hot Tip**  Use the SHIFT or Control key to select multiple data files.

**Open**  This button opens the highlighted file/s or the files listed in the File name field.

See also:  
[File menu](#)
Save As—ADL Shell

Use the **Save As** dialog to save the data, method, report or graphics template individually or together as a Batch file.

**Note**
This dialog appears when you select either the **Save Method As** or **Save As** menu option in the **File** menu. When you select **Save Method As** this dialog appears with 'Methods' displayed in the **Files of type** field. When you select **Save As** this dialog appears with 'Batch' displayed in the **Files of type** field.

Traces (or data) that are currently ‘Visible’ in the Graphics area are saved, regardless of how they are displayed (full screen graph box or tile).

For example, if six graph boxes are displayed in the Graphics area and each graph box has one Visible trace then all six traces will be saved.

If you do not wish to save particular traces, make them not Visible in the **Trace Preferences** dialog. To save only the focused trace, select the **Save only focused trace** check box.

The **Save As** dialog comprises the following:

**Look In**
The **Look In** field lists the available folders and files. Click the down arrow on the right of this field to view the location of the current folder in your computer's hierarchy. You can also select a new location simply by selecting a different folder. The folders and files in the selected folder are displayed in the list box below the **Look In** field.

**Note**
To open the folder in the level that is one higher than the current folder, select on the toolbar.

**Look in list box**
The **Look In** list box, which is directly below the **Look In** field, displays the folders and files in the selected location. To view the contents of a folder, double-click it.

- Click to create a new folder. Type the name of the new folder in the highlighted text entry field that appears and press ENTER. Then double-click on the new folder to open it.
- Click to display the directory structure of the drive.
- Click to display the files as a list.
- Click to display files as a detailed list including the file size, file type and the date the file was last modified.
- Click to preview the selected trace before you load it into the application.

**File Name**
Enter the name of the file you want to save in this field. You can also type the full path of a file then select the **Save** button to save the file.

**Files of Type**
Click on the arrow on the right of this field to view a list of the different types of application files that can be opened in the list box.

- **Methods (.*.FMKN)** Choose this file type to only save the current method. Any collected data is not stored.
- **Data (.*.FDKN)** Choose this file type to only save the data currently Visible in the **Trace Preferences** dialog (for any graph box). Note that the method
used to collect the data is not stored with the data.

**Report (*.FRKN)** Choose this file type to only save the contents currently shown in the Report area.

**Graph Template (*.FGKN)** Choose this file type to save the layout of the Graphics area as a template. For example, any bitmaps you have added to your Graphics area will be saved as well as the range and style of the graph axes.

**Batch (*.FBKN)** Choose this file type to save the data that is currently Visible in the Trace Preferences dialog (for any graph box), the method used to collect the data and the currently displayed report all together as a batch file. The graphics setup is also stored.

**Spreadsheet Ascii (*.CSV)** Choose this file type to save the raw intensity data currently Visible in the Trace Preferences dialog (for any graph box) as a comma delimited ASCII Spreadsheet file (*.CSV) file. The data is saved as XY pairs delimited by a comma.

**Note**
This file type can be easily imported into a spreadsheet program. For example, Microsoft Excel. You can also double-click on the stored *.CSV file from Microsoft Explorer to load Excel automatically with the ASCII XY data displayed.

**GRAMS (*.SPC)** Choose this file type to only save the data currently Visible in the Trace Preferences dialog (for any graph box) as a Cary Eclipse GRAMS data file.

See also:

**Save only focused trace** Select this option to save only the focused trace in the graph box. If this option is not selected, all visible traces in the graph box will be saved.

**Save** Click this button to save the file currently present in the File name field in the directory specified in the Look in field.

See also:

**File menu**
Filename extensions—ADL Shell

The Cary Eclipse system uses the filename extension to distinguish between data types (e.g. method or data) and also to differentiate applications. (e.g. Concentration or Kinetics).

The Syntax for file extensions is:

<table>
<thead>
<tr>
<th>Letter</th>
<th>Represents</th>
<th>Available Options</th>
</tr>
</thead>
<tbody>
<tr>
<td>First</td>
<td>Type of analysis</td>
<td>F Fluorescence</td>
</tr>
<tr>
<td>Second</td>
<td>Type of file</td>
<td>B Batch, D Data, G Graph Template, M Method, R Report</td>
</tr>
<tr>
<td>Third and fourth</td>
<td>Application identifier</td>
<td>AB Advanced Reads, CN Concentration, KN Kinetics, LT Lifetimes, SR Simple Reads, SW Scan, VO Validate</td>
</tr>
</tbody>
</table>

The following files may be opened and saved in the ADL Shell Application, Note that the ?? can be replaced with any of the two letter Application identifier codes listed above if you wish to narrow down the list:

<table>
<thead>
<tr>
<th>Extension</th>
<th>Available action</th>
<th>File type</th>
</tr>
</thead>
<tbody>
<tr>
<td>*.FM??</td>
<td>Open/Save</td>
<td>A method file (e.g. *.FMKN) consists of the software setup by which kinetics data is collected.</td>
</tr>
<tr>
<td>*.FD??</td>
<td>Open/Save</td>
<td>A data file (*.FDSR) contains data collected that is currently visible in the Trace Preferences dialog (for any graph box).</td>
</tr>
<tr>
<td>*.FR??</td>
<td>Open/Save</td>
<td>A report file (*.FRKN) contains the report generated by the Cary Eclipse about the data.</td>
</tr>
<tr>
<td>*.FG??</td>
<td>Open/Save</td>
<td>A graph template file (*.FGKN) contains information that sets up the layout of the Graphics area including any bitmaps as well as the range and style of the graph axes.</td>
</tr>
<tr>
<td>*.FB??</td>
<td>Open/Save</td>
<td>A batch file (*.FBKN) contains the collected data that was Visible in the Trace Preferences dialog (for any graph box), the method by which the data was collected and the report and graphics template.</td>
</tr>
<tr>
<td>*.CSV</td>
<td>Open/Save</td>
<td>An ASCII spreadsheet file (*.FCSV) has the same extension for all applications. It consists of raw intensity data (that was currently Visible in the Trace Preferences dialog (for any graph box) saved as XY comma-delimited pairs. This file can be imported into a spreadsheet program such as Microsoft Excel.</td>
</tr>
</tbody>
</table>

**Note**

Selecting All Files in the Windows Open dialog will list all available file types.

See also:

Open
Save As
Edit menu—ADL Shell

Use the Edit menu to copy, cut, paste and delete information. You can also use the menu to access the Report editing commands. The menu is accessible from the menu line at the top of the ADL Shell Application window.

The following commands can be accessed from the Edit menu:

<table>
<thead>
<tr>
<th>Menu command</th>
<th>Button</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Undo</td>
<td><img src="image" alt="Undo" /></td>
<td>If available, this undoes the last action. This command is also accessible by pressing CTRL+Z.</td>
</tr>
<tr>
<td>Cut</td>
<td><img src="image" alt="Cut" /></td>
<td>Select this command to remove the highlighted text and place it on the Windows Clipboard. This command is also accessible by pressing CTRL+X.</td>
</tr>
<tr>
<td>Copy</td>
<td><img src="image" alt="Copy" /></td>
<td>Select this command to copy the highlighted text onto the Windows Clipboard. This command is also accessible by pressing CTRL+C.</td>
</tr>
<tr>
<td>Paste</td>
<td><img src="image" alt="Paste" /></td>
<td>Select this command to insert the contents of the Windows Clipboard (text only) at the cursor. Any highlighted text will be replaced. This command is also accessible by pressing CTRL+V.</td>
</tr>
<tr>
<td>Delete</td>
<td><img src="image" alt="Delete" /></td>
<td>Select this command to remove selected text in the Report area. This option is only available once you select Edit Report and there is text highlighted in the Report area. This command is also accessible by pressing DEL.</td>
</tr>
<tr>
<td>Select All</td>
<td><img src="image" alt="Select All" /></td>
<td>Select this option to highlight all text in the Report area. This command is also accessible by pressing CTRL+A.</td>
</tr>
<tr>
<td>Font</td>
<td><img src="image" alt="Font" /></td>
<td>This enables you to change the font of new or selected text in the report. For existing text, simply highlight the text in the report, select Font and make your changes in the Font dialog. When you select OK, the appearance of the highlighted text in the report will alter to reflect your font changes. You do not have to select Edit Report to use this command. This command is also accessible by pressing CTRL+F.</td>
</tr>
<tr>
<td>Edit Report</td>
<td><img src="image" alt="Edit Report" /></td>
<td>Select this option to edit the report. This command is also accessible by pressing CTRL+R.</td>
</tr>
<tr>
<td>Clear Report</td>
<td><img src="image" alt="Clear Report" /></td>
<td>Select this command to remove the contents of the Report area. If you have saved the report, selecting this option does not delete the contents of the report, the information is simply removed from the Report area.</td>
</tr>
<tr>
<td>Create Report</td>
<td><img src="image" alt="Create Report" /></td>
<td>Select this command to create a new report.</td>
</tr>
</tbody>
</table>

Hot Tip
You can copy a picture, spreadsheet file or document from another application (for example, Microsoft Word or Microsoft Excel) into the report area of the of the ADL Shell application. To edit these objects, simply double click on them to open the associated application in which they were created.

See also:
Menu line
View menu—ADL Shell

The View menu is accessible from the menu line at the top of the ADL Shell Application window.

**Hot Tip**
Highlight a method or data file in Explorer and simply drag and drop it onto the application to open it. Any data will be displayed in the Graphics area.

The following commands can be accessed from the View menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Toolbar</strong></td>
<td>Select this option to toggle the Graphics and Reports toolbar on and off. You can also select to display or hide the toolbar Text Labels.</td>
</tr>
<tr>
<td><strong>Buttons</strong></td>
<td>Select this option to toggle the left-hand button display (button area) on and off.</td>
</tr>
<tr>
<td><strong>Graphics</strong></td>
<td>Select this option to toggle the Graphics area on and off.</td>
</tr>
<tr>
<td><strong>Report</strong></td>
<td>Select this option to toggle the Report area on and off.</td>
</tr>
<tr>
<td><strong>Report Vertical</strong></td>
<td>If the Graphics option is selected, select Report Vertical to display the Graphics area, Report area and/or ADL Editor side by side vertically.</td>
</tr>
<tr>
<td><strong>ADL</strong></td>
<td>Select this option to toggle the ADL Editor on and off.</td>
</tr>
<tr>
<td><strong>ADL Vertical</strong></td>
<td>If the ADL and Report options are selected, select ADL Vertical to display the ADL Editor vertically on the right hand side of the Report area.</td>
</tr>
<tr>
<td><strong>ADL Quick Reference</strong></td>
<td>This displays the Quick Reference window where you can easily add a command in the correct syntax to your ADL program. A detailed description of all the commands in the Quick Reference is available by selecting the Help button.</td>
</tr>
<tr>
<td><strong>ADL Hooks</strong></td>
<td>Select this to activate a link (or hook) to an ADL program at a specific point in the application, and then run that ADL program. For example, ADL hooks have been supplied to run specific programs:</td>
</tr>
<tr>
<td></td>
<td>- on start-up of the Cary Eclipse software;</td>
</tr>
<tr>
<td></td>
<td>- when the system goes online or offline;</td>
</tr>
<tr>
<td></td>
<td>- when the Start button is selected;</td>
</tr>
<tr>
<td></td>
<td>- when a run finishes;</td>
</tr>
<tr>
<td></td>
<td>- when a zero is performed etc.</td>
</tr>
<tr>
<td></td>
<td>For the hook to work, ADL Hooks must be selected, and the appropriate ADL file for the application must be present in the ADL folder in the directory.</td>
</tr>
<tr>
<td></td>
<td><strong>Note</strong></td>
</tr>
<tr>
<td></td>
<td>Full documentation on ADL programming is available in the ADL Online Help.</td>
</tr>
<tr>
<td></td>
<td>Click the button below to view this Help.</td>
</tr>
<tr>
<td><strong>Hints</strong></td>
<td>Select this option to toggle the Hints bubble text on and off. The bubble text appears when you place the arrow cursor over a field, button or text entry field for a short period. It offers a quick explanation of the item’s function.</td>
</tr>
</tbody>
</table>

See also:
- ADL Shell Application
- Graphics area
- ADL Hooks
- How to insert ADL commands using the Quick Reference
Commands menu—ADL Shell

Use the Commands menu to access major operational commands in the ADL Shell application. The menu is accessible from the menu line at the top of the ADL Shell Application window.

Alternatively you can use function key shortcuts for some of the commands. These appear next to the item in the Commands menu.

The Commands menu comprises the following commands:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Start</td>
<td>Select this to start a data collection using the currently set up parameters.</td>
</tr>
<tr>
<td></td>
<td>If the Connect command is displayed in place of the Start command, this indicates that another Cary Eclipse application is in use and has control of the Cary Eclipse instrument. To make the ADL Shell application active so that you can perform a read, you will need to select Connect. However, you will not be able to select Connect if another application is collecting data.</td>
</tr>
<tr>
<td>Stop</td>
<td>Select this to stop the data collection.</td>
</tr>
<tr>
<td></td>
<td>If you have selected Storage On in the Auto Store page of the Setup dialog, all data collected is automatically stored in the file specified in the Batch name field of the Windows Save As dialog before or after the run commenced.</td>
</tr>
<tr>
<td>Clear Zero</td>
<td>Select this option to clear the Zero command. When you select the Zero command (from the Commands menu or by pressing F5), the intensity at that time will be reset to zero and all subsequent results will have the intensity value that was reset to zero subtracted from them. Selecting Clear zero cancels this zeroing (and thus subtraction) effect.</td>
</tr>
<tr>
<td>Pre-scan</td>
<td>Select Pre-scan to quickly determine the optimum instrument parameters to be used for your sample. At the end of this collection sequence the software will report the optimum Ex and Em wavelengths and PMT voltage. Once you are satisfied with the parameters, the software will automatically update your method setup to reflect these new parameters.</td>
</tr>
<tr>
<td>Zero order…</td>
<td>Select the Zero order command to set one of the monochromators to zero order. If you use this command to zero either the excitation or emission monochromators, this will override the Ex. Wavelength or Em. Wavelength set on the Cary page. The Cary Eclipse software will use the zero setting when collecting data. The Zero order command is generally used during alignment procedures.</td>
</tr>
<tr>
<td></td>
<td><strong>Excitation</strong> Select this command to set the excitation monochromator to zero order (white light).</td>
</tr>
<tr>
<td></td>
<td><strong>Emission</strong> Select this command to set the emission monochromator to zero order (white light).</td>
</tr>
<tr>
<td>Reset</td>
<td>Select this command to re-initialize the instrument. This will perform the same initializing procedure that is performed when the instrument is powered on.</td>
</tr>
<tr>
<td>Rapid Result</td>
<td>Select this to display the Rapid Result dialog. Use this dialog to take a reading of a blank and multiple sample readings without changing the application method. This means that between runs, while new samples are being prepared and loaded, other samples can quickly be measured. This command can also be accessed by pressing F6.</td>
</tr>
<tr>
<td>Zero</td>
<td>Select this to zero the current intensity value. The intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before selecting Start if you want your data to start at 0 intensity. This command is also accessible by pressing F5.</td>
</tr>
</tbody>
</table>

**Note**
Before you select **Zero**, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.

| **Go to...** | Select **Go To...** to display the **Go To Wavelength** dialog where you can change the instrument to a new wavelength. Use this command if you want to quickly go to a wavelength to check the intensity reading.
This command is only available if the instrument is not currently collecting data. It can also be accessed by pressing F4.

**Note**
This is not the wavelength used in the application method. If your method uses a wavelength that is different from the 'Go To wavelength', then when you select **Start** the system will change to the wavelength selected. (In most applications, this is set in the Cary page of the **Setup** dialog.) |

| **ADL Command** | Select this to display the **ADL Command Line** window where you can execute an ADL command or ADL program. (You can also press Alt-Y to display this dialog.) |

| **Rerun ADL** | Select this to run the ADL command or program in the ADL Command Line window again. (You can also press Alt-X to rerun programs.) |

**See also:**
- [ADL Shell Application](#)
- [Zero](#)
- [Rapid Result](#)
- [Go To Wavelength](#)
- [ADL Command Line](#)
Go To Wavelength—ADL Shell

Use the Go to Wavelength dialog to temporarily change the instrument to a new excitation or emission wavelength. You would mainly use this dialog if you want to quickly go to a wavelength and check the intensity reading of the sample. The lamp will flash continuously in this mode. This option is only available if the instrument is not currently collecting data.

To access the Go to Wavelength dialog, select the Go To Wavelength command from the Commands menu. You can also select F4 to open the dialog.

Note
This is not the wavelength used in the application method. If your method uses a wavelength that is different from the 'Go to wavelength' setting you select, then when you select Start the system will change to the wavelength selected. (In most applications, this is set in the Cary page of the Setup dialog.)

The Go to Wavelength page comprises the following:

**Excitation**

- **Wavelength**
  Enter the new wavelength in this numeric entry field. When you select Go to the instrument will change to the new value.

- **Zero Order**
  Select Zero Order to drive the Cary Eclipse fluorescence spectrophotometer to 0 nm (white light). You can then use this light for aligning accessories.

**Emission**

- **Wavelength**
  Enter the new wavelength in this numeric entry field. When you select Go to the instrument will change to the new value.

- **Zero Order**
  Select Zero Order to drive the Cary Eclipse fluorescence spectrophotometer to 0 nm (white light). You can then use this light for aligning accessories.

**Go To**

Click Go to once you have entered a new wavelength, to change the instrument to the entered value.

See also:
Function Keys
Zero—ADL Shell

Use the Zero button to zero the current intensity value. The intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before commencing a data collection if you want your data to start at 0 intensity. The Cary Eclipse will prompt you to load a blank before performing the zero.

Before you select Zero, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.

When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the ADL Shell Application window.

To access the Zero option, select the Zero command from the Commands menu. Select the Clear Zero command from the Commands menu to remove zero correction and thus remove the subtraction effect.

See also:
ADL Shell Application Commands menu
Rapid Result—ADL Shell

Use the **Rapid Result** dialog to take a reading of a blank and sample/s, without having to change the current application method. This means that between runs, while new samples are being prepared and loaded, other samples can quickly be measured. The result is printed in the Report area.

To access the **Rapid Result** dialog, select the **Rapid Result** command from the **Commands** menu or press F6.

**Excitation**

- **Wavelength**
  Enter the wavelength at which you want to take a reading.

- **Zero order**
  Select this option to set the excitation monochromator to zero order (white light).

**Emission**

- **Wavelength**
  Enter the wavelength at which you want to take a reading.

- **Zero order**
  Select this option to set the excitation monochromator to zero order (white light).

**Zero**

Click **Zero** to change the wavelength to the entered value and measure a blank.

**Result**

Click **Result** to change the wavelength to the entered value and measure a sample. The X,Y results are corrected for the blank and the results are displayed in the Report window.

See also:

- **Commands menu**
Graph menu—ADL Shell

Introduction

Use the Graph menu to view data in various graphical formats or in multiple graphs in the Graphics area. You can double-click on the Graphics area to toggle the display through graph and report to graph only.

In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area surrounding the graph is referred to as the graph box. A graph will appear in the Graphics area when you select Start to commence a ADL Shell run.

The Graph menu can be accessed in the following ways:

- from the main menu at the top of the ADL Shell Application window
- by clicking the right mouse button in a graph box or the Graphics area

Note
The red trace in any graph box is the one on which all information appears and the one used for the cursor tracking. It is called the focused trace.

Menu commands

The following commands can be accessed from the Graph menu:

<table>
<thead>
<tr>
<th>Menu command</th>
<th>Button</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Print XY point</td>
<td></td>
<td>Select this to print the cursor's current X and Y coordinates in the report area.</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Note:</strong> This option is only available from the Graph menu that appears when you right-click in a graph box.</td>
</tr>
<tr>
<td>Copy Graph</td>
<td></td>
<td>Select <strong>Copy Graph</strong> to copy the entire Graphics area to the Windows Clipboard. All of the graphs in the Graphics area will be copied to the Clipboard.</td>
</tr>
<tr>
<td>Paste to Graph</td>
<td></td>
<td>Select <strong>Paste to Graph</strong> to paste the contents of the Windows Clipboard into the Graphics area. Text is pasted as a label, graphics are pasted as pictures.</td>
</tr>
<tr>
<td>Add Graph</td>
<td>![Add Graph]</td>
<td>Select <strong>Add Graph</strong> to add a new graph box to the Graphics area. You may want to do this if you have, for example, a graph containing existing data and you want to collect some new data in a new graph.</td>
</tr>
<tr>
<td>Remove Graph</td>
<td>![Remove Graph]</td>
<td>Select <strong>Remove Graph</strong> to delete the selected graph box from the Graphics area. The graph box is removed, but the data is not deleted and is still available from the <strong>Trace Preferences</strong> window. You may want to use remove graph if you have a large number of graph boxes open but only want to focus on some of these.</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Note:</strong> You can remove multiple graph boxes by first holding down the CTRL key and selecting the graphs to be removed and then selecting the <strong>Remove Graph</strong> button or menu item. Use the SHIFT key to highlight a range of graphs for removal.</td>
</tr>
<tr>
<td>Clear All Traces</td>
<td>![Clear All Traces]</td>
<td>Select <strong>Clear All Traces</strong> to delete all traces from the graph/s. Note that selecting this option simply turns off the <strong>Visible</strong> property for the traces in the <strong>Trace Preferences</strong> window. You can redisplay the traces by making the traces <strong>Visible</strong> again in the <strong>Trace Preferences</strong> window.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Select <strong>Add Label</strong> to display the <strong>Add Label</strong> dialog where you can enter a text label to...</td>
</tr>
<tr>
<td><strong>Add Label</strong></td>
<td>be placed on a graph or in the background of the Graphics area.</td>
<td></td>
</tr>
<tr>
<td>------------------------</td>
<td>------------------------------------------------------------------------</td>
<td></td>
</tr>
<tr>
<td><strong>Add Picture</strong></td>
<td>Select Add Picture to display the Add Picture dialog where you can select a picture, bitmap or chemical structure to be placed on a graph or in the background of the Graphics area.</td>
<td></td>
</tr>
<tr>
<td><strong>Edit Annotation</strong></td>
<td>Select Edit Annotation to alter the content or look of the selected annotation, (either a label or a picture) in the Graphics area.</td>
<td></td>
</tr>
<tr>
<td></td>
<td><strong>Note:</strong> This option can only be activated if an annotation is selected.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>If the selected annotation is a label, selecting Edit Annotation will open the Edit Label dialog where you can change the text of the selected label.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>If the selected annotation is a picture, selecting Edit Annotation will open the Edit Picture dialog where you can replace the selected picture with a new picture.</td>
<td></td>
</tr>
<tr>
<td><strong>Delete Annotation</strong></td>
<td>Select Delete Annotation to remove an annotation, (such as a label or a picture) from the Graphics area.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>You can use this menu option to remove highlighted annotations in the Graphics area such as labels, pictures and ruler/fit information.</td>
<td></td>
</tr>
<tr>
<td></td>
<td><strong>Note:</strong> This option can only be activated if an annotation is selected.</td>
<td></td>
</tr>
<tr>
<td><strong>Trace Preferences</strong></td>
<td>Select this command to display the Trace Preferences dialog where you can select the trace(s) to display in the selected graph.</td>
<td></td>
</tr>
<tr>
<td><strong>Graph Preferences</strong></td>
<td>Select this command to display the Graph Preferences dialog where you can change the appearance of your graph. Any changes you make to the appearance of the graph will be applied to all graphs.</td>
<td></td>
</tr>
<tr>
<td><strong>Cursor Mode…</strong></td>
<td>Select this command to display the Cursor Modes dialog where you can choose the type of cursor to be used on the screen. For example, you can select to use a free cursor that can move anywhere in the graph or exactly trace the focused scan (red scan). Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the Least Squares method or the Point to Point method.</td>
<td></td>
</tr>
<tr>
<td><strong>Axes Scales…</strong></td>
<td>Select this command to display the Axes Scales dialog where you can set the scale range for the X and Y axes.</td>
<td></td>
</tr>
<tr>
<td><strong>Auto scale graph</strong></td>
<td>To automatically scale the graph, point to one of the following options:</td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Autoscale</em>[XY]* Select Autoscale*[XY]* to automatically scale collected data by scaling the Y height and X width to full screen.</td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Autoscale</em>[X]* Select Autoscale*[X]* to automatically scale collected data by scaling the X width to full screen.</td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Autoscale</em>[Y]* Select Autoscale*[Y]* to automatically scale collected data by scaling the Y height to full screen.</td>
<td></td>
</tr>
<tr>
<td><strong>Zoom Out</strong></td>
<td>Select Zoom Out to Zoom back to the original X and Y ranges which were used to collect the data (i.e. the method setup conditions of the focused scan). For example, the system will revert back to the X and Y ranges set in the method if you zoom in and out during a data collection.</td>
<td></td>
</tr>
<tr>
<td><strong>Single/Multi Graph</strong></td>
<td>Select Single/Multi Graph to toggle between displaying one or all graphs.</td>
<td></td>
</tr>
<tr>
<td></td>
<td><strong>Note:</strong></td>
<td></td>
</tr>
</tbody>
</table>
The graph highlighted in the **Multi Graph** view will be displayed when you toggle the **Single Graph** option.

| **Auto Arrange Graphs** | Select **Auto Arrange Graphs** to view and arrange all graphs simultaneously.  
**Note:** You can move graph boxes manually by clicking the left mouse button outside the graph axes and dragging the graph box to a new position. You can also resize graphs. |
<table>
<thead>
<tr>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>User Data Form</strong></td>
<td>Select <strong>User Data Form</strong> to display the <strong>User Data Form</strong> where you can enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data, such as the pH at which data was collected.</td>
</tr>
<tr>
<td><strong>Calculator</strong></td>
<td>Select <strong>Calculator</strong> to display the <strong>Maths</strong> window where you can perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator.</td>
</tr>
<tr>
<td><strong>History List</strong></td>
<td>This displays a list of the current graphs.</td>
</tr>
</tbody>
</table>

See also:  
Add Label  
Add Picture  
Edit Label  
Edit Picture  
Trace Preferences  
Graph Preferences  
Cursor Modes  
Axes Scales  
Single/Multi Graph  
User Data Form  
Maths  
Graphics toolbar  
Graphics area  
Graphics How To
## Help menu—ADL Shell

Use the **Help** menu to view the online Help and software information for this application.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>ADL Shell Help</td>
<td>Select this to display the ADL Shell Home Page. This contains a list of online Help topics specific to the ADL Shell Application.</td>
</tr>
<tr>
<td>Help Topics</td>
<td>Select this to display the Cary Eclipse Windows Online Help Home Page.</td>
</tr>
<tr>
<td>Tip of the day</td>
<td>View useful tips about using your Cary Eclipse instrument and software.</td>
</tr>
<tr>
<td>Eclipse Tour</td>
<td>Select this to display the Cary Eclipse Multimedia Online Help. This will give you an animated introduction to the Cary Eclipse software and hardware.</td>
</tr>
<tr>
<td>About…</td>
<td>Select <strong>About</strong> to access a dialog that displays information about the application as well as Varian email and Internet contact information. To clear this <strong>About</strong> dialog, select <strong>OK</strong>.</td>
</tr>
</tbody>
</table>
Graphics area—ADL Shell

The Graphics area of the ADL Shell application appears in the top half of the ADL Shell Application window and is used to display traces of collected data. In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area surrounding the graph is referred to as the graph box.

To display or hide the Graphics area, select the Graphics command on the View menu. Double-clicking on the Graphics area enables you to toggle between half or full size display sizes.

You can access the functions associated with the Graphics area by doing the following:

- selecting one of the Graphics toolbar buttons;
- selecting a command from the Graph menu; or
- clicking the right mouse button in the Graphics area and select a command from the menu

**Note**
Not all functions are available on every menu.

You can customize the graphics area to suit your requirements in the following ways:

See also:
- Graph Menu
- Toolbar
- View Menu
- Add Label
- Add Picture
- How to make a graphics template
Graphics Toolbar—ADL Shell

Use the Graphics toolbar buttons to gain quick access to many of the Graph menu options. You can also use the toolbar to select the graph you want to view and to change the look of your graphs.

To display or hide the toolbar, point to the Toolbars option in the View menu and select Graphics. To display or hide the toolbar's text labels, point to the Toolbars option in the View menu and select Text Labels.

**Hot Tip**
To see the name of a button, point to it with the mouse.

The following commands can be accessed from the Graphics toolbar:

<table>
<thead>
<tr>
<th>Button</th>
<th>Menu command</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image" alt="Trace Preferences" /></td>
<td>Trace Preferences</td>
<td>Click this button to display the Trace Preferences window where you can select the trace(s) to display in the selected graph.</td>
</tr>
<tr>
<td><img src="image" alt="Graph Preferences" /></td>
<td>Graph Preferences</td>
<td>Click this button to display the Graph Preferences dialog where you can change the appearance of your graph. Any changes you make to the appearance of the graph will be applied to all graphs.</td>
</tr>
<tr>
<td><img src="image" alt="Cursor Mode" /></td>
<td>Cursor Mode</td>
<td>Click this button to display the Cursor Modes dialog where you can choose the type of cursor to be used on the screen. For example, you can select to use a free cursor that can move anywhere in the graph or exactly trace the focused scan (red scan). Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the Least Squares method or the Point to Point method.</td>
</tr>
<tr>
<td><img src="image" alt="Axes Scales…" /></td>
<td>Axes Scales…</td>
<td>Click this button to display the Axes Scales dialog where you can set the scale range for the X and Y axes.</td>
</tr>
<tr>
<td><img src="image" alt="Autoscale [XY]" /></td>
<td>Autoscale [XY]</td>
<td>Click the Autoscale[XY] button to automatically scale collected data by scaling the Y height and X width to full screen.</td>
</tr>
<tr>
<td><img src="image" alt="Autoscale [X]" /></td>
<td>Autoscale [X]</td>
<td>Click the Autoscale[X] button to automatically scale collected data by scaling the X width to full screen.</td>
</tr>
<tr>
<td><img src="image" alt="Autoscale [Y]" /></td>
<td>Autoscale [Y]</td>
<td>Click the Autoscale[Y] button to automatically scale collected data by scaling the Y height to full screen.</td>
</tr>
<tr>
<td><img src="image" alt="Zoom Out" /></td>
<td>Zoom Out</td>
<td>Click the Zoom Out button to Zoom back to the original X and Y ranges which were used to collect the data (i.e. the method setup conditions of the focused scan). For example, the system will revert back to the X and Y ranges set in the method if you zoom in and out during a data collection.</td>
</tr>
<tr>
<td><img src="image" alt="Add Label" /></td>
<td>Add Label</td>
<td>Click this button to display the Add Label dialog where you can enter a text label to be placed on a graph or in the background of the Graphics area.</td>
</tr>
<tr>
<td><img src="image" alt="Add Picture" /></td>
<td>Add Picture</td>
<td>Click this button to display the Add Picture dialog where you can select a picture, bitmap or chemical structure to be placed on a graph or in the background of the Graphics area.</td>
</tr>
<tr>
<td><img src="image" alt="Single/Multi Graph" /></td>
<td>Single/Multi Graph</td>
<td>Select Single/Multi Graph to toggle between displaying one or all graphs. <strong>Note:</strong> The graph highlighted in the Multi Graph view will be displayed when you toggle the Single Graph option.</td>
</tr>
<tr>
<td><img src="image" alt="Auto" /></td>
<td>Auto</td>
<td>Click the Auto Arrange Graphs button to view and arrange all graphs simultaneously.</td>
</tr>
<tr>
<td>Arrange Graphs</td>
<td>Note: You can move graph boxes manually by clicking the left mouse button outside the graph axes and dragging the graph box to a new position. You can also resize graphs.</td>
<td></td>
</tr>
<tr>
<td>----------------</td>
<td>----------------------------------------------------------------------------------------------------------------------------------</td>
<td></td>
</tr>
<tr>
<td>Add Graph</td>
<td>Click this button to add a new graph box to the Graphics area. You may want to do this if you have, for example, a graph containing existing data and you want to collect some new data in a new graph.</td>
<td></td>
</tr>
<tr>
<td>Remove Graph</td>
<td>Click this button to delete the selected graph box from the Graphics area. The graph box is removed, but the data is not deleted and is still available from the Trace Preferences window. You may want to use remove graph if you have a large number of graph boxes open but only want to focus on some of these.</td>
<td></td>
</tr>
<tr>
<td>Clear All Graphs</td>
<td>Click this button to delete all traces from the graph/s. Note that selecting this option simply turns off the Visible property for the traces in the Trace Preferences window. You can redisplay the traces by making the traces Visible again in the Trace Preferences window.</td>
<td></td>
</tr>
<tr>
<td>User Data Form</td>
<td>Click this button to display the User Data Form where you can enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data, such as the pH at which data was collected.</td>
<td></td>
</tr>
<tr>
<td>Calculator</td>
<td>Click this button to display the Maths window where you can perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator.</td>
<td></td>
</tr>
</tbody>
</table>

See also:
- Add Label
- Add Picture
- Edit Label
- Edit Picture
- Trace Preferences
- Graph Preferences
- Cursor Modes
- Axes Scales
- Single/Multi Graph
- User Data Form
- Maths
- Graphics area
- Graph menu
- Graphics How to
Add Label—ADL Shell

Use the Add Label dialog to place a label on your graph or into the background of the Graphics area.

To add a label to a graph box, you must have at least one trace Visible in the graph box. (You can select which traces you want to make visible in the Trace Preferences dialog.) If you are viewing multiple graphs, the label will be placed in the graph box that is currently selected. However, you can add a label outside of a graph box in the background of the Graphics area at any time.

Q. Can I add a label that will print with all my graphs.
A. Yes, you can add your label to the background of the Graphics area. To do this, right-click on a section of the background (gray area) and select Add Label from the menu. (You will notice that this menu has less options than the menu that appears when you right-click on a graph.) In the Add Label dialog, enter the label to be added to all graphs and select OK. The label will appear in the background of the Graphics area.

To access the Add Label dialog, select the Add Label command from the Graph menu, or select the Add Label button on the Graphics toolbar.

The Add Label dialog comprises the following:

To selected trace
Select this option to associate a label with the selected trace. When you associate a label with a trace and make the trace visible in the Graph box, the label will also be displayed with the trace.

To selected graph
Select this option to associate the label with the selected graph. When you associate a label with a graph, the label will always be displayed in the Graph box, regardless of what traces are visible.

Selected trace
This will identify the trace that is currently selected and display the name of the trace.

Font...
Click Font to open the Windows Font dialog. In this dialog you can specify the font type, style and size to be used for the label.

Label
Use the Label field to enter the text for the label. You can enter as many lines of text as you require.

Hot Tip
If you wish to include certain XY co-ordinates on the graph as a label, position your cursor at the appropriate place, right-click and select Add Label from the menu. When the Add Label dialog appears, the co-ordinate positions will be added to the Label field.

See also:
How to add a label to a graph
How to move a label
How to edit a label
How to delete a label
Add Picture—ADL Shell

Use the Add Picture dialog to insert a picture or bitmap into the Graphics area. The picture can be added as a background picture or to a particular graph box.

To add a picture to a graph box you must have at least one trace Visible in the graph box. (You can select which traces you want to make visible in the Trace Preferences dialog.) If you are viewing multiple graphs, the picture will be placed in the graph box that is currently selected. However, you can add a picture outside of a graph box in the Graphics area at any time.

Q. Can I add a picture that will print with all my graphs.
A. Yes, you can add your picture to the background of the Graphics area. To do this, right-click on a section of the background and select Add Picture from the menu. (You will notice that this menu has less options than the menu that appears when you right-click on a graph.) In the Add Picture dialog, load the picture to be added to all graphs and select OK. The picture will appear in the background of the Graphics area.

To access the Add Picture dialog, select the Add Picture command from the Graph menu, or select the Add Picture button on the Graphics toolbar.

The Add Picture dialog comprises the following commands:

To selected trace Select this option to associate a picture with the selected trace. When you associate a picture with a trace and make the trace visible in the Graph box, the label will also be displayed with the trace.

To selected graph Select this option to associate the picture with the selected graph. When you associate a picture with a graph, the label will always be displayed in the Graph box, regardless of what traces are visible.

Load Click Load to open the Windows Open dialog. Use this dialog to browse your computer for the picture you want to insert. The picture must be one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.

If you are viewing multiple graphs the picture will be placed on the graph that is currently selected. If no graph is selected, the picture will be placed on the Graphics area background.

Note When you save the data (as a Batch or Data file) the label is saved with its associated trace or graph. Labels outside graph boxes can be saved in a graphics template.

See also: How to add a picture to a graph How to move a picture How to delete a picture How to replace a picture
Edit Label—ADL Shell

Use the **Edit Label** dialog to alter the content or look of your label.

To access the **Edit Label** dialog, right-click on the label you wish to change and select **Edit Annotation** from the menu.

The dialog comprises the following:

- **Font**: Select **Font** to open the **Font** dialog, where you can specify the font type, style and size to be used for the label.

- **Label**: Use the **Label** field to alter the text for the label.

See also:
- [How to edit a label](#)
- [How to add a label to a graph](#)
- [How to move a label](#)
Edit Picture—ADL Shell

Use the Edit Picture dialog to change a picture you have inserted into the Graphics area.

To access the Edit Picture dialog, right-click on the picture you wish to change and select Edit Annotation from the menu.

The Edit Picture dialog comprises the following commands:

- **To selected trace/ graph**: Displays whether you originally selected to add the picture to a trace or a graph. This option is not editable.
- **Load**: Click Load to open the Windows Open dialog. Use this dialog to browse your computer for the picture you want to insert. The selected picture will replace the current picture. The picture must be one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.

See also:
- How to replace a picture
- How to add a picture
- How to move a picture
- How to delete a picture
Axes Scales—ADL Shell

Use the **Axes Scales** dialog to manually change the axes range. This can assist in zooming in on a particular part of the graph.

To access the **Axes Scales** dialog, select the **Axes Scales** command from the **Graph** menu, or select the **Axes Scales** button on the Graphics toolbar.

The dialog comprises the following:

**X Axis**

- **Mode**
  View the current units for the X axis.

- **Scale**
  Use the **Scale** field to enter a new range for the X axis, in the format n1 - n2 (where n1 is the start of the scale and n2 is the end of the scale).
  
  **Hot Tip**
  The Cary Eclipse will store the ten most recently entered ranges, which you can access by selecting the down arrow adjacent to the **Scale** field.

**Y Axis**

- **Mode**
  View the current units for the X axis.

- **Scale**
  Use the **Scale** field to enter a new range for the Y axis, in the format n1 - n2 (where n1 is the start of the scale and n2 is the end of the scale).
  
  **Hot Tip**
  The Cary Eclipse will store the ten most recently entered ranges, which you can access by selecting the down arrow adjacent to the **Scale** field.

See also:

- Graph menu
- Graphics toolbar
- How to scale your graph manually
- How to scale your graph automatically
- How to zoom a graph
Cursor Modes—ADL Shell

Use the Cursor Modes dialog to choose the type of cursor to be used on screen. For example, you can select to use a free cursor that can move anywhere in the graph.

To access the Cursor Modes dialog, select the Cursor Modes command from the Graph menu, or select the Cursor Modes button on the Graphics toolbar.

The dialog comprises the following:

Cursor Modes

Free
The Free cursor is not confined to a particular trace on the graph. It can move anywhere in a graph box, and its ‘X,Y’ position is shown in the bottom right-hand side of the Graphics area. You can use the Free cursor to zoom in on a particular part of a graph.

Note
The Free cursor moves from pixel point to pixel point in the selected graph box. Hence you need to be careful when positioning the cursor over a scan to obtain an accurate (X,Y) reading. It is recommended that you use the Track cursor mode for this.

Track
Use the Track cursor to exactly trace the focused (red) scan in a graph box. The cursor position is displayed in the X and Y at the bottom right-hand side of the Graphics area.

Hot Tip
If you wish to fix the cursor on a particular position on the curve, move the cursor to the required position. Click the left mouse button, press ALT on the keyboard, then release the mouse button.

Track Intensity
Select the Track Intensity option if you want to associate an intensity value with the tracking. The cursor will then track from point to point on the continuum between the intensity value you enter. The default value will be 50% of the maximum intensity of the focused continuum.

Peaks/Valleys
Choose from the options of Peaks, Valleys or Peaks Valleys to move the cursor from peak to peak, valley to valley or from one peak/valley to the next.

See also:
Graph menu
Graphics toolbar
How to zoom a graph
How to track the focused trace
Example of a Graphics Template—ADL Shell

By using the Add Label command in the Cary Eclipse graphics system, you can customize a graphics template such as the one shown below:

The above is created as follows:

- The graph on the left is the collected data.
- The text on the top right is a multi-line label (added using the Add Label command).
- The logo is an inserted picture (added using the Add Picture command).

See also:
Add Label
How to make a graphics template
Graph Preferences—ADL Shell

Introduction

Use the Graph Preferences dialog to change the appearance of your graph.

All graphs will appear labeled with the X axis label being horizontal and the Y axis vertical. You cannot change the orientation of these labels. However you can alter the font and size of the axes and their labels.

Any changes you make to the appearance of the graph will apply to all graphs until the settings are changed again.

To access the Graph Preferences dialog, select the Graph Preferences command from the Graph menu, or select the Graph Preferences button on the Graphics toolbar.

Dialog controls

The Graph Preferences dialog comprises the following:

Axes Preferences

Axes Color
Select the color that you want to assign to the axes of the graph/s from the drop down list.

Axes Width
Select the width of the axes from the drop down list. The width, which is in pixels, ranges between one and five pixels.

Grid Style
Select the style of grid for the graph from the drop down list. If you do not wish to include a grid on the graph, select None.

Plot Area Color
Select the color to be used as the background of the graph from the drop down list. If you do not want a background color on the graph, select White.

Trace Width
Select the width of the trace from the drop down list. The width is given in pixels and ranges between one and five pixels.

Font
Click the Font button to display the Windows Font dialog where you can define the font style for the axes of the graph/s. The style and size of the font will be applied to all graph. The style, name and point size of the font appears on the font button.

Preview Box
Preview any changes you make to the axes preferences in the preview box. The changes shown in this box will not be made until you select OK.

Advanced Options

Auto tick marks
The Auto tick marks option enables you to approximately set how many tick marks (or points) will be included on the graph axes. The tick marks generated will be whole, rounded, even numbers. The Auto tick marks option is not available if you have selected Major ticks.

Y-axis
Use the Y-axis slider bar to increase or decrease the number of tick marks (or points) to be included on the Y axis.

X-axis
Use the X-axis slider bar to increase or decrease the number of tick marks (or points) to be included on the X axis.

**Major ticks**
Use the **Major Ticks** option to enable you to set exactly how many ticks (or points) will be added to the X and Y axes. The number of marks you specify will be divided by the axis range, which can result in odd numbers on the axis. Note that any changes are not reflected in the graph bitmap at the top of the screen.

**Decimals**
Use the **Decimals** option to set the number of X and Y decimal places to be displayed on the axes. To set the number of decimal places, select the **Decimal** option and enter the number in the Y-axis and X-axis combo boxes. Note that any changes are not reflected in the graph bitmap at the top of the screen.

**Trace Style**

**Points**
Select **Points** to display each individual data point collected by the Cary Eclipse on the graph.

**Points and solid lines**
Select **Points and solid lines** to display each individual data point collected by the Cary Eclipse on the graph. Additionally, an interpolated line that connects the points will be drawn.

**Solid lines**
If you select **Solid lines**, the Cary Eclipse system will interpolate the data between the actual, collected data points and show the data as a continuous line.

**Style cycles automatically**
If you select **Style cycles automatically**, the Cary Eclipse system will display each of the above three trace styles. This makes it easier to differentiate between traces, especially on black-and-white printers.

**Show Legend**

**Show Legend**
Select **Show Legend** to display a legend in the right side of each graph box. A single left click on a sample name or example line within the legend will make it the focused or red scan. Information about the trace will be displayed below the X axis.

**Hot Tip**
Double-clicking on the legend will automatically display the **Trace Preferences** window, allowing you to view more detailed information about your trace (e.g. audit log, File name, etc.). You can also use the legend to select which trace will be the focused trace.

See also:
- Graph Menu
- Toolbar
- Graphics area
- Graphics How To
Single/Multi Graph—ADL Shell

You can use the **Single/Multi Graph** command to toggle between a display of either one or multiple graph boxes in the Graphics area.

The command is accessible from the **Graph** menu or by selecting the **Single/Multi Graph** button from the Graphics toolbar.

*Example of a single graph box display*

*Example of a multi graph box display*

**Note**

Up to 100 graphs can be displayed in the Graphics area at the one time.

**See also:**

- Graph Menu
- Graphics toolbar
- Graphics How To
Trace Preferences—ADL Shell

A single graph box can display many traces. This window enables you to select the trace(s) to display (make Visible) in the selected graph box.

To access the Trace Preferences dialog, select the Trace Preferences command from the Graph menu, or select the Trace Preferences button on the Graphics toolbar.

The dialog comprises the following:

Hot Tip
The Trace Preferences menu is also available on clicking the right mouse button anywhere in the trace listing.

Trace Preferences table

Visible
Use the Visible field to select which traces will appear on the selected graph. To select or clear the Visible field, simply click the left mouse button in this column. A tick will appear next to traces that are to be displayed in the selected graph.

If you are in single graph view, all selected traces will appear on the one graph. If you are in Multi Graph view the traces will appear on the graph that is currently selected.

Hot Tip
To make all traces Visible, right-click on the table and select the Select All command from the menu. Then right-click on the table again and select the Show Traces command from the menu.

Color
The Color field enables you to view the color of each trace that appears on the selected graph. To change the color of a trace, highlight it, click the right mouse button in the Trace Preferences table and select Change Trace Color. You can then select a color from the Color dialog that is displayed.

The red trace in any graph box is the one on which all information appears. This trace, which is used for the cursor tracking, is called the focused trace and it is always red. When you select a trace in the Color column it will become the focused trace in the selected graph box.

Note
When you change the trace line width in the Graph Preferences dialog it also will change the width of the color trace line in this dialog.

Name of trace
The Name of Trace field contains a description of the traces currently available for display in the selected graph box.

Filename
This field displays the filename and path for the trace. If the name is too long to appear in the field you can use the keyboard arrow keys to scroll to the end of the filename or you can resize the Trace Preferences window. If the file path for a trace shows 'not saved' then the trace is unsaved data and is only stored in memory.

Trace Audit Log

The Trace Audit Log displays information about the highlighted trace. This includes instrument parameters and data form information relating to the trace that has been selected. It also logs all information about data manipulation. When more than one trace is highlighted, the Trace Audit Log will display information about the trace that was first selected.
See also:
- Graph menu
- Graphics toolbar
- Graph Preferences
- Trace Preferences menu
- How to make multiple traces visible in a graph
- How to remove a trace from the trace list
- How to remove a trace from a graph
Trace Preferences menu—ADL Shell

The Trace Preferences menu is displayed when you click the right mouse anywhere on the Trace Preferences table. The menu contains the following commands:

<table>
<thead>
<tr>
<th>Command</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Select All</td>
<td>This selects or highlights all the traces in the listed on the Trace Preferences page.</td>
</tr>
<tr>
<td>Select None</td>
<td>This de-selects all the traces in the listing.</td>
</tr>
<tr>
<td>Show Traces</td>
<td>Select Show Traces to make all of the traces currently highlighted Visible, meaning that they will be displayed in the current graph box.</td>
</tr>
<tr>
<td>Hide Traces</td>
<td>Select Hide Traces to remove the Visible property from all the traces currently highlighted, which will remove them from the current graph box.</td>
</tr>
<tr>
<td>Delete Traces</td>
<td>Select Delete Traces to delete all the highlighted traces in all graphs. Note that if you save the data file before you select Delete Traces, the traces are not deleted from the saved file, and they can be retrieved at a later date. Selecting Delete Traces will delete the traces from the application memory, and it is recommended that you select this option if you need to free up memory on your computer. This is especially relevant for the ADL Shell application where the data files can be quite large.</td>
</tr>
<tr>
<td>Change Trace Color</td>
<td>Select Change Trace Color to display the Windows Color dialog where you can change the color of the currently highlighted trace(s). The focused trace will remain red while it retains the focus, and will revert to the selected color when the focus is moved.</td>
</tr>
</tbody>
</table>

**Note**

If the application is controlled by the GLP Administration application, you will be prompted to save an unsaved data file before it is removed from the application.

See also:
- Trace Preferences
- Graph Preferences
- How to remove a trace from the trace list
- How to remove a trace from a graph
User Data Form—ADL Shell

Use the **User Data Form** to enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data.

To access the **User Data Form**, select the **User Data Form** command from the **Graph** menu, or select the **User Data Form** button on the **Graphics** toolbar. The **User Data Form** command is only available if you have selected a graph box.

The dialog comprises the following:

- **Data Name**: This column lists the names of each visible trace in the highlighted graph box.

  Use the extra columns for recording additional information about the experimental conditions you used to collect the data, eg. pH.

- **Other Columns**: Use this to record the name of the parameter in which you are interested as a column heading, (for example, pH, ionic concentration, substrate concentration etc.).

  **Note**: You must enter a column heading, or else the data in that column will not be saved.

See also:
- **Graph menu**
- **Graphics toolbar**
Function keys—ADL Shell

The function keys at the top of your keyboard perform basic Cary Eclipse operations simply and quickly.

<table>
<thead>
<tr>
<th>Function key</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>This displays the Help for the current dialog or window.</td>
</tr>
<tr>
<td>F4</td>
<td>Press F4 to display the Go to Wavelength dialog.</td>
</tr>
<tr>
<td>F5</td>
<td>Press F5 to perform an instrument <strong>Zero</strong>. The current intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before selecting <strong>Start</strong> if you want your data to start at 0 intensity. This command is also accessible from the Commands menu or by selecting the Zero button. <strong>Note</strong> Before you select <strong>Zero</strong>, ensure that you have placed the blank solution in the cell positions that are currently in the cell path. Select the <strong>Clear Zero</strong> command from the Commands menu to remove zero correction.</td>
</tr>
<tr>
<td>F6</td>
<td>Press F6 to display the <strong>Rapid Result</strong> dialog where you can read a blank and multiple sample readings without changing the application method. This command is also accessible from the Commands menu.</td>
</tr>
<tr>
<td>F9</td>
<td>If the application is online, press F9 to start a data collection. This is equivalent to selecting the Start button. If the application is not online, you will need to press F9 to first connect to the Cary Eclipse. Then press F9 to start the data collection. This command is also accessible from the Commands menu or by selecting the Start button. <strong>Note</strong> You will not be able to perform a <strong>Connect</strong> if another application is collecting data.</td>
</tr>
<tr>
<td>F12</td>
<td>Select this to stop the current measurement. This is equivalent to selecting the <strong>Stop</strong> button. This command is also accessible from the Commands menu or by selecting the Stop button.</td>
</tr>
</tbody>
</table>

See also:  
Go To Wavelength  
Zero  
Rapid Result  
Stop Instrument Button
Instrument Status Readings—ADL Shell

Ordinate status
This double-height field displays the current intensity reading (ordinate value). It is continually updated during a data collection.

Abscissa status
This double-height field displays the current wavelength reading (abscissa value) in nanometres.

See also:
Status line
Print—ADL Shell

Use the Print dialog to select the printer and pages to print of the currently displayed report as well as the number of copies you wish to print.

To access the Print option, select the Print command from the File menu.

All graphical information displayed on screen will be printed with the report, therefore it is advisable to edit the report on the screen prior to printing. You can also preview the report prior to printing by using the Print Preview command.

See also:
- Edit Report
- Print Preview
Print Preview—ADL Shell

Use the Print Preview dialog to view the current report before you print it. From the File menu, select Print Preview to access the dialog.

The contents of the Graphics area appear first in the preview area followed by the contents of the Report area. You can reduce or enlarge the print preview on screen. To do this, click the left mouse button in the preview area to toggle through the five available views. Click the right mouse button in the preview area to return to the full page view.

The page comprises the following commands:

- **First arrow** Use this arrow to view the first page of the report.
- **Previous arrow** Use this arrow to view the previous page of the report.
- **Next arrow** Use this arrow to view the next page of the report.
- **Last arrow** Use this arrow to view the last page of the report.

**Q. Why are the arrow buttons grayed out?**

**A.** The buttons will be grayed out if the report has only one page of data.

- **Print** Use this to display the Windows Print dialog where you can choose the printer and pages to print as well as the number of copies you wish to print.

See also:

File menu
Report Toolbar—ADL Shell

Use the Report toolbar buttons to gain quick access to many of the Edit menu options. You can also use the toolbar to cut, copy and paste information and to change the look of your report.

To display or hide the toolbar, point to the Toolbars option in the View menu and select Reports. To display or hide the toolbar’s text labels, point to the Toolbar option in the View menu and select Text Labels.

**Hot Tip**
To see the name of a button, point to it with the mouse.

The following commands can be accessed from the Reports toolbar:

<table>
<thead>
<tr>
<th>Button</th>
<th>Menu command</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image" alt="Edit Report" /></td>
<td><strong>Edit Report</strong></td>
<td>Click this button to edit the report. This command is also accessible by pressing CTRL+R.</td>
</tr>
<tr>
<td><img src="image" alt="Undo" /></td>
<td><strong>Undo</strong></td>
<td>If available, this undoes the last action. This command is also accessible by pressing CTRL+Z.</td>
</tr>
<tr>
<td><img src="image" alt="Cut" /></td>
<td><strong>Cut</strong></td>
<td>Click this button to remove the highlighted text and place it on the Windows Clipboard. This command is also accessible by pressing CTRL+X.</td>
</tr>
<tr>
<td><img src="image" alt="Copy" /></td>
<td><strong>Copy</strong></td>
<td>Click this button to copy the highlighted text onto the Windows Clipboard. This command is also accessible by pressing CTRL+C.</td>
</tr>
<tr>
<td><img src="image" alt="Paste" /></td>
<td><strong>Paste</strong></td>
<td>Click this button to insert the contents of the Windows Clipboard (text only) at the cursor. Any highlighted text will be replaced. This command is also accessible by pressing CTRL+V.</td>
</tr>
</tbody>
</table>
| ![Delete](image) | **Delete** | Click this button to remove selected text in the Report area. This command is also accessible by pressing DEL.  
**Note** This option is only available once you select Edit Report and there is text highlighted in the Report area. |
| ![Font](image) | **Font** | Click this button to change the font of new or selected text in the report. For existing text, simply highlight the text in the report, select this button and make your changes in the Font dialog. When you select OK, the appearance of the highlighted text in the report will alter to reflect your font changes. This command is also accessible by pressing CTRL+F. |
| ![Preview](image) | **Preview** | Click this button to display the Print Preview dialog where you can view the current report as it will be printed. |

See also:  
[Print Preview](#)
Right mouse menu—ADL Shell

The following commands are accessible by clicking the right mouse button on editable fields in the Cary Eclipse software:

- **Undo**: If available, this reverts to the previous value before the Edit on the field.
- **Cut**: If available, this cuts the highlighted text to the clipboard.
- **Copy**: If available, this copies the highlighted text to the clipboard.
- **Paste**: If available, this pastes the text from the clipboard to the cursor position.
- **Delete**: If available, this deletes the highlighted text/number.
- **Select All**: If available, this selects all entries.
Status line—ADL Shell

The Status Line is displayed at the very bottom of the ADL Shell Application window. The line will display the following information (from left to right):

- **Hardware Status**: This field displays status messages indicating the current instrument activity.
- **Sequence Information**: This field displays status messages indicating the current system activity.
- **Current Results**: This field displays information on the last reading.

ADL Shell Application window
Stop Instrument button—ADL Shell

Select the **Stop** button to end the current data collection.

**Note**

The ADL Shell does not contain a Start button. You will need to program this button and its functionality in your Shell application.
**ADL Hooks—ADL Shell**

The **ADL Hooks** command is accessible from the View menu.

Instead of writing a full ADL program you can use ADL hooks to link into an existing application. This is particularly useful if you want to make small changes to the way an application works, e.g. you want to pop up a dialog for the user to enter some specific data before the run starts. ADL hooks are links at specific points in an application at which you can run an ADL program. For example, ADL hooks have been supplied to run the following ADL programs:

**Note**
Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.

<table>
<thead>
<tr>
<th>Hook</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>AD_PreSeq.ADL</td>
<td>This runs when the Start button is pressed.</td>
</tr>
<tr>
<td>AD_PostSeq.ADL</td>
<td>This runs at the end of the Start button sequence.</td>
</tr>
<tr>
<td>AD_Online.ADL</td>
<td>This runs when Cary Eclipse is detected online.</td>
</tr>
<tr>
<td>AD_OffLine.ADL</td>
<td>This runs when Cary Eclipse ‘goes’ offline.</td>
</tr>
<tr>
<td>AD_Zero.ADL</td>
<td>This runs when the Zero button is pressed.</td>
</tr>
<tr>
<td>AD_Recalc.ADL</td>
<td>This runs when the Recalc button is pressed.</td>
</tr>
<tr>
<td>AD_Shutdown.ADL</td>
<td>This runs when the application is shutting down.</td>
</tr>
<tr>
<td>AD_PreRead.ADL</td>
<td>This runs just before a read is taken.</td>
</tr>
<tr>
<td>AD_PostRead.ADL</td>
<td>This runs just after a read is taken (It has the reading result stored in the UserResult variable.)</td>
</tr>
<tr>
<td>AD_PreSetup.ADL</td>
<td>This runs when the Setup button is pressed.</td>
</tr>
<tr>
<td>AD_PostSetup.ADL</td>
<td>This runs when the OK button on the Setup dialog is pressed.</td>
</tr>
<tr>
<td>AD_CellChange.ADL</td>
<td>This runs instead of the Cary Eclipse driving the Multicell Holder. It enables you to add your own instructions for the Cary Eclipse Multicell Holder or to use your own cell changing accessory.</td>
</tr>
</tbody>
</table>

See also:
[View menu](#)

**ADL Command Line—ADL Shell**

**Introduction to ADL**

Use the ADL Command Line to run Single-line or Multi-line Applications Development Language (ADL) commands that enable you to perform functions over and above that of the existing program functionality.
ADL is not a general purpose programming language. It has been developed specifically by Varian to meet the needs of spectroscopists. It can be used to handle all the data collected from the Cary Eclipse—both photometric values and entire data collections. ADL programs that you develop should be stored with the extension 'ADL'.

To access the ADL Command Line, select the ADL Command item from the Commands menu. You can also press ALT+Y to open the dialog.

**Note**
Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.

### Software controls

**Single Line**
Selecting this lets you enter ADL commands on a single line into the Command Line entry box.

**Q.** Can I enter more than one ADL command on a single line?
**A.** Yes, provided that the commands are separated by a semicolon ‘;’.

**Multi-line**
The Multi-line capability enlarges the Command Line entry box. Use the Multi-line capability to create a more complex ADL program by adding a series of commands over multiple lines. When Multi-line mode is selected, the Quick Reference option becomes available.

**Quick Reference**
Select Quick Reference to open the Quick Reference dialog where you can easily add a command in the correct syntax to your ADL program. This option is only available for Multi-line commands.

[How to insert ADL commands using the Quick Reference](#)

A detailed description of all the commands in the Quick Reference is available by pressing the Help button on the Quick Reference dialog.

**Browse**
Select **Browse** to access the Open dialog where you can browse for and select an existing ADL program to load and run.

**Command Line (entry box)**
This entry box enables you to use ADL commands. You can enter single ADL commands into this field, or you can enter a series of ADL commands. If you have the entry type set to Single Line, you can review previous ADL entries by selecting the down arrow to the right of the list box.

The ADL commands in the Command Line are executed when you select OK.

**Note**
On entering this window, the Cary Eclipse will recall the last command or program executed from the Command Line entry box. The ADL commands entered in the last executed program are typically stored in the Varian\Cary Eclipse WinFLR\ADL directory on the drive on which the Cary Eclipse software
is installed.

**Hot Tip**

You can rerun the ADL commands in the Command Line entry box from any screen in the Kinetics application by pressing Alt-X or selecting **Re-run ADL** from the **Commands** menu.

See also:
- [Commands menu](#)
- [ADL Hooks](#)
ADL Shell How To—ADL Shell

Select the procedure you require below for a step-by-step informative guide on running the application software.

**ADL Shell How To**

- How to use advanced reporting options
- How to export collected data
- How to insert ADL commands using the Quick Reference

**Graphics How To**

- How to alter the look of your graphs
- How to make a graphics template

**Traces**

- How to view a particular trace
- How to select the focused trace
- How to track the focused trace
- How to display a trace that is not appearing in a graph box
- How to track the focused trace
- How to remove a trace from the trace list

**Sizing and scaling**

- How to resize graph boxes
- How to scale your graph manually
- How to scale your graph automatically
- How to zoom a graph

**Labels**

- How to add a label to a graph
- How to edit a label
- How to move a label
- How to delete a label

**Pictures**

- How to add a picture
- How to replace a picture
- How to move a picture
- How to delete a picture

ADL Shell Home Page Index
How to insert ADL commands using the Quick Reference—ADL Shell

The Quick Reference window is accessed from the ADL Command window. It enables you to easily add a command in the correct syntax to your ADL program. To do this:

1. Press Alt-Y to open the ADL Command Line window.
2. Select the Multi-line option. The Command Line entry box will expand to take multiple lines of ADL commands and the Quick Reference check box will appear.
3. Select the Quick Reference check box. The Quick Reference window will appear.
4. If necessary, use the Go To options to jump to the appropriate part of the Quick Reference listing.
5. Highlight the command you require in the Quick Reference.
6. Click the Insert into ADL button. The selected ADL command will appear in the Command Line entry box.
7. Where necessary, alter the defaults supplied by the inserted Quick Reference command in the Command Line entry box to suit your program.
8. Repeat steps 4–7 to add more commands as required.

How to export collected data—ADL Shell

If you want to export data from a Cary Eclipse application into a third-party application, then you simply need to save your data as an ASCII *.CSV file. To do this:

Step 1

Click on the File menu item.

Step 2

Click on Save Data As. The Save As dialog will appear.

Step 3

Click on the down arrow next to the Files of type list box.

Step 4

Select Spreadsheet ASCII (*.CSV).

Step 5
Type your file name into the File name field.

**Step 6**

Press Save to export your data.

**How to use advanced reporting options—ADL Shell**

When you have programmed your ADL Shell reporting options, or when you open an existing file and prepare to print the report, you should be aware of some of the ADL Shell's advanced reporting options.

Options available include:

- Including header and footer information
- Including a logo
- Editing the text in the report
- Viewing the report on screen before printing
- Cutting and pasting to a word processor

Including header and footer information

You can have text appearing at the top and bottom of every page in your report. To specify what the text should be, double-click on the button in the System tray of the Windows Taskbar to display the Company tab in System Information.

Including a logo

You can include a logo on the first page of your report. To specify what the logo should be, double-click on the button in the System tray of the Windows Taskbar to display the Company tab in System Information. If a logo is specified then it will always be included in reports.

**Q.** Why isn't the logo I selected included in my report?

**A.** If you are running the Cary Eclipse software under Windows NT you need to be logged on as an Administrator in order to specify the logo in the System Information application.

Editing the text in the report

You can edit any of the text in the Report area. Just select Edit Report from the Edit menu. You can then add, delete and change text in the Report area.

**Q.** The Edit Report option is not available, why not?

**A.** You do not have reporting editing privileges set in the GLP Administration application. The Administrator of the system will have to change your privileges before you can edit reports.

Viewing the report on the screen before printing

Use the Print Preview selection on the File menu to view the report on screen before printing it. This allows you to get the report right before you print it on paper. Use the arrow keys at the bottom of the Print Preview screen to change pages.
Cutting and pasting to a word processor

To have complete control over how your report looks you may need to export it to a word processor. To do this:

Step 1
Generate a report in your ADL Shell application.

Step 2
Select Edit Report on the Edit menu.

Step 3
Click in the Report area and click and drag to highlight the text you want to export to your word processor.

Step 4
Right click on the Report area and select Copy from the menu.

Step 5
Start up your word processor and use the Paste function (usually under the Edit menu) to place the text into a document.

Step 6
Change back to the ADL Shell. If you want to add a picture of the graphics to your report, you need to set up the graphics the way you would like them to appear and then right click on the graphics area. Select Copy graph from the menu.

Step 7
Change back to your word processor and select Paste again. The graphics will appear as a bitmap.

See also:
How to add a label to a graph
How to add a picture to a graph
How to alter the look of your graphs
How to display a trace that is not appearing

Graphics How to…—ADL Shell

Traces

• How to track the focused trace
• How to select the focused trace
How to display a trace that is not appearing in a graph box
How to remove a trace from a graph
How to remove a trace from the trace List

Sizing and scale

How to resize graph boxes
How to scale your graph manually
How to scale your graph automatically
How to alter the look of your graphs
How to zoom a graph

Labels

How to add a label to a graph
How to edit a label
How to delete a label
How to move a label

Pictures

How to add a picture to a graph
How to move a picture
How to delete a picture
How to replace a picture

How to add a label to a graph—ADL Shell

You can add a label to your graph or Graphics area background. To do this:

Step 1

Click on the graph box you require or click outside the graph box(es) to choose the Graphics area background.

Note
There must be at least one trace Visible in the graph box before you can add a label to it.

Step 2

Select Add Label from the Graph menu or click the Add Label button on the toolbar. The Add Label dialog will appear. You can also access the Graph menu to add a label by clicking the right mouse button anywhere in the Graphics area.

Step 3

Click the Font button. The Windows Font dialog will appear.
Step 4

Select the font type, size and style you require for your label. A sample of your label font appears in the Sample field.

Step 5

Click the OK button when you are satisfied with your selections and return to the Add Label dialog.

Step 6

Enter the label in the text entry field and press OK.

Note
If you are viewing multiple graphs the label will only be placed on the graph that is currently selected.

If necessary, you can move a label so that it is more appropriately placed on your graph.

See also:
How to move a label
How to edit a label

How to add a picture to a graph—ADL Shell

You can add a picture to your graph or the Graphics area background. To do this:

Step 1

Click on the graph box you require or click outside the graph box(es) to choose the Graphics area background.

Note
There must be at least one trace Visible in the graph box before you can add a picture to it.

Step 2

Select Add Picture from the Graph menu or click the Add Picture button on the toolbar. The Add Label dialog will appear. You can also access the Graph menu to add a picture by clicking the right mouse button anywhere in the Graphics area.

Step 3

Click the Load button. The Windows Open dialog will appear.

Step 4

Browse through your directories to find the picture you want to insert. All pictures must be in the bitmap format and will have the extension .BMP.
Step 5

Highlight the .BMP that you want to insert and click Open.

Step 6

The Add Picture dialog will appear showing a preview of the bitmap. If the bitmap is the correct one click the OK button. The bitmap will be placed onto the graph.

Note
You should take care not to add a bitmap that is too big as it will obscure part or all of your graph.

You can move a picture so that it is more appropriately placed on your graph.

See also:
How to move a picture to a graph
How to delete a picture
How to replace a picture

How to alter the look of your graphs—ADL Shell

Step 1

Click the button on the toolbar, or select Graph Preferences… from the Graph menu, or click the right mouse button in a graph box and select Graph Preferences…. The Graph Preferences dialog will appear.

Step 2

From the options available there, choose your preferred appearance for the graphs. A preview box in the top right-hand corner will demonstrate how the graphs will appear.

Step 3

Click OK, and all graphs will change to reflect your choices.

Note
You can also add labels and pictures to your graphs to alter their appearance.

Step 4

To save your graphics setup for re-use at a later date, save it as a graphics template.

See also:
How to add a label to a graph
How to add a picture to a graph
How to move a label
How to delete a label—ADL Shell

You can delete a label from a graph or Graphics area background by doing the following:

Step 1

Position the cursor on the label and click the left mouse button to select it.

Hot Tip
If you are having trouble selecting the label it may be that a trace is interfering with the label selection. Try zooming in and clicking in a different part of the label.

Step 2*

Click with the right mouse button.

Step 3

From the menu select Delete Annotation… The label will be removed from your graph or Graphics area background.

* An alternative to Steps 2 is to click on the Graph option in the menu bar.

See also:
How to add a label to a graph
How to move a label
How to edit a label

How to delete a picture—ADL Shell

You can delete a picture from a graph or Graphics area background by doing the following:

Step 1

Position the cursor on the picture and click the left mouse button to select it.

Hot Tip
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2*

Click with the right mouse button.

Step 3
From the menu select Delete Annotation… The picture will be removed from your graph or Graphics area background.

* An alternative to Steps 2 is to click on the Graph option in the menu bar.

See also:
- How to add a picture to a graph
- How to move a picture
- How to zoom a graph

**How to display a trace that is not appearing in the graph box—ADL Shell**

**Step 1**

Click on the graph in the Graphics area.

**Step 2**

Click the button on the toolbar, or select Trace Preferences… from the Graph menu, or click the right mouse button in a graph box and select Trace Preferences… The Trace Preferences dialog will appear.

**Step 3**

Find that trace that you wish to see and click on the Visible option. A tick should appear. Alternatively, for multiple traces you can:

a. Hold down the CTRL key click on all the traces that you want to display.

b. Click the right mouse button to display the Trace Preferences menu.

c. Select Show Traces to turn on the Visible option for each file.

**Step 4**

Press OK to exit the Trace Preferences window. The desired trace will appear.

See also:
- How to remove a trace from a graph
- How to remove a trace from the trace list

**How to edit a label—ADL Shell**

**Step 1**

Place the cursor on the label and click the left mouse button to select the label.

**Hot Tip**
Step 2

Click the right mouse button and select **Edit Annotation** from the drop-down menu. Alternatively you can select the **Edit Annotation** command from the **Graph** menu. The **Edit Label** dialog is displayed.

Step 3

Use the **Edit Label** dialog to change the label.

For example, if you want to change the font size of the label, select the **Font** button. In the **Size** field, scroll through the available sizes for the font and select the size you require. Alternatively, highlight the currently selected size in the **Size** entry field and enter the size you require. Click the **OK** button in this dialog to return to the **Edit Label** dialog.

Step 4

Click **OK** to clear the **Edit Label** dialog. Your label will appear with the new font size.

See also:
- How to add a label to a graph
- How to move a label
- How to delete a label

**How to make a graphics template - ADL Shell—ADL Shell**

A graphics template lets you set up and store the style of a graphics report for future use. You can do this by adding labels and pictures to the background of the Graphics area and altering any axes styles etc. One advantage of this over the application's built-in reporting features is that you have more flexibility as to where you can position graph boxes and any text that you may want to appear (e.g. operator name, batch number etc.) and also where you add pictures, e.g. company logos etc.

Step 1

Start the application and open the data file from which you want to make a graphics template. Alternatively, collect some new data.

Step 2

Make as many graph boxes as you require in your template by using the **Add Graph** function.

Step 3

Adjust the graph boxes to the correct size and position them as you want them to appear in the template.

Step 4

Do any of the following, if required:
- Change the appearance of the graph boxes in the Graph Preferences dialog
- Use the Add Label function to append text to the Graphics area
- Use the Add Picture function to add pictures, such as your company's logo to the Graphics area

Step 5

From the **File** menu, select **Print Preview**. Use the **Print Preview** dialog displayed to check the template that you have created.

Step 6

When you are satisfied with the appearance of the graphics, select the **Save As…** command from the **File** menu. The **Save As** dialog is displayed.

Step 7

A list of stored data files will appear. Select the down arrow at the right of the **Files of type** field and select 'Graph Template' to list all the template files. Ensure that the **Save only focused trace** check box is cleared.

Step 8

Enter a name for the template in the **File name** field.

Step 9

Select **OK** to save the graphic template. The template will be used each time you create a new file.

**Note**

You can also make a graphic template in a similar manner from existing data. No data will be saved in the template.

**See also:**
- Example of a Graphics Template
- How to add a label to a graph
- How to add a picture to a graph
- How to move a label
- How to move a picture

**How to move a label—ADL Shell**

**Step 1**

Place the cursor on the label.

**Step 2**

Click and hold down the left mouse button.

**Hot Tip**
Step 3

Drag the label to the appropriate position on the graph or Graphics area background.

Step 4

Release the mouse button. The label will appear at the new position.

Note

Labels are transparent so as not to interfere with any traces on graphs.

See also:

How to add a label to a graph
How to edit a label
How to delete a label

How to move a picture—ADL Shell

You can move a picture so that it is more appropriately placed on your graph or Graphics area background.

Step 1

Place the cursor on the picture.

Step 2

Click and hold down the left mouse button.

Hot Tip

If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 3

Drag the picture to the correct position.

Step 4

Release the mouse button.

See also:

How to add a picture to a graph
How to delete a picture
How to replace a picture
How to remove a trace from a graph—ADL Shell

Step 1
Click on the graph in the Graphics area.

Step 2
Click the button on the toolbar, or select Trace Preferences… from the Graph menu, or click the right mouse button in a graph box and select Trace Preferences…. The Trace Preferences dialog will appear.

Step 3
Find that trace that you no longer wish to see and click on the Visible option. The tick should disappear. Alternatively, for multiple traces you can:

a. Hold down the CTRL key click on all the traces that you no longer want to display.

b. Click the right mouse button to display the Trace Preferences menu.

c. Select Hide Traces to turn off the Visible option for each file.

Step 4
Press OK to exit the Trace Preferences window. The sample trace will be removed from the graph.

See also:
How to display a trace that is not appearing in the graph box
How to remove a trace from the trace list

How to remove a trace from the trace list—ADL Shell

Deleting a trace from the Trace Preferences window (as opposed to making it invisible) frees up application memory and helps system performance.

Step 1
Click on any graph in the Graphics area.

Step 2
Click the button on the toolbar, or select Trace Preferences… from the Graph menu, or click the right mouse button in a graph box and select Trace Preferences…. The Trace Preferences dialog will appear.

Step 3
Select the trace to be removed by clicking on it.
Step 4

Click the right mouse button to display the Trace Preferences menu.

Step 5

Select Delete Traces to delete the sample from the trace list. The software will prompt you to confirm this.

Step 6

Press OK to exit the Trace Preferences window. The sample trace will be removed from all graphs.

See also:
How to remove a trace from a graph
How to display a trace that is not appearing in the graph box

How to replace a picture—ADL Shell

You can change a picture that you have added to a graph or Graphics area background using the following:

Step 1

Move the cursor to the picture and click the left mouse button to select it.

Hot Tip
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2*

Click the right mouse button.

Step 3

Select Edit Annotation from the Graph menu that appears.

Step 4

Click the Load button. The Windows Open dialog will appear.

Step 5

Browse through your directories to find the picture to replace the current bitmap. All pictures must be in the bitmap format and have the extension .BMP.

Step 6

Highlight the .BMP that you want to insert and click Open.
Step 7

The Edit Picture dialog will appear showing a preview of the bitmap. If the bitmap is the correct one click the OK button. The bitmap will be placed onto the graph.

Note
You should take care not to add a bitmap that is too big as it will obscure part or all of your graph.

If you need to, move the picture so that it is more appropriately placed on your graph.

* An alternative to Step 2 is to click on the Graph option in the menu bar.

See also:
How to add a picture to a graph
How to move a picture
How to delete a picture

How to resize graph boxes—ADL Shell

Step 1

Click on the graph that you want to resize so that the highlight box appears around it.

Step 2

Move the cursor to the edge of the highlight box and place it over one of the square marks (e.g. ■) in the box until it turns into a double headed arrow (←→).

Step 3

Click and hold the left mouse button and drag the box to the desired size.

Step 4

Release the left mouse button.

See also:
How to alter the look of your graphs

How to scale your graph manually—ADL Shell

Step 1

Click on the graph whose axes you want to change.

Step 2

Click the button on the toolbar, or select Axes Scales… from the Graph menu, or click the right mouse button in a graph box and select Axes Scales… The Axes Scales… dialog will appear.
Step 3

Type in your new ranges for the X and Y axes in the format ‘n-n’, or choose an option for each from the drop-down list.

Step 4

Press OK. The axes on the selected graph will change to reflect your chosen settings.

**Hot Tip**
Click on the Zoom Out option or button to resize the graph to its original settings.

See also:
- How to zoom a graph
- How to scale a graph automatically

**How to scale your graph automatically—ADL Shell**

Step 1

Click on the graph whose axes you want to change.

Step 2

Make sure that the trace that you wish to autoscale is the focused trace.

Step 3

Click the  button on the toolbar if you want the application to autoscale both X and Y axes. Alternatively click on the  button or the  button to autoscale only the X or Y axis. (You can also select these options from the Graph menu.)

The application will scale the focused trace according to the option selected.

**Hot Tip**
Click on the Zoom Out option or button to resize the graph to its original settings.

See also:
- How to zoom a graph
- How to scale your graph manually

**How to select the focused trace—ADL Shell**

To select the scan that you want to be the focused (red) trace, do the following:

- Click the left mouse button on the trace that you want to become the focused trace in a graph box.
• Click the left mouse button on the trace that you want to become the focused trace in the legend, to the right of the graph.

• Click on the trace that you want to become the focused trace in the Color column of the Trace Preferences window.

**Note**
Check the Show Legend check box in the Graph Preferences dialog to display the legend in each graph box.

See also:
- How to track the focused trace
- How to view a particular trace

**How to track the focused trace—ADL Shell**

To track the focused (red) trace in the graph:

**Step 1**

Click on the button on the toolbar, or select Cursor Mode from the Graph menu. The Cursor Modes dialog will appear.

**Step 2**

Select Track and click the OK button.

**Step 3**

Place the blue tracking cursor in the graph box you require.

**Step 4**

Move the cursor left and right across the graph box to trace the focused (red) scan. The cursor coordinates change (in the X, Y field at the bottom left hand corner of the Graphics area) as you move along the curve.

**Hot Tip**
If you wish to fix the cursor on a particular position on the curve, move the cursor to the required position. Press 'Alt' on the keyboard, then release the mouse button.

See also:
- How to select the focused trace
- How to view a particular trace
How to zoom a graph—ADL Shell

To do this, use the following procedure:

Step 1

Click on the button on the toolbar, or select Cursor Mode from the Graph menu. The Cursor Modes dialog will appear.

Step 2

Select Free and click the OK button.

Step 3

Click and hold down left mouse button in the highlighted graph box.

Step 4

Drag the mouse sideways and up or down. You will see a zoom box made up of a dotted line appear on the screen. (See picture below.)

Step 5

Release the left mouse button. The area defined by the zoom box will be redrawn to fill the entire graph box, and the scans within the box will be enlarged accordingly. The abscissa (X) and ordinate (Y) coordinates displayed below the graph will also be changed to reflect the positions of the corners of the zoom box.

You can repeat this process if you need to zoom in even further.

Hot Tip

To move the box on the screen, hold down the left and right mouse buttons together. You can then position the box anywhere in the graph box. Releasing the right mouse button will still allow you to resize the zoom box.

Step 5

Release the left mouse button. The area defined by the zoom box will be redrawn to fill the entire graph box, and the scans within the box will be enlarged accordingly. The abscissa (X) and ordinate (Y) coordinates displayed below the graph will also be changed to reflect the positions of the corners of the zoom box.

You can repeat this process if you need to zoom in even further.

Note

The system may limit the extent to which you can Zoom.

See also:

How to scale your graph manually
How to scale your graph automatically
## Advanced Reads Home Page

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Advanced Reads Application

Introduction

Use the Advanced Reads application to set up methods and read multiple samples in a single run.

The features of the application include:

- finding the mean of multiple readings of the sample solution
- finding the mean of multiple sample aliquots of the sample solution
- three different data modes—fluorescence, phosphorescence and chemi-/bio-luminescence
- the ability to use various accessories

To access the Advanced Reads Application, select the Windows Start button, point to Programs, Cary Eclipse and then select Advanced Reads.

Software controls

The application comprises the following:

**Instrument Status Readings**

The Instrument Status Readings, which display the Y and X values of the graph axes are displayed in the top left and right corners of the Advanced Reads Application window.

- **Y value**
  Displays the current Y value, usually intensity. This value is continually updated during a data collection.

- **X values**
  Displays the Excitation and Emission wavelengths.

**Start and Stop buttons**

The Start and Stop buttons at the top of the Advanced Reads Application window are used to start and stop instrument analyses. These buttons can also be accessed from the Commands menu.

Tell me more about the Start and Stop buttons

**Report area**

In the Advanced Reads Report area you can view the current report. The report may include the results of your data collection and the method parameters used to collect the results. You can select the information to include in your report in the Reports tab of the Setup dialog.

You can copy the generated report onto the Windows clipboard. To do this, highlight the report (or the relevant section) and press CTRL+C, Alternatively, you can select the Copy button on the Report Toolbar. You can then paste the report into a word processor such as Microsoft® Word.

Hot Tip

You can edit the Advanced Reads Report by pressing CTRL+R or selecting the Edit Report command from the Edit menu. Alternatively, you can select the Edit button on the Report Toolbar.

**Commands**

The Advanced Reads Application window contains the following commands, all of which can
also be accessed from the various menus in the application.

*Click on the buttons to view information about them:*

```
[Setup ...]  [Zero]  [Rread]  [Pre-scan ...]  
[Clear report]  [Print ...]  [G-Factor]
```

**Hot Tip**
By becoming familiar with these commands (Setup, Zero, Rread, etc.), you can remove the Command buttons from the Advanced Reads window. This will automatically enlarge the Report area, making it easier to view results. To do this, clear the Buttons item in the View menu.

**Menus**
The Advanced Reads Application contains the following menus:

File, Edit, View, Commands, Setup, Help

**See also:**
Advanced Reads home page
Keyboard function keys
Report Toolbar
Status Line
Validation of your Cary Eclipse for the Advanced Reads application

Three files are shipped with the Advanced Reads application to enable you to validate the software. The files are stored on your CD in a Validation subdirectory.

To validate your software, you should Open each file in turn and select the Recalculate button. The results calculated by the Cary Eclipse should match those given in the validation file listed below. These results were determined by exporting the data to a spreadsheet program and performing the same calculation as that used in the Advanced Reads application.

- Advanced Reads validate_re replicates.FBAB
- Advanced Reads validate_duplicates.FBAB
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Getting ready to read samples—Advanced Reads

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- Setup menu
- Cary page
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  - User collect example
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- Options page
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- Samples page
  - Polarizer
  - G-Factor
  - How to import sample labels
- Reports page
  - Clear Report
  - How to use the advanced reporting options
- Auto Store page
  - How to email the results

Setup—Advanced Reads

The Setup dialog enables you to set up your Cary Eclipse prior to a read. It also allows you to set up the Samples, Accessories, Report and Storage parameters associated with the read.

To access the Setup dialog select the Setup button from the Simple Reads Application window or the Setup command from the Setup menu.

- Cary
- Options
- Accessories
- Samples
- Reports
- Auto Store

Setup—Advanced Reads

Use the Setup menu to access the parameters for setting up a Advanced Read.
Use the **Cary** page to set up the instrument parameters associated with the data collection.

From the **Setup** menu, select **Cary** to access the **Cary** page. You can also access the page from the **Setup** dialog by selecting the **Cary** tab. To access the **Setup** dialog, select the **Setup** button from the Advanced Reads Application window.

The page comprises the following:

### Instrument setup

#### Data mode

**Fluorescence**

Fluorescence refers to the emission of light from an excited single state. The fluorescence signal is measured when the lamp flashes.

**Bio-/Chemi-luminescence**

Bio-/Chemi-luminescence refers to bio- or chemi-luminescence, where the emission is generated via a chemical or biochemical reaction. The lamp does not flash in Bio-
/Chemi-luminescence mode.

**Phosphorescence**
Phosphorescence refers to the emission of light from an excited triplet state. Phosphorescence is measured after the lamp has stopped flashing.

**Options**
Select **Options** to open the **Options** dialog where you can set parameters such as the Decay, Delay and Gate times for your sample.

The **Options** button is only visible if you have selected **Bio-/Chemi-luminescence** or **Phosphorescence** as the **Data Mode**.

**Wavelength setup**

**Multi-wavelength**
Select the **Multi-wavelength** option if you wish to compare the intensity of a sample at up to six different wavelengths. You can use it to compare the emission intensity of different excitation wavelengths and the excitation intensity of different emission wavelengths. Note that in Bio-/Chemi-luminescence mode, you can only compare emission wavelengths.

If you select **Multi-wavelength**, the following options will be available:

**Number of wavelengths**
Enter the number of wavelengths you wish to monitor. You can monitor up to six at one time. The number of wavelengths you select are represented as rows in the multi-wavelength table.

**Multiwavelength table**
Enter the excitation and emission wavelengths for each wavelength in the table. (Note that in Bio-/Chemi-luminescence mode, you can only enter the emission wavelengths. If you have selected more than two wavelengths to monitor, use the up and down arrows to the right of the table to access all of the wavelengths.

**Ex. Wavelength (nm)**
Set the wavelength of the excitation monochromator, which is the wavelength value at which you want to excite your sample. Click the arrow to the right of the field to quickly select the most recent wavelength entries.

Alternatively, to set the excitation monochromator to zero order (white light), from the **Commands** menu, point to **Zero Order** and select **Excitation**. A tick next to this option indicates that it is selected. Clear this option to return to monochromator light and use the value set in the **Ex. Wavelength** field.

**Em. Wavelength (nm)**
Set the wavelength of the emission monochromator, which is the wavelength value at which your sample emits. Click the arrow to the right of the field to quickly select the most recent wavelength entries.

Alternatively, to set the emission monochromator to zero order (white light), from the **Commands** menu, point to **Zero Order** and select **Emission**. A tick next to this option indicates that it is selected. Clear this option to return to monochromator light and use the value set in the **Em. Wavelength** field.
**User Collect**  Select User Collect to display the **User Collect** field. The **Ex. Wavelength** and **Em. Wavelength** fields will not be available. Note that this option is not available if you select **Multi-wavelength** mode.

Use the **User Collect** entry field to enter an ADL command which applies a mathematical expression to the intensity value collected. The most recent user collect entries will be saved in the drop down list to a maximum of 10.

The Syntax for ADL commands is:

```
Read (Ex. wavelength, Em. Wavelength)
```

**Hot Tip**

You can perform multi-ratio and multi-equation reads using the User Collect option. That is, you can calculate more than one ratio or equation in the one read, rather than performing separate reads for each ratio or equation.

For multi-ratios, use the format:

```
Read(355, 405)/Read(355, 480); Read(355, 500)/Read(355, 600)
```

An example of a multi-equation might be (without a line break):

```
2.1*Read(340, 510)/Read(405, 600); 7.5*Read(350,450)-4.5*Read(525, 610) +25.5*Read(600, 750)
```

The semi-colon (;) separates each ratio or equation. In addition to the results of each ratio or equation, the raw readings for each wavelength used will also be reported.

**Ex. Slit (nm)**  Set the slit width of the excitation monochromator. The **Ex. Slit** option is not available if you have selected **Bio-/Chemi-luminescence** as the **Data mode**.

*As a general rule:*

At a fixed emission slit width, increasing the excitation slit width will result in an increase in the fluorescence excitation intensity.

**Em. Slit (nm)**  Set the slit width of the emission monochromator

*As a general rule:*

At a fixed excitation slit width, a two-fold increase in the emission slit width will result in a four-fold increase in the emission intensity. Accordingly, a two-fold decrease in the emission slit width will result in a four-fold decrease in the emission intensity.

**Ave time (s)**  Set the amount of time, in seconds, for which data is averaged. To do this, enter the amount in the Ave. time field, or use the up/down arrows to the right of the field to increase or decrease the time. The averaged result is displayed in the report.

When setting the Ave Time, consider the following:
Status Display

Select this check box to display the Status Display window, where you can view the status of various parameters during the data collection.

About collection modes—Advanced Reads

In the Advanced Reads application, you can choose from three types of Data Modes: Fluorescence, Bio-/Chemi-luminescence or Phosphorescence. The Data Mode option is set on the Cary page of the Setup dialog.

Fluorescence

Fluorescence refers to the emission of light from an excited single state (lifetime $10^{-11} - 10^{-7}$ sec). The fluorescence signal is measured when the lamp flashes.

If you select Fluorescence mode, the Cary Eclipse will do the following:

- Generate emission with light source
- Measure the emission with the light source on

Bio-/Chemi-luminescence

- The longer the Ave Time, the longer the signal will be averaged
- A longer Ave Time will result in more precise results as the averaging of the signal reduces any contribution of background ‘noise’.

Averaging is calculated as follows:

For fluorescence:
the number of readings averaged are taken from consecutive flashes at 80 Hz where:
number of consecutive flashes = averaging time/0.0125.

For phosphorescence:
the intensity readings are taken from different decays, where:
number of decays = averaging time/gate time.

For bio-/chemi-luminescence:
a signal is collected every Gate time, and (averaging time/gate time) signals are averaged.

Understanding Average Time and Gate Time
Bio-/Chemi-luminescence refers to bio- or chemi-luminescence, where the emission is generated via a chemical or biochemical reaction. The lamp does not flash in Bio-/Chemi-luminescence mode.

- Generate emission with light source off
- Measure emission with light source off

Phosphorescence

Phosphorescence refers to the emission of light from an excited triplet state (lifetime $10^{-3}$ to $10^2$ sec). Phosphorescence is measured after the lamp has stopped flashing.

- Generate emission with light source
- Measure emission with light source off

See also:

Cary page

User Collect examples—Advanced Reads

You can extend the functionality of your Cary Eclipse instrument by using the built-in Read command.
This command enables you to change the instrument to a different wavelength and take a reading. You can combine this command with the mathematical operators: ‘+’, ‘–’, ‘*’ and ‘/’. This is illustrated in the examples below.

**Read(340, 400)–Read(340, 450)**

This reads the intensity at Excitation monochromator = 340, Emission monochromator = 400 and subtracts the intensity read at Excitation monochromator = 340 and Emission monochromator = 450. This reading is performed at each data point.

**Bio-/Chemi-luminescence/Phosphorescence Options—Advanced Reads**

Use the **Bio-/Chemi-luminescence/Phosphorescence Options** dialog to set up the parameters associated with these two measurement modes.

To access the dialog, select the **Options** button from the Cary page.

**Dialog controls**

The **Bio-/Chemi-luminescence/Phosphorescence Options** dialog comprises the following options:

- **Total decay time**
  - Enter, in seconds, the total decay time for your sample. The total decay time is the time that it takes for the phosphorescence signal to decay to dark levels (zero). The lamp will not flash again until the total decay time has elapsed.
  - The range is 0.001–1800.000 s.
  - **Note**
    - This option is not available in Bio-/Chemi-luminescence mode.

- **No. of flashes**
  - Enter the number of lamp flashes required, which is how many times the lamp flashes consecutively before the delay time is counted.
    - If you are measuring phosphorescence samples that have a relatively long decay time (that is, a number of seconds), you can setup the Cary Eclipse to pulse the lamp multiple times, as this will increase the intensity of the phosphorescent signal. For short lived phosphorescent samples, multiple flashes will not impact on the intensity of the signal, and therefore only one flash is required.
    - **How many flashes should I perform?**
    - **Note**
      - This option is not available in Bio-/Chemi-luminescence mode.
Delay time  Enter, in milliseconds, the delay time for your sample. The delay time is the time that elapses between the last flash and the beginning of data collection. The default value is 0.100 ms. Setting a delay time ensures that there is no lamp effects in the phosphorescence decay curve.
The range is 0.000–5000.000 ms.

Note
This option is not available in Bio-/Chemi-luminescence mode.

Gate time  Enter, in milliseconds, the gate time for your sample. The gate time is the overall reading time of the emission signal.
The range is from 10μs–10s.

Understanding Average Time and Gate Time for Phosphorescence scans
Understanding Gate Time for Bio-Chemi-luminescence scans

Example of the Bio-/Chemi-luminescence/Phosphorescence Options

See also:
Cary page

Understanding Gate Time for Bio-/Chemi-luminescence Scans—Advanced Reads

Gate Time is set on Options page, which is accessible from the Cary page.
The following diagram highlights the **Gate Time** setting.

The **Gate Time** is set to 10 and the **Intensity** is equal to $I_T$.

![Diagram showing the relationship between Intensity and Gate Time](image)

See also:
- Understanding Average Time and Gate Time for Phosphorescence scans
- Advanced Reads Application
- Cary page
- Options dialog
- User Collect example

### Understanding Average Time and Gate Time for Phosphorescence Scans—Advanced Reads

For Phosphorescence scans, there is a relationship between Average Time (the time for which data is averaged) and Gate Time (the overall reading time of the emission signal).

Averaging is calculated as follows:

The intensity readings are taken from different decays, where:

**Number of Decays** = **Averaging Time/Gate Time**

**Note:**

**Gate Time** is not used if you select **Fluorescence** as the **Data Mode**.

**Average Time** is set on the **Cary** page of the **Setup** dialog. **Gate Time** is set on **Options** page, which is accessible from the **Cary** page.

The following diagrams highlight the relationship between **Average Time** and **Gate Time**.

**Example 1**
When the Average Time and Gate Time are set to the same value:

<table>
<thead>
<tr>
<th>Average Time/Gate Time</th>
<th>1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intensity</td>
<td>1 (Area $I_1$)</td>
</tr>
</tbody>
</table>

where:

- Total Decay Time: 20 ms
- Delay Time: 10 ms
- Average Time: 5 ms
- Gate Time: 5 ms

**Example 2**

When the Average Time is set to five times the Gate Time:

<table>
<thead>
<tr>
<th>Average Time/Gate Time</th>
<th>5</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intensity</td>
<td>$A_1 + A_2 + A_3 + A_4 + A_5$</td>
</tr>
</tbody>
</table>

\[ \frac{\text{Area } I_1}{5} \]
where:

Total Decay Time 20 ms
Delay Time 10 ms
Average Time 5 ms
Gate Time 25 ms

See also:
How many flashes should I perform?—Advanced Reads

The **Number of Flashes** option is set in the **Bio-/Chemi-luminescence/Phosphorescence Options** dialog. To access the dialog, select the **Options** button from the **Cary page**.

**Note**
The **Options** button is only visible on the **Cary page** if you have selected **Bio-/Chemi-luminescence** or **Phosphorescence** as the **Data mode**.

If you are measuring phosphorescence samples that have a relatively long decay time (that is, a number of seconds), you can setup the Cary Eclipse to pulse the lamp multiple times, as this will increase the intensity of the phosphorescent signal (refer to Example 1, below). For short lived phosphorescent samples, multiple flashes will not impact on the intensity of the signal, and therefore only one flash is required (refer to Example 2, below).

**Example 1**
The above example shows that the sample does not decay before the instrument lamp flashes again. Therefore setting multiple flashes will result in an increase in the intensity of the signal.
**Example 2**
The above example shows that the sample decays in less time than the time it takes the instrument to flash the lamp again. Therefore, setting more than 1 flash, in the **No. of Flashes** field will not increase the intensity of the signal.

See also:
[Bio-Chemi-luminescence/Phosphorescence Options](#)

**Options—Advanced Reads**

Use the Options page to set up your filters and set the detector voltage.

From the **Setup** menu, select **Options** to access the **Options** page. You can also access the page from the **Setup** dialog by selecting the **Options** tab. To access the **Setup** dialog, select the **Setup** button from the Advanced Reads Application window.

<table>
<thead>
<tr>
<th><strong>Excitation filter</strong></th>
<th>Set the excitation filter for the Advanced Reads measurement. To do this, select the down arrow to the right of the field and select an option from the list.</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Auto</strong></td>
<td>If you select the <strong>Auto</strong> option, the filter wheel will be automatically moved to the appropriate position for the selected excitation wavelength. It is recommended that you set the filter to <strong>Auto</strong> (which is the default setting).</td>
</tr>
<tr>
<td><strong>Open</strong></td>
<td>If you select <strong>Open</strong> no filter is used.</td>
</tr>
<tr>
<td><strong>Closed</strong></td>
<td>If you select <strong>Closed</strong>, a blank filter is used to prevent light from getting through.</td>
</tr>
<tr>
<td><strong>250–395, 335–620, 550–1110, 695–1110</strong></td>
<td>If you are looking at a wavelength range that is within one of these filter ranges, you can select your filter manually. Otherwise it is recommended that you set the filter to <strong>Auto</strong>.</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th><strong>Emission filter</strong></th>
<th>Set the emission filter for the Advanced Reads measurement. To do this, select the down arrow to the right of the field and select an option from the list.</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Auto</strong></td>
<td>If you select the <strong>Auto</strong> option, the filter wheel will be automatically moved to the appropriate position for the selected emission wavelength. It is recommended that you set the filter to <strong>Auto</strong> (which is the default setting).</td>
</tr>
<tr>
<td><strong>Open</strong></td>
<td>If you select <strong>Open</strong> no filter is used.</td>
</tr>
</tbody>
</table>
**Closed**
If you select Closed, a blank filter is used to prevent light from getting through.

250–395, 295–1110, 360–1110, 430–1110, 550–1110
If you are looking at a wavelength range that is within one of these filter ranges, you can select your filter manually. Otherwise it is recommended that you set the filter to Auto.

**3%T Attenuator**
Select 3%T Attenuator to decrease the fluorescence intensity and thus the amount of light going to the emission PMT.

Set the EHT or sample detector PMT. To do this, select one of the following:

- **Low**
  Sets the PMT Detector voltage to a pre-defined value of 400 v. Typically you would select this setting if you are reading highly emitting samples.

- **Medium**
  Sets the PMT Detector voltage to a pre-defined value of 600 v. Typically you would select this setting if you are reading moderately emitting samples.

- **High**
  Sets the PMT Detector voltage to a pre-defined value of 800 v. Typically you would select this setting if you are reading low emitting samples.

- **Manual**
  Enables you to enter any PMT voltage in the adjacent Volts field.

**Volts**
Enter the PMT voltage in the adjacent field. Note that this option is only available if you have selected Manual as the PMT Detector voltage.

**Tip**
Increasing or decreasing the PMT Detector Voltage by 10% will double or halve the fluorescence intensity observed.

**Status Display**
Select this check box to display the Status Display window. This window allows you to view the status of various parameters during the data collection.

See also:
- [Setup Status Display window](#)

---

**Accessories—Advanced Reads**

Use the Accessories page to set up the accessory parameters associated with the data collection.

From the Setup menu, select Accessories to access the Accessories page. You can also access the page from the Setup dialog by selecting the Accessories tab. To access the Setup dialog, select the Setup button from the Advanced Reads Application window.
The Accessories page will vary, depending upon what accessory button you have selected from the following:

- Multicell/Temperature Control
- Wellplate
- Polarizer

See also: Cary

**Multicell/Temperature accessories—Advanced Reads**

The Multicell/Temperature accessory page is displayed when you select the Multicell/Temperature tab on the Accessories page.

The page comprises the following:

**Multicell Holder**

Select this option to use the Multicell Holder with the Cary Eclipse.

**Note**

If you select the Multicell holder accessory, you will not be able to select the Wellplate reader accessory.

**Multi-zero**

If you select Multi-zero, a Multi-zero will be performed when the Zero button is selected in the Advanced Reads Application window. A reading will be taken at the set Averaging Time at the specified wavelengths of one or both monochromators and at every selected cell position. These readings will be offset to zero and subsequent readings will have the corresponding offset value subtracted from each cell.

**Reset**

Select Reset to drive the Multicell Holder to cell position 1, which is its fully retracted or zero position.

**Go to cell…**

Select Go to cell to open the Multi Cell Changer dialog. You can use this dialog to manually move the Multicell Holder accessory to a new cell. To do this, simply select the corresponding button of the cell you require in the Multi Cell Changer dialog. The Multicell Holder will move the selected cell to the measurement position in the sample compartment.

The Multi Cell Changer dialog is ‘modeless’ which means that it can remain open even once you have cleared the Setup dialog. This enables you to use the Rapid Read command to perform a reading at the cell specified in the Cell Changer.

If you move to a new cell after you have performed a Multi-zero scan, the Cary Eclipse will display the intensity reading corrected by the determined zero-offset amount for that cell.
Temperature Control

Select **Temperature Control** to specify the Temperature parameters for your analysis.

**Note**
You can use this option to check the intensity of a cell or perform a read at a particular cell.

The Temperature Control option does not control temperature accessories that are not listed in the Device field. For example, if you have selected the **Temperature Control** option and set the Device to **None** because you have a different temperature control accessory, the Cary Eclipse software will not control that accessory. However, it will monitor the temperature and prompt the software to start when the accessory reaches the temperature listed in the **Temperature** field.

**Device**
Select the device that will be used to control the water temperature.

The options available will depend upon whether or not you have selected the **Multicell holder** option.

If you *have* selected the Multicell holder, the following **Device** options will be available for selection:

- Temperature controller
- PCB 150 water bath
- None

If you *have not* selected the Multicell holder, the following **Device** options will be available for selection:

- Single cell peltier
- PCB 150 Water bath
- None

**None**
Select this option if you are using a Temperature Control device not listed in this field (for example, a third party water bath). Note that the Cary Eclipse software will not control the accessory. The software will simply monitor the temperature and prompt the software to start when the accessory reaches the temperature listed in the **Temperature** field.

**PCB 150 water bath**
Select this option if you are using the PCB 150 water bath.

**Single cell peltier**
Select this option if you are using the Single cell peltier.

**Temperature controller**
Select this option if you are using the Temperature controller.

**Temperature**
Set the temperature at which the analysis will be performed. The default value is 20.00° C.

When you select OK to close the Setup dialog, all buttons in the Advanced Reads Application window will be disabled until the temperature recorded by the Monitor device is within ± 0.5°C of the temperature range.
If Zero or Start is selected, and the temperature is not within ± 0.5°C of the temperature range, then the system will wait for the temperature to fall within the temperature range before continuing with the measurement. Whilst waiting for the temperature, there will be a display on the status line at the bottom of the Advanced Reads Application window giving the current temperature and the elapsed time waiting to reach the set temperature.

**Port**
Select from COM1, COM2, COM3 or COM4.
This option is only available if you have selected PCB 150 Water bath as the Device.

**Monitor**
Select which device will be used to monitor the reaction temperature. The options in this field will change depending on the Device selected.

<table>
<thead>
<tr>
<th>Device selected</th>
<th>Monitor options available</th>
</tr>
</thead>
<tbody>
<tr>
<td>None</td>
<td>Probe 1, Probe 2, Probe 3, Probe 4</td>
</tr>
<tr>
<td>PCB 150 water bath</td>
<td>PCB 150, Probe 1, Probe 2, Probe 3, Probe 4</td>
</tr>
<tr>
<td>Single cell peltier</td>
<td>Block, Probe 1, Probe 2, Probe 3, Probe 4</td>
</tr>
<tr>
<td>Temperature controller</td>
<td>Block, Probe 1, Probe 2, Probe 3, Probe 4</td>
</tr>
</tbody>
</table>

**Temperature Display**

Select this option to view Temperature Display information in the Status Display window. The Temperature Display option is only available if you have selected the Temperature Control option.

**Block/PCB 150**
Select this option to view the temperature of the Block or the PCB 150 in the Status Display window. This option is not available if you have selected None as the Device option.

Note that the PCB 150 option is only displayed if you have selected the PCB 150 Water Bath as the Device option without the Multicell holder.

**Probes**
Select one or more of these options to view the temperature of the Probes in the Status Display window.

**Status Display**

Select this check box to display the Status Display window. Use this window to view the status of various parameters during the data collection.

See also:
- PCB 150
- Multi Cell Changer
- Advanced Reads Application
- Cary
- Accessories
- Status Display

**Multi Cell Changer—Advanced Reads**

Use the Multi Cell Changer dialog to manually move the Multicell Holder accessory to a new cell. To
access the **Multi Cell Changer** dialog, select the **Go to cell** button from the **Multicell/Temperature Accessories** page.

The dialog comprises the following:

**Multicell changer**  To manually move the Multicell Holder accessory to a new cell, simply select the corresponding button of the cell you require in the Multi Cell Changer dialog. The Multicell Holder will move the selected cell to the measurement position in the sample compartment.

The **Multi Cell Changer** dialog is ‘modeless’ which means that it can remain open even once you have cleared the **Setup** dialog. This enables you to use the **Rapid Read** command to perform a reading at the cell specified in the cell changer.

If you move to a new cell after you have performed a Multi-zero scan, the Cary Eclipse will display the intensity reading corrected by the determined zero-offset amount for that cell.

**Note**
You can use this option to check the intensity of a cell or perform a scan at a particular cell.

See also:
- **Multicell/Temperature Accessories**
- **Rapid Read**

**Wellplate accessory—Advanced Reads**

The **Wellplate** accessory page is displayed when you select the **Wellplate** tab on the **Accessories** page.

The page comprises the following:

**Wellplate reader**

Select this option to use the wellplate reader with the Cary Eclipse.

**Q. Why is the wellplate option unavailable?**

**A.** The multicell holder, temperature control and/or polarization accessory has been selected. As you cannot place any of these accessories and the wellplate reader in the Cary Eclipse sample compartment at the same time, you must clear other accessory selections before selecting the wellplate reader.

**Plate format**  Select the type of wellplate to be used. The default options are 6 wells, 12 wells, 24 wells, 48 wells, 96 wells, 192 wells or 384 wells. The bitmap representing the wellplate will change according to the number of wells chosen.

**Auto-zero**  Select this option to perform an auto-zero when the **Zero** button is selected from the Advanced Reads Application window. A zero reading will be taken at the selected well position using the current method setup. To select the well position to be zeroed, use the **Select Well** option.
These readings will be offset to zero and subsequent readings will have this single value subtracted from each well in the sequence. The reading is taken only once, stored, and subtracted from the ratioed intensity reading.

Select well
Choose **Select well** to open the **Go To Well** dialog where you can select the well position to be zeroed. This option is only available if you have selected **Auto-zero**.

Well
Displays the well position of the well selected for auto-zeroing. This field is not editable.

Sample names
Select **Sample Names** to open the **Wellplate Sample List** where you can enter the sample names.

Go to well
Select this option to open the **Go To Well** dialog.

Well position
Displays the well position that the mouse cursor is pointing to. This field is not editable.

Reset
Select **Reset** to move the wellplate reader to the first well position defined by the read sequence.

Park
Select **Park** to move the wellplate reader to the **Park** position. The wellplate reader accessory is moved clear of the sample compartment to enable you to easily access all wells.

Wellplate reader diagram
The wellplate reader diagram represents the number of wells present for the chosen wellplate configuration.

The labels for the rows will be 1 .. 12 and for the columns A .. H (for a 96 wellplate). Similar labels will occur for different wellplates according. The sequence for numbering the read order will be from A1 to A12, B12 - B1, C1 - C12…etc for a 96 wellplate. A similar sequence will be for other plates.

To select or clear a single well, simply click or right-click on it.

To select or clear a block of wells, hold down the cursor and drag around the block required. All wells within the area will be selected or cleared.

**Status Display**

Select this check box to display the **Status Display** window. This window allows you to view the status of various parameters during the data collection.

**See also:**
- Accessories page
- Go To Well dialog
- Wellplate Sample List dialog
- About the Wellplate Reader
- Status Display

**Go To Well—Advanced Reads**

You can use the **Go To Well** dialog to manually move the Wellplate Reader accessory to a new well. This enables you to check the intensity of a well or perform a read at a particular well.

The **Go To Well** page is accessible from the **Wellplate Accessories** page by selecting the **Select well** or
Go To Well button.

The Go To Well dialog is ‘modeless’ which means that it can remain open even once you have cleared the Setup dialog. This enables you to use the Rapid Read command to perform a reading at the selected well.

If you move to a new well after you have performed a Multi-zero, the Cary Eclipse will display the intensity reading corrected by the determined zero-offset amount for that well.

The page comprises the following:

**Wellplate reader diagram**

The Wellplate Reader diagram represents the number of wells present for the chosen wellplate configuration.

The labels for the rows will be 1 .. 12 and for the columns A .. H (for a 96 wellplate). Similar labels will occur for different wellplates according. The sequence for numbering the read order will be from A1–A12, B12–B1, C1–C12, etc for a 96 wellplate. A similar sequence will be for other plates.

To move the Wellplate Reader accessory to a new well, simply click on the corresponding well on this diagram. The Wellplate Reader will move the selected well to the measurement position in the sample compartment.

To select or clear a single well, simply click or right-click on it.

To select or clear a block of wells, hold down the cursor and drag around the block required. All wells within the area will be selected or cleared.

**X, Y, Well**

Displays the Well Position that the mouse cursor is pointing to. This field is not editable.

**Reset**

Select Reset to move the Wellplate Reader to the first well position, which is well A1.

**Park**

Select Park to move the Wellplate Reader to the Park position. When the accessory is in the park position you have full access to the Wellplate Reader platform. This is useful for inserting or removing wellplates from the accessory.

See also:
- Wellplate Accessories page
- About the Wellplate Reader

**Wellplate Sample List—Advanced Reads**

Use the Wellplate Sample List dialog to enter or modify the sample names of each well in the wellplate reader.

The Wellplate Sample List is accessible from the Wellplate Accessories page by selecting the Sample Names button.

The page comprises the following:

**No. of**

Displays the total number of samples that have been selected from the Wellplate Reader.
How to import sample names into the Wellplate sample list—Advanced Reads

If you want to import sample names into the Wellplate sample list in the Advanced Reads application, then you simply need to enter your data in a *.txt file and then import this file into the Wellplate sample list dialog. The data should be entered in the following manner:

<table>
<thead>
<tr>
<th>Sample Name</th>
<th>Becomes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Samp A</td>
<td>Samp A</td>
</tr>
<tr>
<td>Sample B</td>
<td>Sample B1</td>
</tr>
<tr>
<td>Sample C</td>
<td>Sample B2</td>
</tr>
<tr>
<td>Sample D</td>
<td>Sample B3</td>
</tr>
<tr>
<td>Sample E</td>
<td>Sample B4</td>
</tr>
</tbody>
</table>

To modify the name of a sample, highlight the sample name and type in a new name. You can enter up to 20 characters for each name.

**Well**
Displays the position of the well corresponding to the Sample Name. This field is not editable.

**Import names**
Select Import Names to display the Open dialog which you can use to select a text file to import into the Sample Names column.

**Increment**
Select Increment to copy and increment the sample names in the Sample Name list. This will save you from entering each sample name individually when the names consist of a descriptor followed by a number.

To use the increment option, highlight the name that you want to increment and select Increment. The Cary Eclipse will increment the names of all the files proceeding the first highlighted name.

**Example:**
In this example, Sample B1 is highlighted when the Increment button is selected.

See also:
- Wellplate Accessories page
- About the Wellplate Reader

How to import sample names into the Wellplate sample list—Advanced Reads

If you want to import sample names into the Wellplate sample list in the Advanced Reads application, then you simply need to enter your data in a *.txt file and then import this file into the Wellplate sample list dialog. The data should be entered in the following manner:
Each sample name and Well position in the imported file must be on a separate line. Enter the Well position on the line immediately after its corresponding Sample Name. Note that any tabs in the file are ignored.

To illustrate this procedure, the following Sample name file (shown on the left) will be used. The picture on the right shows the wells that will be selected for inclusion in the Wellplate sample list. Note that you do not need to include the well positions, as they are entered automatically (in this case Sample_name1–Sample_name6 will correspond to wells A1–A6 and Sample_name7–Sample_name12 will correspond to wells B1–B6).

<table>
<thead>
<tr>
<th>[leave blank]</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sample_name1</td>
</tr>
<tr>
<td>Sample_name2</td>
</tr>
<tr>
<td>Sample_name3</td>
</tr>
<tr>
<td>Sample_name4</td>
</tr>
<tr>
<td>Sample_name5</td>
</tr>
<tr>
<td>Sample_name6</td>
</tr>
<tr>
<td>Sample_name7</td>
</tr>
<tr>
<td>Sample_name8</td>
</tr>
<tr>
<td>Sample_name9</td>
</tr>
<tr>
<td>Sample_name10</td>
</tr>
<tr>
<td>Sample_name11</td>
</tr>
<tr>
<td>Sample_name12</td>
</tr>
</tbody>
</table>

Figure 1
An example of a Sample name text file.

Figure 2
An example of the wells selected on the Wellplate Accessories page.

To import sample names:

Step 1

Enter your data into a text editor (e.g. Notepad) and save it as a text (*.TXT) file. Refer to Figure 1 for an example of the correct format for entering the text.

Note:
The first line of the text file you create is not included when you import the sample names list. Therefore you should leave this line blank.

Step 2

From the Advanced Reads Application window, select the Setup button to display the Setup dialog.

Step 3

Select the Accessories tab to display the Accessories page. Select the Wellplate tab and from the Wellplate page.

Step 5

From the Wellplate diagram, select the wells to be included in your analysis. (Refer to Figure 2.) These
wells will be included in the **Wellplate sample list**.

Step 6

Select the **Sample names** button to display the **Wellplate sample list**.

Step 7

Select the **Import Names** button. The **Open** dialog is displayed.

Step 8

Browse for you text file and select **Open** to open the file. The Sample Names and their corresponding Well position will appear in the **Sample Name** table.

See also:

- [Wellplate sample list](#)
- [Wellplate Accessories page](#)
- [About the Wellplate Reader](#)

**Polarizer accessory—Advanced Reads**

The Polarizer accessory page is displayed when you select the **Polarizer** tab on the **Accessories** page.

The page comprises the following:

**Polarizer**

Select this option to use the Polarizer with the Cary Eclipse.

Q. Why is the **Polarization** option unavailable?

A. The Wellplate accessory has been selected. As you cannot place both the Wellplate accessory and the Polarization accessory in the Cary Eclipse sample compartment at the same time, you can only select one of these accessories.

**Measurement**

Select from the following measurement types: **Polarization**, **Anisotropy** or **Other**.

**G Factor**

The G Factor is a value that reflects the polarization bias of the emission monochromator. It is dependant upon the emission wavelength. If you do not select the G Factor option, then a G-factor value must be collected. If you do select the G Factor option then the value entered is used at the emission wavelength in subsequent polarization or anisotropy calculations.

The G Factor option is not available if you select **Other** as the Measurement type.

**Determining the G-factor**

A check to see whether there is a valid G Factor that corresponds to the emission wavelength is performed when you select OK to exit the Setup dialog. If the G Factor is not valid, the following prompt is displayed:

‘The current G-factor is not valid at the emission wavelength. Press the G Factor button
Status Display

Select this check box to display the Status Display window. This window allows you to view the status of various parameters during the data collection.

G-Factor—Advanced Reads

Select G-Factor to calculate the G Factor. You will be prompted to move your polarizers (Em and Ex) and perform the \( I_{HV} \) and \( I_{HH} \) measurements. At the end of the sequence the system will calculate the subsequent polarization measurements which will be performed when you select the Start button.

The G-Factor command is only available if you have selected the Polarizer accessory on the Accessories page. Once this option is selected, you can then select the G-Factor button from the Advanced Reads Application window or the G-Factor command from Commands menu.

Samples—Advanced Reads

Use the Samples page to specify a list of sample names that will be used during your analysis.

From the Setup menu, select Samples to access the Samples page. You can also access the page from the Setup dialog by selecting the Samples tab. To access the Setup dialog, select the Setup button from the Advanced Reads Application window.

The page comprises the following:

<table>
<thead>
<tr>
<th>No. of samples</th>
<th>Enter the number of samples to be analyzed in this field. The Sample Names list below will expand to reflect your entry.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Replicates</td>
<td>This sets the number of replicate readings to be taken for each sample.</td>
</tr>
</tbody>
</table>

A single solution can lack homogeneity that can significantly alter the results of an analysis. For these reasons, common laboratory practice is to take multiple readings of the same sample. A maximum of five replicate readings can be taken for each sample.

Q. Why is the Replicates numerical data entry field grayed out?
A. Select the Replicates option to activate this field. You cannot have Replicates
Sample averaging

This sets the number of readings to be taken from different aliquots of the same sample/standard solution. The readings are then averaged. The results of every reading, as well as the final averaged result, are included in the report.

Using sample averaging helps minimize solution make-up errors. For example, readings from multiple solutions of samples prepared in supposedly identical manners may yield different results. Sample averaging minimizes this source of error.

For duplicates, enter ‘2’ in the numerical data entry field. For triplicates, enter ‘3’.

Q. Why is the Sample Averaging numerical data entry field grayed out?

A. Select the Sample Averaging option to activate this field. You cannot have Sample Averaging and Replicates selected at the same time.

Sample name (list)
Enter your sample names in this field. To do this, highlight the Sample name and type in the new name. You can enter up to 20 characters for each name.

IncrementSelect Increment to copy and increment the sample names in the Sample names list. This will save you from entering each sample name individually when the names consist of a descriptor followed by a number.

To use the increment option, highlight the name that you want to increment and select Increment. The Cary Eclipse will increment the names of all the files proceeding the first highlighted name.

Example:
In this example, Sample Name ‘B’ is highlighted when the Increment button is selected.

<table>
<thead>
<tr>
<th>Sample Name</th>
<th>Becomes</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>A</td>
</tr>
<tr>
<td>B</td>
<td>B1</td>
</tr>
<tr>
<td>C</td>
<td>B2</td>
</tr>
<tr>
<td>D</td>
<td>B3</td>
</tr>
<tr>
<td>E</td>
<td>B4</td>
</tr>
</tbody>
</table>

Import Names
Click this button to display the Windows Open dialog which you can use to import an ASCII text file of sample names.

Note
Each sample name in the imported file must be on a separate line; any tabs in the file are ignored. If you want to include duplicates or triplicates, then they must be on a separate line immediately after the initial sample name and called ‘Dup’ and ‘trip’ (maintain the case as shown). Place the sample number immediately after the Dup or trip identifier.

Status Display
Select this check box to display the Status Display window. Use this window to view the status of various parameters during the data collection.

See also:
How to import sample labels
How to import sample labels—Advanced Reads

If you want to import sample data into the Advanced Reads application, then you simply need to enter your data in a *.txt file and then open this in the Advanced Reads application. The data should be entered in the following manner:

Each sample name in the imported file must be on a separate line, and any tabs in the file are ignored. If you want to include duplicates or triplicates, then they must be on a separate line immediately after the initial sample name and called ‘Dup’ and ‘trip’ (maintain the case as shown). Place the sample number immediately after the Dup or trip identifier.

An example file is given below.

```
Sample_name1, Dup1, trip1,
Sample_name2, Dup2, trip2,
Sample_name3, Dup3, trip3,
Sample_name4, Dup4, trip4,
Sample_name5, Dup5, trip5,
```

Step 1

Enter your data into a text editor and save it as a text file.

Step 2

Open the Advanced Reads Application window.

Step 3

From the Setup menu, select Cary. The Cary page is displayed.

Step 4

On the Cary page, set the Replicates field to that the number of replicates specified in your sample file.

Step 5
Click on the **Samples** tab. The **Samples** page is displayed.

Step 6

Click **Import Names**. The **Open** dialog is displayed.

Step 7

Search for and highlight your *.txt file. Select **Open** to open your *.txt file. The sample names will appear in the **Samples Name** list.

See also:
- [Cary page](#)
- [Samples page](#)

**Reports—Advanced Reads**

Use the **Reports** page to set up the report parameters associated with your data collection.

From the **Setup** menu, select **Reports** to access the **Reports** page. You can also access the page from the **Setup** dialog by selecting the **Reports** tab. To access the **Setup** dialog, select the **Setup** button from the Advanced Reads Application window.

The page comprises the following:

**Operator**

- **Name** Enter the instrument operator name in this text entry field. Use the arrow to the right of this field to view and select one of the last ten entries in this field.

  **Note** In GLP mode the system will automatically enter the operator’s name and this field will not be editable.

- **Comment** Enter operator details and comments in this text entry field.

**Options**

This group enables you to select the information to include in your report.

- **Auto Print** Select **Auto Print** to print a report automatically during the data collection. If **Auto Print** is selected, then the system will send the report information to the specified printer(s) as well as the Report area. However if **Auto Print** is not selected, the report will only be sent to the Report area.

- **Parameters** Select this option to include the method setup parameters within your report.

- **Company Logo** Check this option to include your company logo in the report.

  A company logo will only be included if you have added one on the **Company** page of the System Information Application.
**Hot Tip**
If you have not included a Company logo, select the Header/Footer button to open the System Information Application and use the Add Logo option to add a logo.

**Header/Footer**
Select the Header/Footer button to open the System Information application where you can enter the Header and Footer information to be included in your report. You can also enter any additional comments, and add a logo to your report. Once you have made all your changes, select OK to exit the System Information application and return to the Reports page of the Advanced Reads application.

**Status Display**
Select this check box to display the **Status Display** window, where you can view the status of your instrument during the data collection.

**Hot Tip**
You can copy a picture, spreadsheet file or document from another application (for example, Microsoft Word or Microsoft Excel) into the report area of the Lifetimes application. To edit these objects, simply double click on them to open the associated application in which they were created.

**See also:**
[Status Display](#)

**Clear Report—Advanced Reads**
Select **Clear Report** to remove the contents of the Report area. If you have saved the report, selecting this option does not delete the contents of the report, the information is simply removed from the Report Area.

You can select the **Clear Report** button from the Advanced Reads Application window or the **Clear Report** command from Commands menu.

**See also:**
[Advanced Reads Application](#)
[Commands menu](#)

**How to use advanced reporting options—Advanced Reads**
This outlines ways to fully customize your reports. Options available include:

- Including header and footer information
- Including a logo
- Editing the text in the report
- Viewing the report on screen before printing
- Cutting and pasting to a word processor
Including header and footer information

You can have text appearing at the top and bottom of every page in your report. To specify what the text should be, do the following:

Step 1

On the Windows Taskbar, click the Start button, point to Programs, Cary Eclipse and then select System Information. The System Information application appears with the Company page displayed.

Step 2

Enter the header and footer information in the Header and Footer fields.

Step 3

Click OK to implement your changes.

Including a logo

You can include a logo on the first page of your report. To specify what the logo should be, do the following:

Step 1

On the Windows Taskbar, click the Start button, point to Programs, Cary Eclipse and then select System Information. The System Information application appears with the Company page displayed.

Step 2

Enter a logo using the Add Logo button. This logo will always be included on the front page of reports.

Step 3

Click OK to implement your changes.

Q. Why isn't the logo I selected included in my report?
A. If you are running the Cary Eclipse software under Windows NT you need to be logged on as an Administrator in order to specify the logo in the System Information application.

Editing the text in the report

You can edit any of the text in the Report area. Just select Edit Report from the Edit menu. You can then add, delete and change text in the Report area.

Q. The Edit Report option is not available, why not?
A. You do not have reporting editing privileges set in the GLP Administration application. The Administrator of the system will have to change your privileges before you can edit reports.
Viewing the report on the screen before printing

You can view a report before printing it. To do this, from the File menu select Print Preview. You can then ensure that the report format is correct before you print it.

Hot Tip
Use the arrow keys at the bottom of the Print Preview screen to change pages.

Cutting and pasting to a word processor

To have complete control over how your report looks you may wish to copy it to a word processor program, such as Microsoft Word. To do this:

Step 1
Open a report.

How to open a previously stored report

Step 2
From the Edit menu, choose Edit Report.

Step 3
Click in the Report area and highlight the text you want to export to your word processor.

Step 4
Right-click on the Report area and select Copy from the menu.

Step 5
Start up your word processor and use the Paste function (usually under the Edit menu) to place the text into a document.

Auto Store—Advanced Reads

Use the Auto Store page to specify if and when data will be stored, and in what format. You can also use this page to set up the system to automatically send an email at the end of a read.

From the Setup menu, select Auto store to access the Auto store page. You can also access the page from the Setup dialog by selecting the Auto store tab. To access the Setup dialog, select the Setup button from the Advanced Reads Application window.

The page comprises the following:
Storage

Off
Select this if you do not wish to be automatically prompted to save collected data.

Note
If you wish to store your unsaved data manually, choose Save Data As… from the File Menu to save your data after the run is complete. You can save your data as a Batch or Data file.

On; Prompt
At Start
Select this to display the Windows Save As dialog at the start of the collect where you can enter the Batch filename for your data.

On; Prompt
At End
Select this to display the Windows Save As dialog at the end of a collect where you can enter the Batch filename for your data.

Email

Select the Email check box and enter the Email details to send an email to a person once the read is completed. This useful facility can be used to alert you to the completion of the read, particularly if you are working away from the laboratory where the Cary Eclipse instrument is. You can also attach the collected data to the email.

Note
You must set up the email Host and Port options on the Email options page of the System Information application in order for the email facility to work correctly.

To
Enter the recipient's email address. You can enter more than one recipient in this field.

From
Enter your email address (or the email address associated with the computer on which the Cary Eclipse software is installed).

Subject
Enter the subject of your email (or leave this field blank if you do not wish to include a subject).

Cc
Enter the email address of any recipient you want to copy the email to.

Message
Enter your message.

Attached…
Use the Attached button include a file attachment in your email. To do this, select Attached and browse your computer to select a file, using the Windows® Open dialog that appears.

Attach current data
Select this option to attach the data that has been collected during the run to the email.

Status display

Select this check box to display the Status Display window, where you can view the status of your instrument during the data collection.

See also:
Advanced Reads Application
Save As
Status Display
How to email your results—Advanced Reads

You may wish to send an email to a person once the read is completed. This useful facility can be used to alert you to the completion of the read, particularly if you are working away from the laboratory where the Cary Eclipse instrument is.

To do this:

Step 1

Ensure that you have set up the Host and Port options on the Email options page of the System Information application in order for the email facility to work correctly.

Step 2

From the Setup menu, select Auto Store. The Auto Store page is displayed.

Step 3

Select the Email check box to enable the email options.

Step 4

In the To field, enter the recipient's email address (for example, anne.person@varianinc.com). To enter multiple recipients, simply type a comma ',' between each email address.

Step 5

In the From field, enter the email address of the computer on which the Cary Eclipse software is installed. This may be your email address, or a communal email address, if the Cary Eclipse software is installed on a computer that has multiple users.

Step 6

Enter information in the Subject and Message fields as required.

Step 7

Select Attach current data. The data that has been collected during the run will be attached to the email.

Hot Tip

You can attach other documents to your email. To do this, select the Attached button and browse your computer to select a file, using the Windows® Open dialog that is displayed.

Step 8

Select OK to clear the Auto Store dialog. Your email will be sent once the run has been completed.
See also:

Auto Store
Performing scans—Concentration

- Starting a read
- Present Standard dialog
- Present Sample dialog
- Cell Loading Guide
- Start/Stop buttons
- Pre-scan dialog
- Reread
- Rapid Result dialog
- Zero
- Go To dialog
- Commands menu
- How to perform a calibration
- How to perform a calibration and manually measure concentrations
- How to perform a calibration and measure concentrations using the Multicell Holder

Information about starting a run—Concentration

When you select Start to commence a Concentration run, the **Standard/Sample Selection** dialog will appear. Once you have selected your solutions, various prompting dialogs will appear. The sequence that occurs depends upon your storage and accessory selections.

Storage selections

<table>
<thead>
<tr>
<th>If you have selected … as the Storage option on the Auto Store page</th>
<th>The following occurs</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>On, prompt at start</strong></td>
<td>The Windows <strong>Save As</strong> dialog will appear once you have made your Standard and Sample selections. You can save the current method, collected data, report and graph template as a Batch file.</td>
</tr>
<tr>
<td><strong>On, prompt at End</strong></td>
<td>The Windows <strong>Save As</strong> dialog will appear at the end of the data collection. You can save the current method, collected data, report and graph template as a Batch file.</td>
</tr>
<tr>
<td><strong>Off</strong></td>
<td>The Windows <strong>Save As</strong> dialog will not appear.</td>
</tr>
</tbody>
</table>

Accessory selections

<table>
<thead>
<tr>
<th>If you have selected … on the Accessories page</th>
<th>The following is displayed</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>No accessories</strong></td>
<td>If you have included Standards in the analysis, the <strong>Present Standard</strong> dialog will appear after the <strong>Standard/Sample Selection</strong> dialog, prompting you to load the standard. You should manually present your first standard solution then select OK to commence the read. Separate <strong>Present Standard</strong> dialogs will appear for each standard. After reading the standards, the <strong>Present Sample</strong> dialog will appear for each selected sample.</td>
</tr>
<tr>
<td><strong>Multicell Holder</strong></td>
<td>The <strong>Cell Loading Guide</strong> dialog will appear. You should load the cells as depicted then select OK to commence the read. After</td>
</tr>
</tbody>
</table>
Present Standard—Concentration

Use the Present Standard dialog to enable reading each standard. The dialog will appear for each selected sample.

To read the standard, place the solution in the sample compartment and then select OK. The Cary Eclipse will measure the standard, plot the value in the Graphics area and record the measurement in the Report area.

Switch to samples

Select this check box and select OK to stop reading the standards and commence reading the samples. The Cary Eclipse will display the Present Sample dialog where you can start measuring sample concentration.

For example, if you have set up the run to include five standards, and after the third standard the calibration is satisfactory, it may not be necessary to read the rest of the standards.

Note:
The Present Standard dialog will only be displayed once you select Start to commence an analysis (and have selected your standards and samples) if you do not have any accessories selected.

See also:
Standard/Sample Selection
Cell Loading Guide
Present Standard
Present Sample

Present Sample—Concentration

After reading the standards, the Present Sample dialog will appear for each selected sample. The Present Sample dialog prompts you to place your sample in the sample compartment. Separate Present Sample dialogs will appear for each sample.

To read the standard, place the solution in the sample compartment and then select OK. The Cary Eclipse will measure the sample, plot the value in the Graphics area and record the measurement in the Report area.

Note:
The Present Sample dialog will only be displayed once you select Start to commence an analysis (and have selected your standards and samples) if you do not have any accessories selected.

See also:
Standard/Sample Selection
Present Sample
Information about starting a Concentration run

Cell Loading Guide—Concentration
If you are using a Multicell Holder, the **Cell Loading Guide** dialog appears after the **Standard/Sample Selection** dialog when you select the **Start** button to commence an analysis.

The **Cell Loading Guide** dialog prompts you to insert blank samples in the Multicell Holder to collect the scans. You can also use the dialog to name the samples in each selected cell in the Multicell Holder.

The dialog comprises the following:

**Loading Guide/Sample Name entry**

Enter the names of the sample to be used in the concentration run in each available field. This sample name will be used as the Name of Trace in the Trace Preferences dialog.

**Note**

The Cell Loading Guide dialog appears only if you have selected the **Multicell Holder** option on the **Accessories** page of the **Setup** dialog. Only cells selected in the Multicell Holder diagram on the **Accessories** page will be available in the **Cell Loading Guide**.

In the adjacent example, cells 1—3 will be available in the Loading Guide dialog, as they have been selected in the Multicell Holder diagram.

You should load the cells as depicted then select **OK** to commence the read. After reading the standards, the **Cell Loading Guide** will prompt you to load your samples.

See also:

- **Accessories page**
- **Standard/Sample Selection dialog**
- **Present Standard**
- **Present Sample**

**Start/Stop instrument buttons—Concentration**

The following buttons may be displayed at the top of the Concentration Application window. The **Start** and **Connect** buttons will change depending upon the status of the instrument. For example, if the instrument is ready for a run, the **Start** button will be visible to indicate that the system is ready to commence a run.

Select **Start** to commence a concentration run using the current **Setup** dialog parameters.

If any unsaved data exists in the Graphics area you will be prompted to save the data. If you choose not to save, any unsaved data, pictures and labels will be deleted when you start the concentration run.

If another application is already online with the Cary Eclipse, the **Start** button will change to **Connect**, and you will need to select **Connect** to bring this application online before the **Start** button will re-appear. (You will not be able to connect if the other application is collecting data.)

Select this to end a concentration run.

If you have selected **Storage On** in the **Auto Store** page of the **Setup** dialog, all data collected is automatically stored in the file specified in the Batch name field of the **Windows Save As** dialog before or after the run commenced.

See also:
Pre-scan—Concentration

Select Pre-scan to determine the optimum instrument parameters to be used for your sample. At the end of this collection sequence the software will report the optimum Ex and Em wavelengths and PMT voltage. Once you are satisfied with the parameters, the software will automatically update your method setup to reflect these new parameters.

To access the Pre-scan option, select the Pre-scan command from the Commands menu, or select the Pre-scan button from the Concentration Application window.

The Pre-scan dialog comprises the following:

Type of scatter

Select the type of scatter you wish to search for from the following:

- **Rayleigh scatter**: Rayleigh scatter refers to the scatter of the excitation light. It is displayed at the peak of the excitation wavelength.
- **Raman scatter**: Raman scatter refers to scatter due to the solvent. Its peak is at a fixed energy from the excitation wavelength, and is solvent dependent. Thus, the type of solvent used must be selected from the Solvent list.
- **2nd order**: Second order scatter refers to the scatter of higher order excitation light. Its peak is a multiple of the excitation wavelength.

See also:
- Commands menu
- Concentration Application

Reread—Concentration

The Reread option is only available once you have completed a data collection. Use the Reread option to reread samples that have already been run.

The Reread option is accessible by selecting the Reread button from the Concentration Application window, or from the Commands menu by selecting Reread. When you select the Reread option, the Standard/Sample Selection dialog is displayed.

See also:
- Standard/Sample Selection dialog
- Commands menu
- Concentration Application

Rapid Result—Concentration

Use the Rapid Result dialog to take a reading of a blank and sample/s, without having to change the current application method. This means that between runs, while new samples are being prepared and loaded, other samples can quickly be measured. The result is printed in the Report area.
To access the Rapid Result dialog, select the Rapid Result command from the Commands menu or press F6.

**Excitation**

*Wavelength*

Enter the wavelength at which you want to take a reading.

*Zero order*

Select this option to set the excitation monochromator to zero order (white light).

**Emission**

*Wavelength*

Enter the wavelength at which you want to take a reading.

*Zero order*

Select this option to set the excitation monochromator to zero order (white light).

**Zero**

Click Zero to change the wavelength to the entered value and measure a blank.

**Result**

Click Result to change the wavelength to the entered value and measure a sample. The X,Y results are corrected for the blank and appear in the reports area.

---

**Zero—Concentration**

Use the Zero button to zero the current intensity value. The intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before commencing a data collection if you want your data to start at 0 intensity. The Cary Eclipse will prompt you to load a blank before performing the zero.

Before you select Zero, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.

When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the Concentration Application window.

Select the Clear Zero command from the Commands menu to remove zero correction and thus remove the subtraction effect.

---

**Go To Wavelength—Concentration**

Use the Go To Wavelength dialog to temporarily change the instrument to a new excitation or emission wavelength. This dialog is useful if you want to quickly go to a wavelength and check the intensity reading of the sample. The lamp will flash continuously in this mode. This option is only available if the instrument is not currently collecting data.

To access the Go To Wavelength dialog, select the Go To… command from the Commands menu. You can also select F4 to open the dialog.

---

**Note**

This is not the wavelength used in the application method. If your method uses a wavelength that is different from the 'Go to wavelength' setting you select, then when you select Start the system will
change to the wavelength selected (as set on the Cary page of the Setup dialog).

The Go To Wavelength page comprises the following:

**Excitation**  
*Wavelength*  
Enter the new wavelength in this numeric entry field. When you select Go To the instrument will change to the new value.

*Zero Order*  
Select Zero Order to drive the Cary Eclipse fluorescence spectrophotometer to 0 nm (white light). You can then use this light for aligning accessories.

**Emission**  
*Wavelength*  
Enter the new wavelength in this numeric entry field. When you select Go To the instrument will change to the new value.

*Zero Order*  
Select Zero Order to drive the Cary Eclipse fluorescence spectrophotometer to 0 nm (white light). You can then use this light for aligning accessories.

**Go To**  
Click Go To once you have entered a new wavelength, to change the instrument to the entered value.

See also:  
[Function Keys](#)

## Commands menu—Concentration

Use the Commands menu to access major operational commands in the Concentration application. The menu is accessible from the menu line at the top of the Concentration Application window.

Alternatively you can use function key shortcuts for some of the commands. These appear next to the item in the Commands menu. Note that not all commands appear as both button and menu items.

**Hot Tip**  
If you access these commands using the Commands menu, rather than from the application buttons, you can remove these buttons from your application window. To do this, clear the Buttons command in the View menu. This will enlarge the Graphics area, making viewing and selection of scans easier.

Tell me how to remove or display the command buttons

The Commands menu comprises the following commands:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
</table>
| **Start** | Select this to start a data collection using the currently set up parameters.  
**Note**  
If the Connect command is displayed in place of the Start command, this indicates that another Cary Eclipse application is in use and has control of the Cary Eclipse instrument. To make the Concentration application active so that you can perform a read, you will need to select Connect. However, you will not be able to select Connect if another application is collecting data. |
| **Stop** | Select this to stop the data collection.  
If you have selected Storage On in the Auto Store page of the Setup dialog, all data |
collected is automatically stored in the file specified in the Batch name field of the Windows
Save As dialog before or after the run commenced.

<table>
<thead>
<tr>
<th>Clear Zero</th>
<th>Select this option to clear the Zero command. When you select the Zero command (from the Commands menu or by pressing F5), the intensity at that time will be reset to zero and all subsequent results will have the intensity value that was reset to zero subtracted from them. Selecting Clear zero cancels this zeroing (and thus subtraction) effect.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pre-scan</td>
<td>Select Pres-can to determine the optimum instrument parameters to be used for your sample. At the end of this collection sequence the software will report the optimum Ex and Em wavelengths and PMT voltage. Once you are satisfied with the parameters, the software will automatically update your method setup to reflect these new parameters.</td>
</tr>
<tr>
<td>Zero order…</td>
<td>Select the Zero order command to set one of the monochromators to zero order. If you use this command to zero either the excitation or emission monochromators, this will override the Ex. Wavelength or Em. Wavelength set on the Cary page. The Cary Eclipse software will use the zero setting when collecting data. The Zero order command is generally used during alignment procedures.</td>
</tr>
<tr>
<td><strong>Excitation</strong></td>
<td>Select this command to set the excitation monochromator to zero order (white light).</td>
</tr>
<tr>
<td><strong>Emission</strong></td>
<td>Select this command to set the emission monochromator to zero order (white light).</td>
</tr>
<tr>
<td>Reset</td>
<td>Select this command to re-initialize the instrument. This will perform the same initializing procedure that is performed when the instrument is powered on.</td>
</tr>
<tr>
<td>Reread</td>
<td>Select this to display the Rapid Result dialog. Use this dialog to take a reading of a blank and multiple sample readings without changing the application method. This means that between runs, while new samples are being prepared and loaded, other samples can quickly be measured. This command can also be accessed by pressing F6.</td>
</tr>
<tr>
<td>Zero</td>
<td>Select this to zero the current intensity value. The intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before selecting Start if you want your data to start at 0 intensity. This command is also accessible by pressing F5.</td>
</tr>
<tr>
<td><strong>Note</strong></td>
<td>Before you select Zero, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.</td>
</tr>
<tr>
<td>Go to…</td>
<td>Select Go To to display the Go To Wavelength dialog where you can change the instrument to a new wavelength. Use this command if you want to quickly go to a wavelength to check the intensity reading. This command is only available if the instrument is not currently collecting data. It can also be accessed by pressing F4.</td>
</tr>
<tr>
<td><strong>Note</strong></td>
<td>This is not the wavelength used in the application method. If your method uses a wavelength that is different from the 'Go To wavelength', then when you select Start the system will change to the wavelength selected. (In most applications, this is set in the Cary tab of the Setup dialog.)</td>
</tr>
<tr>
<td>ADL Command</td>
<td>Select this to display the ADL Command Line window where you can execute an ADL command or ADL program. (You can also press Alt-Y to display this dialog.)</td>
</tr>
</tbody>
</table>
How to perform a calibration—Concentration

This demonstrates how to perform and store a fluorescence multi-standard calibration, without measuring any sample concentrations.

Follow the steps through, adjusting them to meet the requirements of your particular data collection.

- Start the Concentration application
- Set up data collection parameters
- Set up instrument parameters
- Set up the display options
- Set up accessories
- Set up the calibration
- Set up your samples
- Set up reporting and printing requirements
- Set up storage of collected data
- Set up visual system monitoring
- Finish Setup
- Zero the instrument
- Perform the calibration

Start the Concentration application

Step 1

Click the Windows Start button, point to Programs, Cary Eclipse and then select Scan. The Scan Application is displayed.

Note

If you are running a GLP system, you may be prompted to enter a password before accessing the Concentration application.

Set up data collection parameters

Step 2 [Setup dialog]

Select Setup to display the Setup dialog and specify the method parameters for a new method.

To do this:

Set up instrument parameters

Step 3 [Setup dialog | Cary page]
a. In the **Data mode** field set the mode to **Fluorescence**.

b. In the **Ex. Wavelength** field, set the wavelength of the excitation monochromator, which is the wavelength value at which you want to excite your sample.

c. In the **Em. Wavelength** field, set the wavelength of the emission monochromator, which is the wavelength value at which your sample emits.

d. Set the slit width of the excitation monochromator.
   Note that as a general rule, at a fixed emission slit width, increasing the excitation slit width will result in an increase in the fluorescence excitation intensity.

e. Set the slit width of the emission monochromator.
   Note that as a general rule, at a fixed excitation slit width, a two-fold increase in the emission slit width will result in a four-fold increase in the emission intensity. Accordingly, a two-fold decrease in the emission slit width will result in a four-fold decrease in the emission intensity.

f. In the **Ave Time** field enter the required value. A good starting point is 0.1 sec.

**Set up the display options**

Step 4 [Setup dialog | Options page]

a. Select the **Options** tab. The **Options** page is displayed.

b. Enter the minimum and maximum Y values to be displayed on the graph axis during the data collection.

c. Set the **Excitation filter** for the concentration measurement. It is recommended that you set the filter to **Auto** (the default setting). This will automatically move the filter wheel to the appropriate position for the selected excitation wavelength.

d. Set the **Excitation filter** and the **Emission filter** for the concentration measurement. In order to minimize steps in the spectra, it is recommended that you set the filter to **Open** (the default setting).

e. Set the EHT or sample detector PMT. The setting you choose will depend upon your sample. For example, a **Low** setting is recommended for highly emitting samples, while a **High** setting is recommended for low emitting samples.

**Set up accessories**

Step 5 [Setup dialog | Accessories page]

Select the **Accessories** tab and ensure that no accessories are selected.

**Set up the calibration**

Step 6 [Setup dialog | Standards page]

a. Select the **Standards** tab to access the **Standards** page where you can set up the standards and their parameters associated with the data collection.

b. Set the appropriate **Units** for your standards for reporting purposes.

c. In the **Standards** field, enter the number of standards that you are using in your calibration. The table
below this field will expand or contract to match your choice.

d. In the Standards table, enter the concentration of each standard in the Conc column.

e. Select the type of curve fitting required for your calibration in the Fit Type group.

f. Enter the required $R^2$ value or correlation coefficient in the Min $R^2$ field. The closer the number is to 1.000 the better the fit. Typically, 0.95 is used.

Set up your samples

Step 7 [Setup dialog | Samples page]

As this is a calibration only, set the No. of Samples field to ‘0’.

Set up reporting and printing requirements

Step 9 [Setup dialog | Reports page]

a. Select the Reports tab to display the Reports page where you can specify your reporting requirements for this method.

b. Enter your name in the Name field.

c. Enter a comment relating to your experiment in the Comment field (optional).

d. Set up your report style by selecting the appropriate check boxes in the Options group.

For example, select the Auto Print check box to obtain a printout of your report automatically. Select the Parameters check box to include your experimental parameters in the report. Select the Graph check box to include a calibration graph in the generated report.

Note

If Auto Print is selected, then the system will send the report information to the specified printer as well as the Report area. However if Auto Print is not selected, the report will only be sent to the Report area and can be viewed by selecting the Report command from the View menu.

Set up storage of collected data

Step 10 [Setup dialog | Auto Store page]

a. Select the Auto Store tab to display the Auto Store page.

b. Set the Storage option to Off. The method (with calibration), collected data and report will not be automatically saved. However, you can manually save it all at the end of the collection.

c. Select the Auto Convert option you require. If you select ASCII (csv) or ASCII (csv) with log, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format. The data file will be saved in the current folder.

Set up visual system monitoring
Step 11 [Setup dialog]

Select the **Status Display** check box on any of the Setup pages to display various information fields on your current reaction.

**Finish Setup**

Step 12

Once you are satisfied with your method setup select **OK** to confirm any changes you have made and clear the **Setup** dialog.

**Zero the instrument**

Step 13

a. From the Concentration Application window, select the **Zero** button to zero the system. Alternatively, select the **Zero** command from the **Commands** menu. The **Zero** dialog will be displayed.

b. Place a blank in the sample compartment and select **OK**.

**Perform the calibration**

Step 14

a. Select the **Start** button or select **Start** from the Commands menu. The **Standard/Sample Selection** dialog will be displayed.

b. Select the standards and samples to be used in the analysis. By default, all standards are selected.

   **Hot Tip**
   If you accidentally included samples on the **Samples** page, highlight them and use the arrow keys to move them to the **Solutions Available** list so they will not be read.

c. Select **OK** to clear the **Standard/Sample Selection** dialog.

d. The **Present Standard** dialog will be displayed to prompt you to place the appropriate standard in the sample compartment. Press **OK** to measure the standard.

e. Repeat Step 14 d until you have measured all the standards. The Cary Eclipse will calculate the calibration and the correlation coefficient.

   **Note**
   If the set correlation coefficient ($R^2$) value is not met, the Cary Eclipse will prompt you with ‘Min $R^2$ test failed’.

Step 15

a. Once the run is finished, select the **Save As** command from the **File** menu.

b. In the **File name** field, enter a file name for this Concentration run.
c. Select **Save** and save the method with the calibration as a Method (*.FMCN) file.

See also:
- How to change the curve of best fit used in the calibration
- How to delete a standard from the calibration
- How to import samples labels, weights and volumes
- How to export collected data
- How to generate a report from previously calculated data

**How to perform a calibration and manually measure concentrations—Concentration**

This demonstrates how to perform a fluorescence multi-standard calibration and measure sample concentrations using no accessories.

Follow the steps through, adjusting them to meet the requirements of your particular data collection.

- Start the Concentration application
- Set up data collection parameters
- Set up instrument parameters
- Set up display options
- Set up accessories
- Set up the calibration
- Set up your samples and increment names
- Set up your samples for weight and volume correction
- Set up reporting and printing requirements
- Set up storage of collected data
- Set up visual system monitoring
- Finish Setup
- Zero the instrument
- Perform the calibration
- Measure sample concentration
- Save your data

**Start the Concentration application**

Step 1

Click the Windows **Start** button, point to **Programs, Cary Eclipse** and then select **Scan**. The Scan Application is displayed.

**Note**

If you are running a GLP system you may be prompted to enter a password before accessing the Scan Application.

**Set up data collection parameters**

Step 2 [Setup dialog]

Select **Setup** to display the **Setup** dialog and specify the method parameters for a new method.

To do this:
Set up instrument parameters

Step 3 [Setup dialog | Cary page]

a. In the **Data mode** field set the mode to **Fluorescence**.

b. In the **Ex. Wavelength** field, set the wavelength of the excitation monochromator, which is the wavelength value at which you want to excite your sample.

c. In the **Em. Wavelength** field, set the wavelength of the emission monochromator, which is the wavelength value at which your sample emits.

d. Set the slit width of the excitation monochromator.
   Note that as a general rule, at a fixed emission slit width, increasing the excitation slit width will result in an increase in the fluorescence excitation intensity.

e. Set the slit width of the emission monochromator.
   Note that as a general rule, at a fixed excitation slit width, a two-fold increase in the emission slit width will result in a four-fold increase in the emission intensity. Accordingly, a two-fold decrease in the emission slit width will result in a four-fold decrease in the emission intensity.

f. In the **Ave Time** field enter the required value. A good starting point is 0.1 sec.

Set up the options

Step 4 [Setup dialog | Options page]

a. Select the **Options** tab. The **Options** page is displayed.

b. Enter the minimum and maximum Y values to be displayed on the graph axis during the data collection.

c. Set the **Excitation filter** for the concentration measurement. It is recommended that you set the filter to **Auto** (the default setting). This will automatically move the filter wheel to the appropriate position for the selected excitation wavelength.

d. Set the **Excitation filter** and the **Emission filter** for the concentration measurement. In order to minimize steps in the spectra, it is recommended that you set the filter to **Open** (the default setting).

e. Set the EHT or sample detector PMT. The setting you choose will depend upon your sample. For example, a **Low** setting is recommended for highly emitting samples, while a **High** setting is recommended for low emitting samples.

Set up accessories

Step 5 [Setup dialog | Accessories page]

Select the **Accessories** tab and ensure that no accessories are selected.

Set up the calibration

Step 6 [Setup dialog | Standards page]

a. Select the **Standards** tab to access the **Standards** page where you can set up the standards and their
parameters associated with the data collection.

b. Set the appropriate Units for your standards for reporting purposes.

c. In the Standards field, enter the number of standards that you are using in your calibration. The table below this field will expand or contract to match your choice.

d. In the Standards table, enter the concentration of each standard in the Conc column.

e. Select the type of curve fitting required for your calibration in the Fit Type group.

f. Enter the required $R^2$ value or correlation coefficient in the Min $R^2$ field. The closer the number is to 1.000 the better the fit. Typically, 0.95 is used.

**Set up your samples and increment names**

Step 7 [Setup dialog | Samples page]

a. Select the Samples tab to display the Samples page. This page enables you to enter a list of sample names that will be used during your analysis.

b. Enter the number of samples that you are going to use in the No. of Samples field. The table below this field will expand or contract to match your choice.

c. In the Sample names table enter the name of each sample. You can enter up to 20 characters for each name.

d. If you would like the samples to have the same name with a different numeric extension, enter the name in the first sample position and then select the Increment button.

**Set up weight and volume correction**

Step 8 [Setup dialog | Samples page]

a. Select the Weight/Volume Corrections check box.

b. Enter the theoretical sample weight in the Method Weight field. This is the weight of the sample specified in your method. (The Actual Weight is entered for each sample in the Sample Names list.)

c. Enter the units in the Weight Units field.

d. Enter the theoretical sample volume in the Method Volume field. This is the volume to which the method tells you to make the sample. (The Actual Volume is entered for each sample in the Sample Names list.)

e. Enter the units in the Volume Units field.

f. In the Sample Names table, enter the Actual Weight and Actual Volume for each sample.

**Set up reporting and printing requirements**

Step 9 [Setup dialog | Reports page]
a. Select the **Reports** tab to display the **Reports** page where you can specify your reporting requirements for this method.

b. Enter your name in the **Name** field.

c. Enter a comment relating to your experiment in the **Comment** field (*optional*).

d. Set up your report style by selecting the appropriate check boxes in the **Options** group.

   For example, select the **Auto Print** check box to obtain a printout of your report automatically. Select the **Parameters** check box to include your experimental parameters in the report. Select the **Graph** check box to include a calibration graph in the generated report.

**Note**

If **Auto Print** is selected, then the system will send the report information to the specified printer as well as the Report area. However if **Auto Print** is not selected, the report will only be sent to the Report area and can be viewed by selecting the **Report** command from the **View** menu.

**Set up storage of collected data**

**Step 10** [Setup dialog | Auto Store page]

a. Select the **Auto Store** tab to display the **Auto Store** page.

b. Set the Storage option to **Off**. The method (with calibration), collected data and report will not be automatically saved. However, you can manually save it all at the end of the collection.

c. Select the **Auto Convert** option you require. If you select **ASCII (csv)** or **ASCII (csv) with log**, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format. The data file will be saved in the current folder.

**Set up visual system monitoring**

**Step 11** [Setup dialog]

Select the **Status Display** check box on any of the Setup pages to display various information fields on your current reaction.

**Finish Setup**

**Step 12**

Once you are satisfied with your method setup select **OK** to confirm any changes you have made and clear the **Setup** dialog.

**Zero the instrument**

**Step 13**

a. From the Concentration Application window, select the **Zero** button to zero the system. Alternatively, select the **Zero** command from the **Commands** menu. The **Zero** dialog will be displayed.
b. Place a blank in the sample compartment and select **OK**.

### Perform the calibration

**Step 14**

a. Select the **Start** button or select **Start** from the Commands menu. The **Standard/Sample Selection** dialog will be displayed.

b. Select the standards and samples to be used in the analysis. By default, all standards and samples are selected.

c. Select **OK** to clear the **Standard/Sample Selection** dialog.

d. The **Present Standard** dialog will be displayed to prompt you to place the appropriate standard in the sample compartment. Press **OK** to measure the standard.

e. Repeat Step 14 d until you have measured all the standards. The Cary Eclipse will calculate the calibration and the correlation coefficient.

#### Note

If the set correlation coefficient ($R^2$) value is not met, the Cary Eclipse will prompt you with ‘Min R$^2$ test failed’. When you select **OK**, the Cary Eclipse will then prompt you with ‘There is no valid calibration. Proceed in Intensity (a.u.)?’. If you select **Cancel**, the Concentration run will finish. If you select **OK**, the Cary Eclipse will measure the intensity of any presented samples, but will not generate a concentration.

### Measure sample concentration

**Step 15**

a. Once all the standards have been read, the **Present Sample** dialog will be displayed to prompt you to place the appropriate sample in the sample compartment. Press **OK** to measure the sample and calculate its concentration. (If replicates have been nominated, then the concentration is calculated after the final sample replicate is read.)

b. Repeat Step 15 b for the remaining samples.

### Save your data

**Step 16**

a. Once the run is finished, select the **Save As** command from the **File** menu.

b. In the **File name** field, enter a file name for this Concentration run.

c. Select **Save**. The data will be stored as a Batch file.

#### See also:

- How to change the curve of best fit used in the calibration
- How to delete a standard from the calibration
- How to import samples labels, weights and volumes
- How to perform a calibration and measure concentrations using the Multicell Holder accessory
How to perform a calibration and measure concentrations using the Multicell Holder—Concentration

This demonstrates how to perform a fluorescence multi-standard calibration and measure sample concentrations using the Multicell Holder.

Follow the steps through, adjusting them to meet the requirements of your particular data collection.

- Start the Concentration application
- Set up data collection parameters
- Set up instrument parameters
- Set up display options
- Set up Multicell Holder accessory
- Set up the calibration
- Set up your samples and increment names
- Set up weight and volume correction
- Set up reporting and printing requirements
- Set up storage of collected data before run
- Set up visual system monitoring
- Finish Setup
- Zero the instrument
- Perform the calibration and measure samples
- Save your data

Start the Concentration application

Step 1

Click the Windows Start button, point to Programs, Cary Eclipse and then select Scan. The Scan Application is displayed.

Note

If you are running a GLP system, you may be prompted to enter a password before accessing the Concentration application.

Set up data collection parameters

Step 2 [Setup dialog]

Select Setup to display the Setup dialog and specify the method parameters for a new method.

To do this:

Set up instrument parameters

Step 3 [Setup dialog | Cary page]

a. In the Data mode field set the mode to Fluorescence.

b. In the Ex. Wavelength field, set the wavelength of the excitation monochromator, which is the wavelength value at which you want to excite your sample.
c. In the **Em. Wavelength** field, set the wavelength of the emission monochromator, which is the wavelength value at which your sample emits.

d. Set the slit width of the excitation monochromator.
   Note that as a general rule, at a fixed emission slit width, increasing the excitation slit width will result in an increase in the fluorescence excitation intensity.

e. Set the slit width of the emission monochromator.
   Note that as a general rule, at a fixed excitation slit width, a two-fold increase in the emission slit width will result in a four-fold increase in the emission intensity. Accordingly, a two-fold decrease in the emission slit width will result in a four-fold decrease in the emission intensity.

f. In the **Ave Time** field enter the required value. A good starting point is 0.1 sec.

**Set up the display options**

Step 4 [Setup dialog | Options page]

a. Select the **Options** tab. The **Options** page is displayed.

b. Enter the minimum and maximum Y values to be displayed on the graph axis during the data collection.

c. Set the **Excitation filter** for the concentration measurement. It is recommended that you set the filter to **Auto** (the default setting). This will automatically move the filter wheel to the appropriate position for the selected excitation wavelength.

d. Set the **Excitation filter** and the **Emission filter** for the concentration measurement. In order to minimize steps in the spectra, it is recommended that you set the filter to **Open** (the default setting).

e. Set the EHT or sample detector PMT. The setting you choose will depend upon your sample. For example, a **Low** setting is recommended for highly emitting samples, while a **High** setting is recommended for low emitting samples.

**Set up Multicell Holder accessory**

Step 5 [Setup dialog | Accessories page]

a. Select the **Accessories** tab to display the **Accessories** page.

b. Select the **Multicell Holder** check box to enable the accessory.

c. Ensure that you have this accessory installed prior to commencing the run.

d. On the Cell Holder diagram select the cells you require.

e. Select the **Multi Zero** check box to enable the Multi Zero facility.

f. Make sure that no other accessories are selected on this page.

**Set up the calibration**

Step 6 [Setup dialog | Standards page]
a. Select the **Standards** tab to access the **Standards** page where you can set up the standards and their parameters associated with the data collection.

b. Set the appropriate **Units** for your standards for reporting purposes.

c. Select **Replicates** or **Std Averaging** if required, and enter the appropriate number.

d. In the **Standards** field, enter the number of standards that you are using in your calibration. The table below this field will expand or contract to match your choice.

e. In the **Standards** table, enter the concentration of each standard in the **Conc** column.

f. Select the type of curve fitting required for your calibration in the **Fit Type** group.

g. Enter the required $R^2$ value or correlation coefficient in the **Min $R^2$** field. The closer the number is to 1.000 the better the fit. Typically, 0.95 is used.

**Set up your samples and increment names**

Step 7 [Setup dialog | Samples page]

a. Select the **Samples** tab to display the **Samples** page. This page enables you to enter a list of sample names that will be used during your analysis.

b. Enter the number of samples that you are going to use in the **No. of Samples** field. The table below this field will expand or contract to match your choice.

c. In the **Sample names** table enter the name of each sample. You can enter up to 20 characters for each name.

d. If you would like the samples to have the same name with a different numeric extension, enter the name in the first sample position and then select the Increment button.

**Set up weight and volume correction**

Step 8 [Setup dialog | Samples page]

a. Select the **Weight/Volume Corrections** check box.

b. Enter the theoretical sample weight in the **Method Weight** field. This is the weight of the sample specified in your method. (The **Actual Weight** is entered for each sample in the **Sample Names** list.)

c. Enter the units in the **Weight Units** field.

d. Enter the theoretical sample volume in the **Method Volume** field. This is the volume to which the method tells you to make the sample. (The **Actual Volume** is entered for each sample in the **Sample Names** list.)

e. Enter the units in the **Volume Units** field.

f. In the **Sample Names** table, enter the **Actual Weight** and **Actual Volume** for each sample.

**Set up reporting and printing requirements**
Step 9 [Setup dialog | Reports page]

a. Select the **Reports** tab to display the **Reports** page where you can specify your reporting requirements for this method.

b. Enter your name in the **Name** field.

c. Enter a comment relating to your experiment in the **Comment** field *(optional).*

d. Set up your report style by selecting the appropriate check boxes in the **Options** group.

   For example, select the **Auto Print** check box to obtain a printout of your report automatically. Select the **Parameters** check box to include your experimental parameters in the report. Select the **Graph** check box to include a calibration graph in the generated report.

**Note**

If **Auto Print** is selected, then the system will send the report information to the specified printer as well as the Report area. However if **Auto Print** is not selected, the report will only be sent to the Report area and can be viewed by selecting the **Report** command from the **View** menu.

Set up storage of collected data before run

Step 10 [Setup dialog | Auto Store page]

a. Select the **Auto Store** tab to display the **Auto Store** page.

b. Set the **Storage** option to **Off**. The method (with calibration), collected data and report will not be automatically saved. However, you can manually save it all at the end of the collection.

c. Select the **Auto Convert** option you require. If you select **ASCII (csv)** or **ASCII (csv) with log**, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format. The data file will be saved in the current folder.

Set up visual system monitoring

Step 11 [Setup dialog]

Select the **Status Display** check box on any of the Setup pages to display various information fields on your current reaction.

Finish Setup

Step 12

Once you are satisfied with your method setup select **OK** to confirm any changes you have made and clear the **Setup** dialog.

Zero the instrument

Step 13

a. From the Concentration Application window, select the **Zero** button to zero the system. Alternatively,
select the **Zero** command from the **Commands** menu. The **Zero** dialog will be displayed.

b. Place a blank in the sample compartment and select **OK**.

**Perform the calibration and measure samples**

**Step 14**

a. Select the **Start** button or select **Start** from the Commands menu. The **Standard/Sample Selection** dialog will be displayed.

b. Select the standards and samples to be used in the analysis. By default, all standards and samples are selected.

c. Select **OK** to clear the **Standard/Sample Selection** dialog.

d. The **Cell Loading Guide** dialog will be displayed. Load your standards as indicated, then select **OK** to start reading the standards. If additional **Cell Loading Guide** dialogs are displayed, continue to load your standards as indicated and select **OK** to continue loading the standards. Cary Eclipse will measure the standards and calculate the calibration curve.

e. A **Cell Loading Guide** will be displayed for the samples. Load your samples as indicated and select **OK**. The Cary Eclipse will then measure the samples and calculate their concentration.

**Note**

If the set correlation coefficient (R²) value is not met, the Cary Eclipse will prompt you with ‘Min R² test failed’. When you select **OK**, the Cary Eclipse will then prompt you with ‘There is no valid calibration. Proceed in Intensity (a.u.)?’. If you select **Cancel**, the Concentration run will finish. If you select **OK**, the Cary Eclipse will measure the intensity of any presented samples, but will not generate a concentration.

**Save your data**

**Step 15**

a. Once the run is finished, select the **Save As** command from the **File** menu.

b. In the **File name** field, enter a file name for this Concentration run.

c. Select **Save**. The data will be stored as a Batch file.

**See also:**
- How to change the curve of best fit used in the calibration
- How to delete a standard from the calibration
- How to import samples labels, weights and volumes
Viewing run progress and results—Concentration

- Reports page
- Reports toolbar
- How to generate a report from previously stored data
- How to use the advanced reporting options
- View menu
- Edit menu
- Status display
- Print Preview dialog
- Print

Reports—Concentration

Use the Reports page to set up the report parameters associated with data collection.

The Reports page is accessible from the Setup dialog by selecting the Reports tab. To access the Setup dialog, select the Setup button from the Concentration Application window or the Setup command from the Setup menu.

The Reports page can also be accessed by selecting the Recalculate button and then selecting the Reports tab.

The page comprises the following:

Operator

Name  Enter the instrument operator name in this text entry field. Use the arrow to the right of this field to view and select one of the last ten entries in this field.

Note  In GLP mode the system will automatically enter the operator’s name and this field will not be editable.

Comment  Enter operator details and comments in this text entry field.

Options

This group enables you to select the information to include in your report.

Auto Print  Select Auto Print to print a report automatically during the data collection.

If Auto Print is selected, then the system will send the report information to the specified printer(s) as well as the Report area. However if Auto Print is not selected, the report will only be sent to the Report area and can be viewed by selecting Report in the View menu.

To automatically obtain a copy of the graphics, you will need to select both the Auto Print and Graph options (see below). In this case, all graphical information displayed
on screen will be printed with the report. Therefore it is advisable to arrange the graphical information as you require on screen, prior to printing. For example, if you require a single graph print out, then the **Single/Multi Graph** option in the **Graph** menu should be changed to the correct setting.

**Parameters**

Select this option to include the method setup parameters within your report.

**Graph**

Select Graph to include any graphs or graphic information associated with your report. If you require a printout of the displayed graphics only, you must ensure that *only* this option is selected. Press the Clear Report button to clear any information in the Report area before you print otherwise this will be included in your printout.

**% Page Height**

Enter the size at which you want the graph to be printed, as a percentage of total page height. For example, if you want graphical information to take up half of the page, enter 50% in this field.

**Company Logo**

Check this option to include your company logo in the report.

A company logo will only be included if you have added one on the **Company** page of the System Information Application.

**Hot Tip**

If you have not included a Company logo, select the Header/Footer button to open the System Information Application and use the Add Logo option to add a logo.

**Header/Footer**

Select the **Header/Footer** button to open the System Information application where you can enter the Header and Footer information to be included in your report. You can also enter any additional comments, and add a logo to your report. Once you have made all your changes, select **OK** to exit the System Information application and return to the **Reports** page of the Concentration application.

**Status Display**

Select this check box to display the **Status Display** window where you can view the status of your instrument during the data collection.

**Hot Tip**

You can copy a picture, spreadsheet file or document from another application (for example, Microsoft Word or Microsoft Excel) into the report area of the of the Lifetimes application. To edit these objects, simply double click on them to open the associated application in which they were created.

See also:
- Setup dialog
- Recalculate
- View menu
- Single/Multi Graph
- Status Display window

**Report Toolbar—Concentration**

Use the **Report** toolbar buttons to gain quick access to many of the **Edit** menu commands. You can also
use the toolbar to cut, copy and paste information and to change the look of your report.

To display or hide the toolbar, point to the Toolbars option in the View menu and select Report. To display or hide the toolbar's text labels, point to the Toolbar option in the View menu and select Text Labels.

Hot Tip
To see the name of a button, point to it with the mouse.

The following commands can be accessed from the Report toolbar:

<table>
<thead>
<tr>
<th>Button</th>
<th>Menu command</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Edit Report</td>
<td>Click this button to edit the report. This command is also accessible by pressing CTRL+R.</td>
<td></td>
</tr>
<tr>
<td>Undo</td>
<td>If available, this undoes the last action. This command is also accessible by pressing CTRL+Z.</td>
<td></td>
</tr>
<tr>
<td>Cut</td>
<td>Click this button to remove the highlighted text and place it on the Windows Clipboard. This command is also accessible by pressing CTRL+X.</td>
<td></td>
</tr>
<tr>
<td>Copy</td>
<td>Click this button to copy the highlighted text onto the Windows Clipboard. This command is also accessible by pressing CTRL+C.</td>
<td></td>
</tr>
<tr>
<td>Paste</td>
<td>Click this button to insert the contents of the Windows Clipboard (text only) at the cursor. Any highlighted text will be replaced. This command is also accessible by pressing CTRL+V.</td>
<td></td>
</tr>
<tr>
<td>Delete</td>
<td>Click this button to remove selected text in the Report area. This command is also accessible by pressing DEL. Note This option is only available once you select Edit Report and there is text highlighted in the Report area.</td>
<td></td>
</tr>
<tr>
<td>Font</td>
<td>Click this button to change the font of new or selected text in the report. For existing text, simply highlight the text in the report, select this button and make your changes in the Font dialog. When you select OK, the appearance of the highlighted text in the report will alter to reflect your font changes. This command is also accessible by pressing CTRL+F.</td>
<td></td>
</tr>
<tr>
<td>Preview</td>
<td>Click this button to display the Print Preview dialog where you can view the current report as it will be printed.</td>
<td></td>
</tr>
</tbody>
</table>

How to generate a report from previously calculated data—Concentration

Step 1

Select Open… from the File menu to display the Windows Open dialog.
Step 2

A list of previously stored files will appear. If this is not a list of batch files, select the down arrow at the right of the **Files of type** list box and select 'Batch' to list all the batch files.

Step 3

Select the Batch file you require then select **Open**.

The system will automatically retrieve the collected data into the Graphics area. The method that was used to collect the data will also be retrieved.

**Note**

You can also retrieve data from Methods (*.FMKN), Data (*.FDKN), Report (*.FRKN), Spreadsheet ASCII (*.CSV), GRAMS (*.SPC) and Rich Text Format (*.RTF) file types.

Step 4

Select the **Recalculate** button to display the **Setup** dialog.

Step 5

Select the **Reports** tab and on the **Reports** page specify the information you would like to include in the report. Select **Auto Print** if you require the new report to print to the Reports area and to a printer.

Step 6

Click **OK** to display the results in the Report area (and send to the printer if **Auto Print** is selected).

**Note**

If you require a printout of only the current graphics in the Graphics area of a previously stored data file:

a. Select the **Clear Report** option in the Commands menu to clear any text that may be displayed in the Report area.

b. Press the **Print** button to obtain a printout of the graphics.

Alternatively tick only the **Graph** check box in the **Reports** page to obtain a printout of graphics only.

**See also:**

*How to use advanced reporting options*

**How to use advanced reporting options—Concentration**

This outlines ways to fully customize your reports. Options available include:

- Including header and footer information
- Including a logo
- Editing the text in the report
Including header and footer information

You can have text appearing at the top and bottom of every page in your report. To specify what the text should be, do the following:

Step 1

On the Windows Taskbar, click the Windows Start button, point to Programs, Cary Eclipse and then select System Information. The System Information application appears with the Company page displayed.

Step 2

Enter the header and footer information in the Header and Footer fields.

Step 3

Click OK to implement your changes.

Including a logo

You can include a logo on the first page of your report. To specify what the logo should be, do the following:

Step 1

On the Windows Taskbar, click the Windows Start button, point to Programs, Cary Eclipse and then select System Information. The System Information application appears with the Company page displayed.

Step 2

Enter a logo using the Add Logo button. This logo will always be included on the front page of reports.

Step 3

Click OK to implement your changes.

Q. Why isn't the logo I selected included in my report?

A. If you are running the Cary Eclipse software under Windows NT you need to be logged on as an Administrator in order to specify the logo in the System Information application.

Editing the text in the report

You can edit any of the text in the Report area. Just select Edit Report from the Edit menu. You can then add, delete and change text in the Report area.
Q. The Edit Report option is not available, why not?
A. You do not have reporting editing privileges set in the GLP Administration application. The Administrator of the system will have to change your privileges before you can edit reports.

Viewing the report on the screen before printing

You can view a report before printing it. To do this, from the File menu select Print Preview. You can then ensure that the report format is correct before you print it.

Hot Tip
Use the arrow keys at the bottom of the Print Preview screen to change pages.

Cutting and pasting to a word processor

To have complete control over how your report looks you may wish to copy it to a word processor program, such as Microsoft Word. To do this:

Step 1
Open a report.

Step 2
From the Edit menu, choose Edit Report.

Step 3
Click in the Report area and highlight the text you want to export to your word processor.

Step 4
Right-click on the Report area and select Copy from the menu.

Step 5
Start up your word processor and use the Paste function (usually under the Edit menu) to place the text into a document.

See also:
How to add a label to a graph
How to add a picture to a graph
How to alter the look of your graphs
How to display a trace that is not appearing

View menu—Concentration

The View menu is accessible from the menu line at the top of the Concentration Application window.

Hot Tip
Highlight a method or data file in Explorer and simply drag and drop it onto the application to open it. Any data will be displayed in the Graphics area.

The following commands can be accessed from the **View** menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Toolbars</strong></td>
<td>Select this option to toggle the Graphics and Reports toolbar on and off. You can also select to display or hide the toolbar Text Labels.</td>
</tr>
<tr>
<td><strong>Buttons</strong></td>
<td>Select this option to toggle the left-hand button display (button area) on and off.</td>
</tr>
<tr>
<td><strong>Graphics</strong></td>
<td>Select this option to toggle the Graphics area on and off.</td>
</tr>
<tr>
<td><strong>Report</strong></td>
<td>Select this option to toggle the Report area on and off.</td>
</tr>
<tr>
<td><strong>Report Vertical</strong></td>
<td>If the Graphics option is selected, select <strong>Report Vertical</strong> to display the Graphics area, Report area and/or ADL Editor side by side vertically.</td>
</tr>
<tr>
<td><strong>ADL</strong></td>
<td>Select this option to toggle the ADL Editor on and off.</td>
</tr>
<tr>
<td><strong>ADL Vertical</strong></td>
<td>If the ADL and Report options are selected, select <strong>ADL Vertical</strong> to display the ADL Editor vertically on the right hand side of the Report area.</td>
</tr>
<tr>
<td><strong>ADL Quick Reference</strong></td>
<td>This displays the <strong>Quick Reference</strong> window which you can use to easily add a command in the correct syntax to your ADL program. A detailed description of all the commands in the Quick Reference is available by selecting the <strong>Help</strong> button on this window.</td>
</tr>
</tbody>
</table>
| **ADL Hooks** | Select this to activate a link (or hook) to an ADL program at a specific point in the application, and then run that ADL program. For example, ADL hooks have been supplied to run specific programs:  
  - on start-up of the Cary Eclipse software;  
  - when the system goes online or offline;  
  - when the Start button is selected;  
  - when a run finishes;  
  - when a zero is performed etc.  
  For the hook to work, ADL Hooks must be selected, and the appropriate ADL file for the application must be present in the Varian/Cary Eclipse WinFLR/ADL folder on the drive on which the Cary Eclipse software is installed.  
  **Note**  
  Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help. |
<p>| <strong>Hints</strong>     | Select this option to toggle the Hints bubble text on and off. Hint text appears when you place the arrow cursor over a field, button or text entry field for a short period. It offers a quick explanation of the item’s function. |</p>
<table>
<thead>
<tr>
<th>Command</th>
<th>Button</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Undo</td>
<td><img src="image" alt="Undo" /></td>
<td>If available, this undoes the last action that was performed in the report area. This command is also accessible by pressing CTRL+Z.</td>
</tr>
<tr>
<td>Cut</td>
<td><img src="image" alt="Cut" /></td>
<td>Select this command to remove the highlighted text and place it on the Windows Clipboard. This command is also accessible by pressing CTRL+X.</td>
</tr>
<tr>
<td>Copy</td>
<td><img src="image" alt="Copy" /></td>
<td>Select this command to copy the highlighted text onto the Windows Clipboard. This command is also accessible by pressing CTRL+C.</td>
</tr>
<tr>
<td>Paste</td>
<td><img src="image" alt="Paste" /></td>
<td>Select this command to insert the contents of the Windows Clipboard (text only) at the cursor. Any highlighted text will be replaced. This command is also accessible by pressing CTRL+V.</td>
</tr>
<tr>
<td>Delete</td>
<td><img src="image" alt="Delete" /></td>
<td>Select this command to remove selected text in the Report area. This command is also accessible by pressing DEL. <strong>Note</strong> This option is only available once you select Edit Report and there is text highlighted in the Report area.</td>
</tr>
<tr>
<td>Select All</td>
<td><img src="image" alt="Select All" /></td>
<td>Select this option to highlight all text in the Report area. This command is also accessible by pressing CTRL+A.</td>
</tr>
<tr>
<td>Font</td>
<td><img src="image" alt="Font" /></td>
<td>This enables you to change the font of new or selected text in the report. For existing text, simply highlight the text in the report, select Font and make your changes in the Font dialog. When you select OK, the appearance of the highlighted text in the report will alter to reflect your font changes. This command is also accessible by pressing CTRL+F. <strong>Note</strong></td>
</tr>
</tbody>
</table>

See also:
- Graphics area
- ADL Hooks
- How to insert ADL commands using the Quick Reference
- Graphics area
- Status Display

**Edit menu—Concentration**

Use the **Edit** menu to copy, cut, paste and delete information. You can also use the menu to access the Report editing commands. The menu is accessible from the menu line at the top of the Concentration Application window.

The following commands can be accessed from the **Edit** menu:

**Note**
These commands are also accessible from the Report Toolbar. From the **View** menu, point to **Toolbars** and select **Report** to display the Report Toolbar.
Status Display—Concentration

Use the Status Display window to view the status of your scan during the data collection. Depending upon settings you have for the concentration run, the following parameters may be displayed:

- Current Excitation Wavelength (nm)
- Current Emission Wavelength (nm)
- Excitation Slit Width (nm)
- Emission Slit Width
- PMT Voltage (V)

To access the Status Display, select Status Display from the View menu, or from any page of the Setup dialog. You can choose to view the Status Display at any time during a data collection.

See also:
- View menu
- Setup
- Status line

Print Preview—Concentration

Use the Print Preview dialog to view the current report before you print it. From the File menu, select Print Preview to access the dialog.

The contents of the Graphics area appear first in the preview area followed by the contents of the Report area. You can reduce or enlarge the print preview on screen. To do this, click the left mouse button in the preview area to toggle through the five available views. Click the right mouse button in the preview area to return to the full page view.

The page comprises the following commands:

<table>
<thead>
<tr>
<th>Command</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Edit Report</td>
<td>Select this option to edit the report. This command is also accessible by pressing CTRL+R.</td>
</tr>
<tr>
<td>Recalculate</td>
<td>Use the pages accessible from the Recalculate option to perform re-calculations on data that has been analyzed.</td>
</tr>
<tr>
<td>Create Report</td>
<td>Select this command to create a new report.</td>
</tr>
</tbody>
</table>

See also:
- Concentration Application
- Recalculate

Hot Tip
You can copy a picture, spreadsheet file or document from another application (for example, Microsoft Word or Microsoft Excel) into the report area of the Concentration application. To edit these objects, simply double click on them to open the associated application in which they were created.
Print—Concentration

Use the Print dialog to select the printer and pages to print of the currently displayed report as well as the number of copies you wish to print. From the File menu, select Print to access the dialog.

All graphical information displayed on screen will be printed with the report, therefore it is advisable to edit the report on the screen prior to printing. You can also preview the report prior to printing by using the Print Preview command.

If you require a printout of the graphics in the Graphics area only, select the Clear Report button to erase the contents of the Report area before you print.

Q. Why are the arrow buttons grayed out?
A. The buttons will be grayed out if the report has only one page of data.

See also:
- Print

Print—Concentration

Use the Print dialog to select the printer and pages to print of the currently displayed report as well as the number of copies you wish to print. From the File menu, select Print to access the dialog.

All graphical information displayed on screen will be printed with the report, therefore it is advisable to edit the report on the screen prior to printing. You can also preview the report prior to printing by using the Print Preview command.

If you require a printout of the graphics in the Graphics area only, select the Clear Report button to erase the contents of the Report area before you print.

Note
The options on the Reports page are used to generate the contents of the Report area during a run or on a recalculation. If Auto Print has been selected the Cary Eclipse will automatically print a report at the end of a data collection.

See also:
- File menu
- Print Preview
- Reports
- Edit Report
Performing Calculations—Concentration

- Recalculate
- How to generate a report from previously stored data
- How to change the curve of best fit used in the calibration
- How to determine the absorptivity of a sample

Recalculate—Concentration

Select the Recalculate button to access Standards and Reports pages that appear in the Setup dialog. These enable you to recalculate the calibration or change the information that appears on the report.

The Recalculate button is accessible from the Concentration Application window. When you select this option, the Recalculate dialog will appear, containing the following page:

- Standards
- Reports

See also:
Concentration Application window
Setup

How to generate a report from previously calculated data—Concentration

Step 1
Select Open… from the File menu to display the Windows Open dialog.

Step 2
A list of previously stored files will appear. If this is not a list of batch files, select the down arrow at the right of the Files of type list box and select 'Batch' to list all the batch files.

Step 3
Select the Batch file you require then select Open.

The system will automatically retrieve the collected data into the Graphics area. The method that was used to collect the data will also be retrieved.

Note
You can also retrieve data from Methods (*.FMKN), Data (*.FDKN), Report (*.FRKN), Spreadsheet ASCII (*.CSV), GRAMS (*.SPC) and Rich Text Format (*.RTF) file types.

Step 4
Select the Recalculate button to display the Setup dialog.
Step 5

Select the **Reports** tab and on the **Reports** page specify the information you would like to include in the report. Select **Auto Print** if you require the new report to print to the Reports area and to a printer.

Step 6

Click **OK** to display the results in the Report area (and send to the printer if **Auto Print** is selected).

**Note**

If you require a printout of only the current graphics in the Graphics area of a previously stored data file:

a. Select the **Clear Report** option in the Commands menu to clear any text that may be displayed in the Report area.
b. Press the **Print** button to obtain a printout of the graphics.

Alternatively tick only the **Graph** check box in the **Reports** page to obtain a printout of graphics only.

**See also:**

[How to use advanced reporting options](#)

**How to change the curve of best fit used in the calibration—Concentration**

- [Open stored calibration and sample data](#)
- [Change calibration curve type](#)
- [Start recalculation](#)

Once the standards have been measured and the calibration curve plotted on the screen, you may wish to change the type of curve used to fit the standard points.

**Open stored calibration and sample data**

Step 1

From the **File** menu, select **Open** to display the Windows **Open** dialog.

A list of previously stored batch files will appear. Select the batch file you require then select the **Open** button.

The system will automatically retrieve the method and calibration that was used to measure the samples as well as the sample data.

**Change calibration curve type**

Step 2
Select the Recalculate button to display the Recalculate dialog. This dialog contains the Standards and Reports pages which enable you to set up the recalculation parameters.

Step 3

Select the Standards tab. The Standards page is displayed.

Step 4

Select the Fit Type algorithm by selecting the button that corresponds to the fit type you require.

Start recalculation

Step 5

Select OK to perform the calibration using the selected Fit Type.

The Cary Eclipse will create a calibration curve by fitting the selected fit curve to the data results. The calibration equation and correlation coefficient will also be sent to the Report area along with the recalculated sample results.

See also:
- How to delete a standard from the calibration
- How to export collected data
- How to generate a report from previously calculated data
- How to remove or display the command buttons

**How to determine the absorptivity of a sample—Concentration**

Step 1

Using the Concentration application, prepare a calibration curve using the appropriate standards.

Step 2

Create a calibration curve using Linear Direct as the Fit Type.

Step 3

Now look at the calibration equation that is reported in the calibration report. e.g. \( \text{Abs} = 0.05692 \times \text{Conc} \). Using the Beer-Lambert law, which states that:

\[ A = \varepsilon \ell c \]

where:
- \( A \) = absorbance
- \( \varepsilon \) = absorptivity, \( \text{cm}^{-1} \text{M}^{-1} \)
- \( \ell \) = pathlength, cm
- \( C \) = concentration of the solute, M
You can see that, in this example, $0.05692 = \varepsilon l$, or the slope of the calibration curve.

Knowing the pathlength of your cell, you can therefore calculate the absorptivity.

For example:

If cell pathlength ($l$) = 1 cm, then Absorptivity ($\varepsilon$) = $0.05692 \text{ cm}^{-1}\text{M}^{-1}$
If cell pathlength ($l$) = 0.2 cm, then Absorptivity ($\varepsilon$) = $0.2846 \text{ cm}^{-1}\text{M}^{-1}$

See also:
- How to perform a calibration
- How to perform a calibration and manually measure concentrations
- How to perform a calibration and measure concentrations using the Multicell Holder accessory
Displaying data graphically—Concentration

- Graphics area
- Graphics How To
- Graph menu
- Graphics toolbar
- Graph Labels
  - Add Label dialog
  - Edit Label dialog
  - How to add a label to a graph
  - How to edit a label
  - How to move a label
  - How to resize a label
  - How to delete a label
- Graph Pictures
  - Add Picture dialog
  - Edit Picture dialog
  - How to add a picture
  - How to replace a picture
  - How to move a picture
  - How to resize a picture
  - How to delete a picture
- Trace Preferences dialog
  - Trace Preferences menu
  - How to select the focused trace
  - How to display a trace
  - How to remove a trace from a graph
  - How to remove a trace from the trace list
- Graph Preferences dialog
  - How to alter the look of your graphs
- Cursor Modes dialog
  - How to track the focused trace
- Axes Scales dialog
  - How to scale your graph manually
  - How to scale your graph automatically
- Single/Multi Graph
- Graphics Templates
  - How to create a graphics template

Graphics area—Concentration

The Graphics area of the Concentration Application appears in the top half of the Concentration Application window and is used to display traces of collected data. In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area surrounding the graph is referred to as the graph box. A graph will appear in the Graphics area when you select Start to commence a read.

To display or hide the Graphics area, select the Graphics command on the View menu. Double-clicking on the Graphics area enables you to toggle between half or full size display sizes.
You can access the functions associated with the Graphics area by doing the following:

- selecting one of the **Graphics toolbar** buttons;
- selecting a command from the **Graph** menu; or
- clicking the right mouse button in the Graphics area and select a command from the menu

**Note**
Not all functions are available on every menu.

You can customize the graphics area to suit your requirements in the following ways:

**Note**
All of the commands and dialogs listed are accessible from the **Graph menu**.

<table>
<thead>
<tr>
<th>To…</th>
<th>Use the…</th>
<th>Tell me how…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Change the axes scaling automatically</td>
<td>Autoscale graph command</td>
<td>How to scale your graph automatically</td>
</tr>
<tr>
<td>Change the axes scaling manually</td>
<td>Axes Scales dialog</td>
<td>How to scale your graph manually</td>
</tr>
<tr>
<td>Modify the graph appearance (axes, trace)</td>
<td>Graph Preferences dialog</td>
<td>How to alter the look of your graphs</td>
</tr>
<tr>
<td></td>
<td></td>
<td>How to specify the number of tick marks and decimal places for your X and Y axes</td>
</tr>
<tr>
<td>Add a new graph to the display</td>
<td>Add graph command</td>
<td></td>
</tr>
<tr>
<td>Remove a graph from the display</td>
<td>Remove graph command</td>
<td>How to remove graphs</td>
</tr>
<tr>
<td>View single or multiple graphs</td>
<td>Single/multiple graph command</td>
<td></td>
</tr>
<tr>
<td>Resize a graph box</td>
<td>Cursor</td>
<td>How to resize graph boxes</td>
</tr>
<tr>
<td>Track the focused trace</td>
<td>Cursor Mode dialog</td>
<td>How to track the focused trace</td>
</tr>
<tr>
<td>Select the focused trace</td>
<td>Cursor</td>
<td>How to select the focused trace</td>
</tr>
<tr>
<td>Display a trace in the graph</td>
<td>Trace Preferences dialog</td>
<td>How to display a trace that is not appearing in a graph box</td>
</tr>
<tr>
<td>Remove a trace from a graph</td>
<td>Trace Preferences dialog</td>
<td>How to remove a trace from a graph</td>
</tr>
<tr>
<td>Remove a trace from the trace list</td>
<td>Trace Preferences dialog</td>
<td>How to remove a trace from the trace list</td>
</tr>
<tr>
<td>Zoom in and out on the graph</td>
<td>Zoom In and Zoom Out commands</td>
<td>How to zoom a graph</td>
</tr>
</tbody>
</table>
## Graphics How to—Concentration

### Traces

- How to track the focused trace
- How to select the focused trace
- How to display a trace that is not appearing in a graph box
- How to remove a trace from a graph
- How to remove a trace from the trace List

### Sizing and scale

- How to resize graph boxes
- How to scale your graph manually
- How to scale your graph automatically
- How to alter the look of your graphs
- How to zoom a graph

### Labels

- How to add a label to a graph
- How to edit a label

---

**Note**

The red trace in any graph box is the one on which all information appears and the one used for the cursor tracking. It is called the focused trace.

---

**See also:**

[Graphics Toolbar](#)
Graph menu—Concentration

Introduction

Use the Graph menu to view data in various graphical formats or in multiple graphs in the Graphics area. You can double-click on the Graphics area to toggle the display through graph and report to graph only.

In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area surrounding the graph is referred to as the graph box. A graph will appear in the Graphics area when you click Start to commence the concentration run.

The Graph menu can be accessed in the following ways:

- from the main menu at the top of the Concentration Application window
- by clicking the right mouse button in a graph box or the Graphics area

![Note]
The red trace in any graph box is the one on which all information appears and the one used for the cursor tracking. It is called the focused trace.

Menu commands

The following commands can be accessed from the Graph menu:

<table>
<thead>
<tr>
<th>Menu command</th>
<th>Button</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Print XY point</td>
<td>Select this to print the cursor's current X and Y coordinates in the report area. Note that this option is only available from the Graph menu that appears when you right-click in a graph box.</td>
<td></td>
</tr>
<tr>
<td>Copy Graph</td>
<td>Select Copy Graph to copy the entire Graphics area to the Windows Clipboard. All of the graphs in the Graphics area will be copied to the Clipboard.</td>
<td></td>
</tr>
<tr>
<td>Paste to Graph</td>
<td>Select Paste to graph to paste the contents of the Windows Clipboard into the Graphics area. Text is pasted as a label, graphics are pasted as pictures.</td>
<td></td>
</tr>
<tr>
<td><strong>Add Graph</strong></td>
<td>![Image]</td>
<td>Select <strong>Add graph</strong> to add a new graph box to the Graphics area. You may want to do this if you have, for example, a graph containing existing data and you want to collect some new data in a new graph.</td>
</tr>
</tbody>
</table>
| **Remove Graph** | ![Image] | Select **Remove graph** to delete the selected graph box from the Graphics area. The graph box is removed, but the data is not deleted and is still available from the **Trace Preferences** window. You may want to use remove graph if you have a large number of graph boxes open but only want to focus on some of these.  

**Note**  
You can remove multiple graph boxes by first holding down the CTRL key and selecting the graphs to be removed and then selecting the **Remove Graph** button or menu item. Use the SHIFT key to highlight a range of graphs for removal. |
| **Clear All Traces** | ![Image] | Select **Clear All Traces** to delete all traces from the graph/s. Note that selecting this option simply turns off the Visible property for the traces in the Trace Preferences window. You can redisplay the traces by making the traces Visible again in the Trace Preferences window. |
| **Add Label** | ![Image] | Select **Add Label** to display the **Add Label dialog** where you can enter a text label to be placed on a graph or in the background of the Graphics area. |
| **Add Picture** | ![Image] | Select **Add Picture** to display the **Add Picture dialog** where you can select a picture, bitmap or chemical structure to be placed on a graph or in the background of the Graphics area. |
| **Edit Annotation** | ![Image] | Select **Edit Annotation** to alter the content or look of the selected annotation, (either a label or a picture) in the Graphics area.  

**Note**  
This option can only be activated if an annotation is selected.  

If the selected annotation is a label, selecting Edit Annotation will open the **Edit Label dialog** where you can change the text of the selected label.  
If the selected annotation is a picture, selecting Edit Annotation will open the **Edit Picture dialog** where you can replace the selected picture with a new picture. |
| **Delete Annotation** | ![Image] | Select **Delete Annotation** to remove an annotation, (such as a label or a picture) from the Graphics area. You can use this menu option to remove highlighted annotations in the Graphics area such as labels, pictures and ruler/fit information.  

**Note**  
This option can only be activated if an annotation is selected. |
| **Trace Preferences** | ![Image] | Select this command to display the **Trace Preferences window** where you can select the trace(s) to display in the selected graph. |
| **Graph Preferences** | ![Image] | Select this command to display the **Graph Preferences dialog** where you can change the appearance of your graph. Any changes you make to the appearance of the graph will be applied to all graphs. |
| **Cursor Mode…** | Select this command to display the [Cursor Modes dialog](#) where you can choose the type of cursor to be used on the screen. For example, you can select to use a free cursor that can move anywhere in the graph or exactly trace the focused scan (red scan). Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the Least Squares method or the Point to Point method. |
| **Axes Scales…** | Select this command to display the [Axes Scales dialog](#) where you can set the scale range for the X and Y axes. |
| **Auto scale graph** | To automatically scale the graph, point to one of the following options:  
- **Autoscale[XY]**  
  Select Autoscale[XY] to automatically scale collected data by scaling the Y height and X width to full screen.  
- **Autoscale[X]**  
  Select Autoscale[X] to automatically scale collected data by scaling the X width to full screen.  
- **Autoscale[Y]**  
  Select Autoscale[Y] to automatically scale collected data by scaling the Y height to full screen. |
| **Zoom Out** | Select **Zoom Out** to Zoom back to the original X and Y ranges which were used to collect the data (i.e. the method setup conditions of the focused scan). For example, the system will revert back to the X and Y ranges set in the method if you zoom in and out during a data collection. |
| **Single/Multi Graph** | Select **Single/Multi Graph** to toggle between displaying one or all graphs.  
**Note**  
The graph highlighted in the **Multi Graph** view will be displayed when you toggle the **Single Graph** option. |
| **Auto Arrange Graphs** | Select **Auto Arrange Graphs** to view and arrange all graphs simultaneously.  
**Note**  
You can move graph boxes manually by clicking the left mouse button outside the graph axes and dragging the graph box to a new position. You can also resize graphs. |
| **History List** | This displays a list of the current graphs. |

---

See also:  
[Toolbar](#)  
[Graphics area](#)  
[Graphics How to…](#)

**Graphics Toolbar—Concentration**

Use the **Graphics Toolbar** buttons to gain quick access to many of the **Graph** menu options. You can
also use the toolbar to select the graph you want to view and to change the look of your graphs.

To display or hide the toolbar, point to the **Toolbars** option in the **View** menu and select **Graphics**. To display or hide the toolbar's text labels, point to the **Toolbars** option in the **View** menu and select **Text Labels**.

**Hot Tip**
To see the name of a button, point to it with the mouse.

The following commands can be accessed from the Graphics Toolbar:

<table>
<thead>
<tr>
<th>Button</th>
<th>Menu command</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image" alt="Trace Preferences" /></td>
<td>Trace Preferences</td>
<td>Click this button to display the Trace Preferences window where you can select the trace(s) to display in the selected graph.</td>
</tr>
<tr>
<td><img src="image" alt="Graph Preferences" /></td>
<td>Graph Preferences</td>
<td>Click this button to display the Graph Preferences dialog where you can change the appearance of your graph. Any changes you make to the appearance of the graph will be applied to all graphs.</td>
</tr>
<tr>
<td><img src="image" alt="Cursor Mode" /></td>
<td>Cursor Mode</td>
<td>Click this button to display the Cursor Modes dialog where you can choose the type of cursor to be used on the screen. For example, you can select to use a free cursor that can move anywhere in the graph or exactly trace the focused scan (red scan). Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the Least Squares method or the Point to Point method.</td>
</tr>
<tr>
<td><img src="image" alt="Axes Scales…" /></td>
<td>Axes Scales…</td>
<td>Click this button to display the Axes Scales dialog where you can set the scale range for the X and Y axes.</td>
</tr>
<tr>
<td><img src="image" alt="Autoscale [XY]" /></td>
<td>Autoscale [XY]</td>
<td>Click the Autoscale[X] button to automatically scale collected data by scaling the ordinate (Y) height and abscissa (X) width to full screen.</td>
</tr>
<tr>
<td><img src="image" alt="Autoscale [X]" /></td>
<td>Autoscale [X]</td>
<td>Click the Autoscale[X] button to automatically scale collected data by scaling the abscissa (X) width to full screen.</td>
</tr>
<tr>
<td><img src="image" alt="Autoscale [Y]" /></td>
<td>Autoscale [Y]</td>
<td>Click the Autoscale[Y] button to automatically scale collected data by scaling the ordinate (Y) height to full screen.</td>
</tr>
<tr>
<td><img src="image" alt="Zoom Out" /></td>
<td>Zoom Out</td>
<td>Click the Zoom Out button to Zoom back to the original X and Y ranges which were used to collect the data (i.e. the method setup conditions of the focused scan). For example, the system will revert back to the X and Y ranges set in the method if you zoom in and out during a data collection.</td>
</tr>
<tr>
<td><img src="image" alt="Add Label" /></td>
<td>Add Label</td>
<td>Click this button to display the Add Label dialog where you can enter a text label to be placed on a graph or in the background of the Graphics area.</td>
</tr>
<tr>
<td><img src="image" alt="Add Picture" /></td>
<td>Add Picture</td>
<td>Click this button to display the Add Picture dialog where you can select a picture, bitmap or chemical structure to be placed on a graph or in the background of the Graphics area.</td>
</tr>
<tr>
<td><img src="image" alt="Single/Multi Graph" /></td>
<td>Single/Multi Graph</td>
<td>Select Single/Multi Graph to toggle between displaying one or all graphs.</td>
</tr>
</tbody>
</table>
### Graph Labels—Concentration

- **Add Label dialog**
- **Edit Label dialog**
- **How to add a label to a graph**
- **How to edit a label**
- **How to move a label**
- **How to resize a label**
- **How to delete a label**

### Add Label—Concentration

Use the **Add Label** dialog to place a label on your graph or into the background of the Graphics area.
To add a label to a graph box, you must have at least one trace **Visible** in the graph box. (You can select which traces you want to make visible in the **Trace Preferences** dialog.) If you are viewing multiple graphs, the label will be placed in the graph box that is currently selected. However, you can add a label outside of a graph box in the background of the Graphics area at any time.

Q. Can I add a label that will print with all my graphs.

A. Yes, you can add your label to the background of the Graphics area. To do this, right-click on a section of the background (gray area) and select **Add Label** from the menu. (You will notice that this menu has less options than the menu that appears when you right-click on a graph.) In the **Add Label** dialog, enter the label to be added to all graphs and select **OK**. The label will appear in the background of the Graphics area.

To access the **Add Label** dialog, select the **Add Label** command from the **Graph** menu, or select the **Add Label** button on the Graphics toolbar.

The **Add Label** dialog comprises the following:

- **To selected trace**
  Select this option to associate a label with the selected trace. When you associate a label with a trace and make the trace visible in the Graph box, the label will also be displayed with the trace.

- **To selected graph**
  Select this option to associate the label with the selected graph. When you associate a label with a graph, the label will always be displayed in the Graph box, regardless of what traces are visible.

- **Selected trace**
  This will identify the trace that is currently selected and display the name of the trace.

- **Font**
  Click **Font** to open the Windows Font dialog. In this dialog you can specify the font type, style and size to be used for the label.

- **Label**
  Use the **Label** field to enter the text for the label. You can enter as many lines of text as you require.

**Hot Tip**

If you wish to include certain XY co-ordinates on the graph as a label, position your cursor at the appropriate place, right-click and select **Add Label** from the menu. When the **Add Label** dialog appears, the co-ordinate positions will be added to the **Label** field.

**Note**

When you save the data (as a Batch or Data file) the label is saved with its associated trace or graph. Labels outside graph boxes can be saved in a graphics template. To do this, choose **Graph Template** as the **Save As** type in the **Save As** dialog. See [Example of a Graphics Template](#).

See also:
- **Trace Preferences**
- **Graph menu**
- **Graphics toolbar**
- How to add a label to a graph
- How to move a label
- How to edit a label
- How to resize a label
- How to delete a label
Edit Label—Concentration

Use the Edit Label dialog to alter the content or look of your label.

To access the Edit Label dialog, right-click on the label you wish to change and select Edit Annotation from the menu.

The dialog comprises the following:

Font  Select Font to open the Font dialog, where you can specify the font type, style and size to be used for the label.

Label  Use the Label field to alter the text for the label.

See also:

- How to edit a label
- How to add a label to a graph
- How to resize a label
- How to move a label

How to add a label to a graph—Concentration

You can add a label to your graph or Graphics area background. To do this:

Step 1

Click on the graph box you require or click outside the graph box(es) to choose the Graphics area background.

Note

There must be at least one trace Visible in the graph box before you can add a label to it.

Step 2

Click the Add Label button on the toolbar or right-click on the graph and select Add Label from the menu. The Add Label dialog will appear.

The Add Label command is also accessible from the Graph menu.

Step 3

Click the Font button. The Windows Font dialog appears. Select the font type, size and style you require for your label. A sample of the font style appears in the Sample field. When you are satisfied with your selections, select OK to return to the Add Label dialog.

Step 4

Enter the label in the Label field and select OK. The label will be displayed on the selected graph. If necessary, you can move a label so that it is more appropriately placed on your graph.

See also:
How to edit a label—Concentration

Step 1

Place the cursor on the label and click the left mouse button to select the label.

**Hot Tip**

If you are having trouble selecting the label it may be that a trace is interfering with the label selection. Try zooming in and clicking in a different part of the label.

Step 2

Click the right mouse button and select *Edit Annotation* from the drop-down menu. Alternatively you can select the *Edit Annotation* command from the *Graph* menu. The *Edit Label* dialog is displayed.

Step 3

Use the *Edit Label* dialog to change the label.

For example, if you want to change the font size of the label, select the *Font* button. In the *Size* field, scroll through the available sizes for the font and select the size you require. Alternatively, highlight the currently selected size in the *Size* entry field and enter the size you require. Click the *OK* button in this dialog to return to the *Edit Label* dialog.

Step 4

Click *OK* to clear the *Edit Label* dialog. Your label will appear with the new font size.

See also:

How to add a label to a graph
How to move a label
How to delete a label

How to move a label—Concentration

To move a label so that it is more appropriately placed on your graph or Graphics area background, do the following:

Step 1

Place the cursor on the label.

Step 2

Click and hold down the left mouse button.
Step 3

Drag the label to the correct position and release the mouse button. The label will be displayed at the new position.

Note
Labels are transparent so as not to interfere with any traces on graphs.

See also:
- How to add a label to a graph
- How to edit a label
- How to resize a label
- How to delete a label

How to resize a label—Concentration

To resize a label so that it is more appropriately sized on your graph or Graphics area background, do the following:

Step 1

Select the label by clicking on it with the mouse. A border will be displayed around the label to indicate it is selected.

Hot Tip
If you are having trouble selecting the label, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Position the cursor over one of the corner edges of the label until the cursor becomes a diagonal double-headed arrow. Click and hold down the left mouse button.

Example of label showing the border that indicates it is selected and the arrow to be used for re-sizing

Step 3

Increase or decrease the size of the label by dragging the mouse. The label text will also be resized. Release the mouse button when the label and text are the correct size.

Note
Labels are transparent so as not to interfere with any traces on graphs.
How to delete a label—Concentration

To delete a label from a graph or Graphics area background, do the following:

Step 1

Position the cursor on the label and click the left mouse button to select it.

**Hot Tip**
If you are having trouble selecting the label it may be that a trace is interfering with the label selection. Try zooming in and clicking in a different part of the label.

Step 2

Click with the right mouse button and select **Delete Annotation** from the drop-down menu displayed. Alternatively, you can select the **Delete Annotation** command from the **Graph** menu. The label will be deleted from the graph or Graphics area.

Graph Pictures—Concentration

- Add Picture dialog
- Edit Picture dialog
- How to add a picture
- How to replace a picture
- How to move a picture
- How to resize a picture
- How to delete a picture

Add Picture—Concentration

Use the **Add Picture** dialog to insert a picture or bitmap into the Graphics area. The picture can be added as a background picture or to a particular graph box.

To add a picture to a graph box you must have at least one trace **Visible** in the graph box. (You can select which traces you want to make visible in the **Trace Preferences dialog**.) If you are viewing multiple graphs, the picture will be placed in the graph box that is currently selected. However, you can add a picture outside of a graph box in the Graphics area at any time.
Q. Can I add a picture that will print with all my graphs.
A. Yes, you can add your picture to the background of the Graphics area. To do this, right-click on a section of the background and select **Add Picture** from the menu. (You will notice that this menu has less options than the menu that appears when you right-click on a graph.) In the **Add Picture** dialog, load the picture to be added to all graphs and select OK. The picture will appear in the background of the Graphics area.

To access the **Add Picture** dialog, select the **Add Picture** command from the **Graph menu**, or select the **Add Picture** button on the **Graphics toolbar**.

The **Add Picture** dialog comprises the following commands:

- **To selected trace**
  - Select this option to associate a picture with the selected trace. When you associate a picture with a trace and make the trace visible in the Graph box, the label will also be displayed with the trace.

- **To selected graph**
  - Select this option to associate the picture with the selected graph. When you associate a picture with a graph, the label will always be displayed in the Graph box, regardless of what traces are visible.

- **Load**
  - Click **Load** to open the Windows **Open** dialog. Use this dialog to browse your computer for the picture you want to insert. The picture must be one of the following formats: DIB, JPG, PGEG, BMP, EMF or WMF.

If you are viewing multiple graphs the picture will be placed on the graph that is currently selected. If no graph is selected, the picture will be placed on the Graphics area background.

When you save the data (as a Batch or Data file) the picture is saved with its associated trace or graph. Pictures outside graph boxes can be saved in a graphics template. To do this, choose **Graph Template** as the **Save As** type in the **Save As** dialog. See Example of a Graphics Template.

See also:
- How to add a picture to a graph
- How to move a picture
- How to resize a picture
- How to delete a picture
- How to replace a picture

**Edit Picture—Concentration**

Use the **Edit Picture** dialog to change a picture you have inserted into the Graphics area.

To access the **Edit Picture** dialog, right-click on the picture you wish to change and select **Edit Annotation** from the menu.

The **Edit Picture** dialog comprises the following commands:

- **To selected trace/graph**
  - Displays whether you originally selected to add the picture to a trace or a graph. This option is not editable.

- **Load**
  - Click **Load** to open the Windows **Open** dialog. Use this dialog to browse your computer
for the picture you want to insert. The selected picture will replace the current picture. The picture must be one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.

See also:
How to replace a picture
How to add a picture
How to move a picture
How to resize a picture
How to delete a picture

How to add a picture to a graph—Concentration

You can add a picture to your graph or the Graphics area background. To do this:

Step 1
Click on the graph box you require or click outside the graph box(es) to choose the Graphics area background.

Note
There must be at least one trace Visible in the graph box before you can add a picture to it.

Step 2
Click the Add Picture button on the toolbar or right-click on the graph and select Add Picture from the menu. The Add Picture dialog will appear. The Add Picture command is also accessible from the Graph menu.

Step 3
Click the Load button. The Windows Open dialog appears.

Step 4
Browse through your directories to find the picture you want to insert. All pictures must be in the bitmap format and will have the extension .BMP.

Step 5
Highlight the .BMP that you want to insert and select Open.

Step 6
The Add Picture dialog will appear showing a preview of the bitmap. If the bitmap is the correct one select the OK button. The bitmap will be placed onto the graph.

Note
You should take care not to add a bitmap that is too big as it will obscure part or all of your graph.
You can move a picture so that it is more appropriately placed on your graph.

See also:
- How to move a picture to a graph
- How to delete a picture
- How to replace a picture
- How to resize a picture

**How to replace a picture—Concentration**

To change a picture that you have added to a graph or Graphics area background do the following:

**Step 1**

Move the cursor to the picture and select the left mouse button to select it.

**Hot Tip**
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

**Step 2**

Click the right mouse button and select **Edit Annotation** from the drop-down menu or select **Edit Annotation** from the **Graph** menu. The **Edit Picture** dialog is displayed.

**Step 3**

From the **Edit Picture** dialog, select the **Load** button. The Windows **Open** dialog is displayed.

**Step 4**

Browse through your directories to find the picture to replace the current bitmap. Picture files must be in one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.

**Step 5**

Highlight the file that you want to insert and select **Open** to return to the **Edit Picture** dialog.

**Step 6**

The **Edit Picture** dialog will display a preview of the bitmap. If the bitmap is the correct one select the **OK** button. The bitmap will be placed onto the graph. If you need to, move the picture so that it is more appropriately placed on your graph.

**Note**
- Do not to add a bitmap that is too big as it will obscure part or all of your graph.

See also:
- How to add a picture to a graph
- How to move a picture
How to move a picture—Concentration

To move a picture so that it is more appropriately placed on your graph or Graphics area background, do the following:

Step 1

Place the cursor on the picture.

Step 2

Click and hold down the left mouse button.

**Hot Tip**
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 3

Drag the picture to the correct position and release the mouse button. The picture will be displayed at the new position.

See also:
- How to add a picture to a graph
- How to delete a picture
- How to replace a picture
- How to resize a picture

How to resize a picture—Concentration

To resize a picture so that it is more appropriately sized on your graph or Graphics area background, do the following:

Step 1

Select the picture by clicking on it with the mouse. A border will be displayed around the picture to indicate it is selected.

**Hot Tip**
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Position the cursor over one of the corner edges of the picture until the cursor becomes a diagonal double-headed arrow. Click and hold down the left mouse button.
Example of picture showing the border that indicates it is selected and the arrow to be used for re-sizing

Step 3

Increase or decrease the size of the picture by dragging the mouse. The picture will also be resized. Release the mouse button when the picture is the correct size.

See also:
- How to add a picture to a graph
- How to move a picture to a graph
- How to delete a picture
- How to replace a picture

How to delete a picture—Concentration

To delete a picture from a graph or Graphics area background, do the following:

Step 1

Position the cursor on the picture and click the left mouse button to select it.

Hot Tip
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Click with the right mouse button and select Delete Annotation from the drop-down menu displayed. Alternatively, you can select the Delete Annotation command from the Graph menu. The picture will be deleted from the graph or Graphics area.

See also:
- How to add a picture to a graph
- How to move a picture
- How to zoom a graph
- How to resize a picture

Trace Preferences—Concentration

A single graph box can display many traces. Use the Trace Preferences dialog to select the trace(s) to display (make Visible) in the selected graph box.
To access the Trace Preferences dialog, select the Trace Preferences command from the Graph menu, or select the Trace Preferences button on the Graphics toolbar.

The dialog comprises the following:

**Hot Tip**
The Trace Preferences menu is also available on clicking the right mouse button anywhere in the trace listing.

**Trace Preferences table**

**Visible**
Use the Visible field to select which traces will appear on the selected graph. To select or clear the Visible field, simply click the left mouse button in this column. A tick will appear next to traces that are to be displayed in the selected graph.

If you are in single graph view, all selected traces will appear on the one graph. If you are in Multi Graph view the traces will appear on the graph that is currently selected.

**Hot Tip**
To make all traces Visible, right-click on the table and select the Select All command from the menu. Then right-click on the table again and select the Show Traces command from the menu.

**Color**
The Color field enables you to view the color of each trace that appears on the selected graph. To change the color of a trace, highlight it, click the right mouse button in the Trace Preferences table and select Change Trace Color. You can then select a color from the Color dialog that is displayed.

The red trace in any graph box is the one on which all information appears. This trace, which is used for the cursor tracking, is called the focused trace and it is always red. When you select a trace in the Color column it will become the focused trace in the selected graph box.

**Note**
When you change the trace line width in the Graph Preferences dialog it also will change the width of the color trace line in this dialog.

**Name of trace**
The Name of Trace field contains a description of the traces currently available for display in the selected graph box.

**Filename**
This field displays the filename and path for the trace. If the name is too long to appear in the field you can use the keyboard arrow keys to scroll to the end of the filename or you can resize the Trace Preferences window. If the file path for a trace shows 'not saved' then the trace is unsaved data and is only stored in memory.

**Trace Audit Log**
The Trace Audit Log displays information about the highlighted trace. This includes instrument parameters and data form information relating to the trace that has been selected. It also logs all information about data manipulation. When more than one trace is highlighted, the Trace Audit Log will display information about the trace that was first selected.
The Trace Preferences menu is displayed when you click the right mouse anywhere on the Trace Preferences table. The menu contains the following commands:

- **Select All**: This selects or highlights all the traces in the listed on the Trace Preferences page.
- **Select None**: This de-selects all the traces in the listing.
- **Show Traces**: Select **Show Traces** to make all of the traces currently highlighted *Visible*, meaning that they will be displayed in the current graph box.
- **Hide Traces**: Select **Hide Traces** to remove the *Visible* property from all the traces currently highlighted, which will remove them from the current graph box.
- **Delete Traces**: Select **Delete Traces** to delete all the highlighted traces in all graphs. Note that if you save the data file before you select **Delete Traces**, the traces are not deleted from the saved file, and they can be retrieved at a later date. Selecting **Delete Traces** will delete the traces from the application memory, and it is recommended that you select this option if you need to free up memory on your computer.

  **Note**

  If the application is controlled by the GLP Administration application, you will be prompted to save an unsaved data file before it is removed from the application.

- **Change Trace Color**: Select **Change Trace Color** to display the Windows *Color* dialog where you can change the color of the currently highlighted trace(s). The focused trace will remain red while it retains the focus, and will revert to the selected color when the focus is moved.

See also:
- Trace Preferences
- Graph Preferences
- How to make multiple traces visible in a graph
- How to remove a trace from the trace list
- How to remove a trace from a graph

**Trace Preferences menu—Concentration**

**How to select the focused trace—Concentration**

To select the scan that you want to be the focused (red) trace, do one the following:

- From the Graph, click the left mouse button on the trace that you want to make the focused trace.
- From the Graph Legend (if displayed), click the left mouse button on the trace that you want to make the focused trace.
- From the Trace Preferences dialog, click in the *Color* column of the trace that you want to make...
How to display a trace that is not appearing in the graph box—Concentration

Step 1
Click on the graph in the Graphics area.

Step 2
Select the Trace Preferences button from the Graphics toolbar, or select Trace Preferences from the Graph menu. The Trace Preferences dialog is displayed.

Step 3
Find that trace that you wish to see and select the Visible option. A tick will appear in the Visible column to indicate that it will now be displayed.

If you wish to make multiple traces visible, do the following:

a. Hold down the CTRL key select all the traces that you want to display.
b. Click the right mouse button and select the Show Traces command from the drop-down menu. Ticks will appear next to the selected traces to indicate that they will now be displayed.

Step 4
Press OK to clear the Trace Preferences dialog. The selected trace(s) will be displayed.

How to remove a trace from a graph—Concentration

Step 1
Select a graph in the Graphics area.

Step 2
Click the Trace Preferences button on the Graphics toolbar or select Trace Preferences from the

Graph menu. The Trace Preferences dialog is displayed.

Step 3

Find that trace that you no longer wish to see and select the Visible option. The tick will be removed from the Visible column to indicate that it will not be displayed.

If you wish to make multiple traces visible, do the following:

a. Hold down the CTRL key select all the traces that you no longer want to display.
b. Click the right mouse button and select the Hide Traces command from the drop-down menu. The ticks will be removed from the selected traces to indicate that they will not be displayed.

Step 4

Select OK to clear the Trace Preferences dialog. The trace will be removed from the graph.

See also:
How to display a trace that is not appearing in the graph box
How to remove a trace from the trace list

Graph Preferences—Concentration

Introduction

Use the Graph Preferences dialog to change the appearance of your graph.

All graphs will appear labeled with the X axis label being horizontal and the Y axis vertical. You cannot change the orientation of these labels. However you can alter the font and size of the axes and their labels.

Any changes you make to the appearance of the graph will apply to all graphs until the settings are changed again.

To access the Graph Preferences dialog, select the Graph Preferences command from the Graph menu, or select the Graph Preferences button on the Graphics toolbar.

Dialog controls

The Graph Preferences dialog comprises the following:

Axes Preferences

Axes Color Select the color that you want to assign to the axes of the graph/s from the drop down list.
Axes Width Select the width of the axes from the drop down list. The width, which is in pixels,
ranges between one and five pixels.

**Grid Style** Select the style of grid for the graph from the drop down list. If you do not wish to include a grid on the graph, select **None**.

**Plot Area Color** Select the color to be used as the background of the graph from the drop down list. If you do not want a background color on the graph, select **White**.

**Trace Width** Select the width of the trace from the drop down list. The width is given in pixels and ranges between one and five pixels.

**Font** Click the **Font** button to display the Windows **Font** dialog where you can define the font style for the axes of the graph/s. The style and size of the font will be applied to all graph. The style, name and point size of the font appears on the font button.

**Preview Box** Preview any changes you make to the axes preferences in the preview box. The changes shown in this box will not be made until you select **OK**.

### Advanced Options

**Auto tick marks** The **Auto tick marks** option enables you to approximately set how many tick marks (or points) will be included on the graph axes. The tick marks generated will be whole, rounded, even numbers. The **Auto tick marks** option is not available if you have selected **Major ticks**.

**Y-axis** Use the Y-axis slider bar to increase or decrease the number of tick marks (or points) to be included on the Y axis.

**X-axis** Use the X-axis slider bar to increase or decrease the number of tick marks (or points) to be included on the X axis.

**Major ticks** Use the **Major Ticks** option to enable you to set exactly how many ticks (or points) will be added to the X and Y axes. The number of marks you specify will be divided by the axis range, which can result in odd numbers on the axis. Note that any changes are not reflected in the graph bitmap at the top of the screen.

**Decimals** Use the **Decimals** option to set the number of X and Y decimal places to be displayed on the axes. To set the number of decimal places, select the **Decimal** option and enter the number in the Y-axis and X-axis combo boxes. Note that any changes are not reflected in the graph bitmap at the top of the screen.

### Trace Style

**Points** Select **Points** to display each individual data point collected by the Cary Eclipse on the graph.

**Points and solid lines** Select **Points and Solid Lines** to display each individual data point collected by the Cary Eclipse on the graph. Additionally, an interpolated line that connects the points will be drawn.
How to alter the look of your graphs—Concentration

Step 1

Click the Graph Preferences button on the Graphics toolbar, or select Graph Preferences from the Graph menu. The Graph Preferences dialog is displayed.

Step 2

Use the available options available to choose your preferred appearance for the graphs. A preview box in the top right-hand corner will demonstrate how the graphs will appear.

Step 3

Select OK to clear the Graph Preferences dialog and update all graphs to reflect your selections.

Note

You can also add labels and pictures to your graphs to alter their appearance.

Step 4

To re-use your graphics setup at a later date, save the setup as a graphics template.

See also: How to add a label to a graph
How to add a picture to a graph
How to move a label
How to move a picture
How to resize graph boxes
How to make a graphics template

Cursor Modes—Concentration

Use the **Cursor Modes** dialog to choose the type of cursor to be used on screen. For example, you can select to use a free cursor that can move anywhere in the graph.

To access the **Cursor Modes** dialog, select the **Cursor Modes** command from the **Graph menu**, or select the **Cursor Modes** button on the **Graphics toolbar**.

The dialog comprises the following:

**Cursor Selection**

**Free**

The **Free** cursor is not confined to a particular trace on the graph. It can move anywhere in a graph box, and its ‘X,Y’ position is shown in the bottom right-hand side of the Graphics area. You can use the **Free** cursor to zoom in on a particular part of a graph.

**Note**

The Free cursor moves from point to point in the selected graph box. Hence you need to be careful when positioning the cursor over a trace to obtain an accurate (X,Y) reading. It is recommended that you use the Track cursor mode for this.

**Track**

Use the **Track** cursor to exactly trace the focused (red) trace in a graph box. The cursor position is displayed in the X and Y at the bottom right-hand side of the Graphics area.

**Hot Tip**

If you wish to fix the cursor on a particular position on the curve, move the cursor to the required position. Click the left mouse button, press ‘Alt’ on the keyboard, then release the mouse button.

**Track Intensity**

Select the **Track Intensity** option if you want to associate an intensity value with the tracking. The cursor will then track from point to point on the continuum between the intensity value you enter. The default value will be 50% of the maximum intensity of the focused continuum.

**Peaks**

Choose from the options of **Peaks**, **Valleys** or **Peaks Valleys** to move the cursor from peak to peak, valley to valley or from one peak/valley to the next.

See also:

**How to zoom a graph**
**How to track the focused trace**

**How to track the focused trace—Concentration**

To track the focused (red) trace in the graph:

Step 1
Click the **Cursor Modes** button from the Graphics toolbar, or select **Cursor Mode** from the **Graph** menu. The **Cursor Modes** dialog will appear.

**Step 2**

Select **Track** and select the **OK** button.

**Step 3**

Place the blue tracking cursor in the graph box you require.

**Step 4**

Move the cursor left and right across the graph box to trace the focused (red) scan. The cursor coordinates change (in the X, Y field at the bottom left hand corner of the Graphics area) as you move along the curve.

**Hot Tip**

If you wish to fix the cursor on a particular position on the curve, move the cursor to the required position. Press **ALT** on the keyboard, then release the mouse button.

**See also:**

- How to select the focused trace
- How to view a particular trace

**Axes Scales—Concentration**

Use the **Axes Scale** dialog to manually change the axes range. This can assist in zooming in on a particular part of the graph.

To access the **Axes Scale** dialog, select the **Axes Scale** command from the **Graph menu**, or select the **Axes Scale** button on the **Graphics toolbar**.

The dialog comprises the following:

**X Axis**

| **Mode** | **View the current units for the X axis.** |
| **Scale** | **Use the Scale field to enter a new range for the X axis, in the format n1 - n2 (where n1 is the start of the scale and n2 is the end of the scale).** 

**Hot Tip**

The Cary Eclipse will store the ten most recently entered ranges, which you can access by selecting the down arrow adjacent to the Scale field.

**Y Axis**
How to scale your graph manually—Concentration

Step 1

Click on the graph whose axes you want to change to make it the focused trace.

Step 2

Click the Axes Scales button  on the toolbar, or select Axes Scales from the Graph menu. The Axes Scales dialog is displayed.

Step 3

Type in the new ranges for the X and Y axes in the format ‘n-n’, or choose an option for each from the drop-down list.

Step 4

Select OK. The axes on the selected graph will change to reflect your chosen settings.

How to scale your graph automatically—Concentration

Step 1

Click on the graph whose axes you want to change to make it the focused trace.

Step 2

To autoscale the graph, select one of the following:

- View the current units for the Y axis.
- Use the Scale field to enter a new range for the Y axis, in the format n1 - n2 (where n1 is the start of the scale and n2 is the end of the scale).

**Hot Tip**

The Cary Eclipse will store the ten most recently entered ranges, which you can access by selecting the down arrow adjacent to the Scale field.

See also:

- How to scale your graph manually
- How to scale your graph automatically
- How to zoom a graph

See also:

- How to zoom a graph
- How to scale a graph automatically
Click the **Autoscale XY** button on the toolbar to autoscale both X and Y axes.

Click the **Autoscale X** button on the toolbar to autoscale the X axis only.

Click the **Autoscale Y** button on the toolbar to autoscale the Y axis only.

The focused trace will be scaled according to the option selected.

**Hot Tip**
Select the **Zoom Out** command to resize the graph to its original settings.

See also:
- [How to zoom a graph](#)
- [How to scale your graph manually](#)

**Single/Multi Graph—Concentration**

Select the **Single/Multi Graph** option to toggle between a display of either one or multiple graph boxes in the Graphics area. Note that up to 100 graphs can be displayed in the Graphics area at the one time.

The **Single/Multi Graph** option is accessible from the **Graph** menu or the **Graphics** toolbar.

Example of a single graph box display

Example of a multi graph box display
Example of a Graphics Template—Concentration

By using the **Add Label** command in the Cary Eclipse graphics system, you can customize a graphics template such as the one shown below:

The above is created as follows:

- The graph on the left is the collected data.
- The text on the top right is a multi-line label (added using the **Add Label** command).
- The logo is an inserted picture (added using the **Add Picture** command).

How to make a graphics template—Concentration

A graphics template lets you set up and store the style of a graphics report for future use. You can do this by adding labels and pictures to the background of the Graphics area and altering any axes styles etc. One advantage of this over the application's built-in reporting features is that you have more flexibility as to where you can position graph boxes and any text that you may want to appear (e.g. operator name, batch number etc.) and also where you add pictures, e.g. company logos etc.

Step 1

Start the application and open the data file from which you want to make a graphics template.
Alternatively, collect some new data.

Step 2

Make as many graph boxes as you require in your template by using the Add Graph function.

Step 3

Adjust the graph boxes to the correct size and position them as you want them to appear in the template.

Step 4

Do any of the following, if required:

- Change the appearance of the graph boxes in the Graph Preferences dialog
- Use the Add Label function to append text to the Graphics area
- Use the Add Picture function to add pictures, such as your company's logo to the Graphics area

Step 5

From the File menu, select Print Preview. Use the Print Preview dialog displayed to check the template that you have created.

Step 6

When you are satisfied with the appearance of the graphics, select the Save As… command from the File menu. The Save As dialog is displayed.

Step 7

A list of stored data files will appear. Select the down arrow at the right of the Files of type field and select 'Graph Template' to list all the template files. Ensure that the Save only focused trace check box is cleared.

Step 8

Enter a name for the template in the File name field.

Step 9

Select OK to save the graphic template. The template will be used each time you create a new file.

Note
You can also make a graphic template in a similar manner from existing data. No data will be saved in the template.

See also:
- Example of a Graphics Template
- How to add a label to a graph
- How to add a picture to a graph
How to move a label
How to move a picture
File Management—Concentration

- File menu
- Open dialog
- Save As dialog
- Filename extensions
- ASCII spreadsheet file
- Auto Store page
- How to export collected data

File menu—Concentration

Use the File menu to open or save a method or data file, print or preview a report, or exit the Cary Eclipse application.

**Hot Tip**

Highlight a method or data file in Explorer and simply drag and drop it onto the application to open it. Any data will be displayed in the Graphics area.

The following commands can be accessed from the File menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>New</td>
<td>Select New to clear all method information and report information and create a new file containing the default settings for the application. The Save As dialog will be displayed for you to enter the name of the new file and select a location.</td>
</tr>
<tr>
<td>Open Method…</td>
<td>Select Open Method to display the Open dialog where you can search for and load a stored method.</td>
</tr>
<tr>
<td>Open…</td>
<td>Select Open to display the Open dialog where you can search for and load method, data, report, graph template and batch files, as well as other file types compatible with the Cary Eclipse software. When you load a batch file, the method which collected the data is loaded as well as the associated report and graphics layout.</td>
</tr>
<tr>
<td>Save Method As…</td>
<td>Select this option to display the Save As dialog and store the current method. You can use the Save As dialog to store the method, data, report, or graphics template separately or together as a Batch file. The data can also be saved as a comma delimited ASCII Spreadsheet file.</td>
</tr>
<tr>
<td>Save As…</td>
<td>Select this option to display the Save As dialog and store the collected data. You can use the Save As dialog to store the method, data, report, or graphics template separately or together as a Batch file. The data can also be saved as a comma delimited ASCII file.</td>
</tr>
<tr>
<td>Print Setup…</td>
<td>Select this option to display the Windows Print Setup dialog where you can select the printer, printer page size and orientation.</td>
</tr>
<tr>
<td>Print Preview…</td>
<td>Select this option to display the Print Preview dialog where you can view the current report as it will be printed.</td>
</tr>
<tr>
<td>Print…</td>
<td>Select this option to display the Windows Print dialog where you can choose the printer and pages to print as well as the number of copies you wish to print.</td>
</tr>
<tr>
<td>Explorer…</td>
<td>Select this option to run Windows Explorer. This may help you locate method and data</td>
</tr>
</tbody>
</table>
Open—Concentration

Use this dialog to open a file previously saved by the application. To access the Open dialog, select Open or Open Method from the File menu.

| Look in | The Look In field lists the available folders and files. Click the down arrow on the right of this field to view the location of the current folder in your computer's hierarchy. You can also select a new location simply by selecting a different folder. The folders and files in the selected folder are displayed in the list box below the Look In field. | Click to open the folder in the level that is one higher than the current folder, select on the toolbar. |
| Look in list box | The Look In List Box, which is directly below the Look In field, displays the folders and files in the selected location. To view the contents of a folder, double-click it. | Click to create a new folder. Type the name of the new folder in the highlighted text entry field that appears and press Enter. Then double-click on the new folder to open it. |
| File name | Enter the name of the file you want to open in this field. The easiest way to do this is to find the file in the Look in list box and then select the file. However, you can type the full path of a file and then select Open to open the file. | |
| Files of type | Click on the arrow on the right of this field to view a list of the different types of application files that can be opened in the list box. | Methods (*,FMSW) Choose this file type to open a Method file. Only the method is loaded. No data is loaded. |

See also:
- Save As dialog
- Open dialog
- Print Preview dialog
- Filename Extensions
- ASCII Spreadsheet File
### Save As—Concentration

Use the **Save As** dialog to save the data, method, report or graphics template individually or together as a Batch file.

**Note**
This dialog appears when you select either the **Save method as..** or **Save as...** menu option from the **File** menu. When you select **Save method as..** this dialog appears with 'Methods' displayed in the **Files of type field**. When you select **Save as...** this dialog appears with 'Batch' displayed in the **Files of type field**.

Traces (or data) that are currently **Visible** in the Graphics area are saved, regardless of how they are

<table>
<thead>
<tr>
<th>File Type</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Data (*.FDSW)</strong></td>
<td>Choose this file type to open a Data file. Only the data is loaded. The method that was used to collect the data is not loaded. The last used method or default method will be displayed.</td>
</tr>
<tr>
<td><strong>Report (*.FRSW)</strong></td>
<td>Choose this file type to open a Report file. You can view the report in the Report area.</td>
</tr>
<tr>
<td><strong>Graph Template (*.FGSW)</strong></td>
<td>Choose this file to open a Graph Template file. The file will set up the layout of the Graphics area including any bitmaps as well as the range and style of the graph axes.</td>
</tr>
<tr>
<td><strong>Batch (*.FBSW)</strong></td>
<td>Choose this file type to open a Batch file. A Batch file includes data, the method used to collect the data, report and graphics setup.</td>
</tr>
<tr>
<td><strong>Spreadsheet Ascii (*.CSV)</strong></td>
<td>This will open ASCII files that contain single comma delimited files with two or more columns.</td>
</tr>
<tr>
<td><strong>ADL (*.ADL)</strong></td>
<td>This will open and run program files generated in the Applications Development Language (ADL).</td>
</tr>
<tr>
<td><strong>All files (<em>.</em>)</strong></td>
<td>This enables you to view all files in the directory so that you can open any compatible file.</td>
</tr>
</tbody>
</table>

Tell me more about Filename Extensions

- **Overlay Data**
  Select this check box to overlay individual data files in one graph box. If this is not checked the data from each file will appear in individual graph boxes. This option is only available if the **Open** dialog has been accessed by selecting **Open...** from the **File** menu.

  **Hot Tip**
  Use the SHIFT or CTRL key to select multiple data files.

- **Open**
  This button opens the highlighted file/s or the files listed in the **File name** field.

See also:
- **File menu**
- **ASCII Spreadsheet File**

### Save As—Concentration

Use the **Save As** dialog to save the data, method, report or graphics template individually or together as a Batch file.

**Note**
This dialog appears when you select either the **Save method as..** or **Save as...** menu option from the **File** menu. When you select **Save method as..** this dialog appears with 'Methods' displayed in the **Files of type field**. When you select **Save as...** this dialog appears with 'Batch' displayed in the **Files of type field**.

Traces (or data) that are currently **Visible** in the Graphics area are saved, regardless of how they are
displayed (full screen graph box or tile). For example, if six graph boxes are displayed in the Graphics area and each graph box has one visible trace then all six traces will be saved.

If you do not wish to save particular traces, make them not Visible in the Trace Preferences dialog. To save only the focused trace, select the Save only focused trace check box.

The dialog comprises the following:

**Look In** The Look In field lists the available folders and files. Click the down arrow on the right of this field to view the location of the current folder in your computer's hierarchy. You can also select a new location simply by selecting a different folder. The folders and files in the selected folder are displayed the list box below the Look In field.

- Click to open the folder in the level that is one higher than the current folder, click on the toolbar.

**Look in list box** The Look In List Box, which is directly below the Look In field, displays the folders and files in the selected location. To view the contents of a folder, double-click it.

- Click to create a new folder. Type the name of the new folder in the highlighted text entry field that appears and press Enter. Then double-click on the new folder to open it.

- Click to display the directory structure of the drive.

- Click to display the files as a list.

- Click to display files as a detailed list including the file size, file type and the date the file was last modified.

- Click to preview the selected trace before you load it into the application.

**File Name** Enter the name of the file you want to save in this field. You can also type the full path of a file then select the Save button to save the file.

**Files of Type** Click on the arrow on the right of this field to view a list of the different types of application files that can be opened in the list box.

- **Methods (/*.FMSW)** Choose this file type to only save the current method. Any collected data is not stored.

- **Data (/*.FDSW)** Choose this file type to only save the data currently Visible in the Trace Preferences dialog (for any graph box). Note that the method used to collect the data is not stored with the data.

- **Report (/*.FRSW)** Choose this file type to only save the contents currently shown in the Report area.

- **Graph Template (/*.FGSW)** Choose this file type to save the layout of the Graphics area as a template. For example, any bitmaps you have added to your Graphics area will be saved as well as the range and style of the graph axes.

- **Batch (/*.FBSW)** Choose this file type to save the data that is currently Visible in the Trace Preferences dialog (for any graph box), the method
used to collect the data and the currently displayed report all together as a Batch file. The graphics setup is also stored.

**Spreadsheet Ascii (\*.

Choose this file type to save the raw intensity data currently Visible in the Trace Preferences dialog (for any graph box) as a comma delimited ASCII Spreadsheet file (\*.

**Note**

This file type can be easily imported into a spreadsheet program. For example, Microsoft Excel. You can also double-click on the stored *.

**GRAMS (\*.

Choose this file type to only save the data currently Visible in the Trace Preferences dialog (for any graph box) as a Cary Eclipse GRAMS data file.

Tell me more about Filename Extensions

**Save**

Click this button to save the file currently present in the File name field in the directory specified in the Look in field.

See also:

File menu

Trace Preferences dialog

ASCII spreadsheet files

### Filename extensions—Concentration

The Cary Eclipse system uses the filename extension to distinguish between data types (e.g. method or report) and also to differentiate applications. (e.g. Concentration or Kinetics).

The Syntax for file extensions is:

<table>
<thead>
<tr>
<th>Letter</th>
<th>Represents</th>
<th>Available Options</th>
</tr>
</thead>
<tbody>
<tr>
<td>First</td>
<td>Type of analysis</td>
<td>F Fluorescence</td>
</tr>
<tr>
<td>Second</td>
<td>Type of file</td>
<td>B Batch</td>
</tr>
<tr>
<td></td>
<td></td>
<td>D Data</td>
</tr>
<tr>
<td></td>
<td></td>
<td>G Graph Template</td>
</tr>
<tr>
<td></td>
<td></td>
<td>M Method</td>
</tr>
<tr>
<td></td>
<td></td>
<td>R Report</td>
</tr>
<tr>
<td>Third and fourth</td>
<td>Application identifier</td>
<td>AB Advanced Reads</td>
</tr>
<tr>
<td></td>
<td></td>
<td>CN Concentration</td>
</tr>
<tr>
<td></td>
<td></td>
<td>KN Kinetics</td>
</tr>
<tr>
<td></td>
<td></td>
<td>LT Lifetimes</td>
</tr>
<tr>
<td></td>
<td></td>
<td>RT Ratio</td>
</tr>
<tr>
<td></td>
<td></td>
<td>SR Simple Reads</td>
</tr>
</tbody>
</table>
The following files may be opened and saved in the Concentration Application:

**Note**
Selecting *All Files* in the Windows Open dialog will list all available file types.

<table>
<thead>
<tr>
<th>Extension</th>
<th>Available action</th>
<th>File type</th>
</tr>
</thead>
<tbody>
<tr>
<td>*.FMCN</td>
<td>Open/Save</td>
<td>A <em>method</em> file (*.FMSW) consists of the software setup by which data is collected.</td>
</tr>
<tr>
<td>*.FDCN</td>
<td>Open/Save</td>
<td>A <em>data</em> file (*.FDSW) contains data collected that is currently visible in the Trace Preferences dialog (for any graph box).</td>
</tr>
<tr>
<td>*.FRCN</td>
<td>Open/Save</td>
<td>A <em>report</em> file (*.FRSW) contains the report generated by the Cary Eclipse about the data.</td>
</tr>
<tr>
<td>*.FGCN</td>
<td>Open/Save</td>
<td>A <em>graph template</em> file (*.FGSW) contains information that sets up the layout of the Graphics area including any bitmaps as well as the range and style of the graph axes.</td>
</tr>
<tr>
<td>*.FBCN</td>
<td>Open/Save</td>
<td>A <em>batch file</em> (*.FBSW) contains the collected data that was Visible in the Trace Preferences dialog (for any graph box), the method by which the data was collected and the report and graphics template.</td>
</tr>
<tr>
<td>*.CSV</td>
<td>Open/Save</td>
<td>An <em>ASCII spreadsheet</em> file (*.CSV) has the same extension for all applications. It consists of raw intensity data (that was currently Visible in the Trace Preferences dialog (for any graph box) saved as XY comma-delimited pairs. This file can be imported into a spreadsheet program such as Microsoft Excel.</td>
</tr>
<tr>
<td>Data.*</td>
<td>Open</td>
<td>A <em>Cary DOS</em> (Data.*) file is the equivalent of a Cary Windows Data file—it does not contain any Batch information.</td>
</tr>
<tr>
<td>*.ADL</td>
<td>Open</td>
<td>An <em>ADL</em> (*.ADL) file is a program file generated in the Applications Development Language (ADL).</td>
</tr>
<tr>
<td>*.SPC</td>
<td>Open/Save</td>
<td>A <em>Cary GRAMS</em> file (*.SPC ) is the equivalent of a Cary Eclipse Windows Data file—it does not contain any Batch information.</td>
</tr>
<tr>
<td>*.FADL</td>
<td>Open</td>
<td>An <em>Eclipse ADL</em> (*.FADL) file is a program file generated in the Applications Development Language (ADL) specifically for the Cary Eclipse. Eclipse ADL files will be associated with the relevant Eclipse application. When creating ADL files for the Cary Eclipse, it is recommended that you save them with the...</td>
</tr>
</tbody>
</table>
ASCII Spreadsheet file (*.CSV) — Concentration

An ASCII Spreadsheet file consists of raw intensity data saved as XY comma-delimited pairs. You can choose to import or export Cary Eclipse files to ASCII files, which can then be imported in spreadsheet programs such as Microsoft Excel.

The format is as follows:

- Line 1 contains the sample name plus a comma
- Line 2 contains X and Y labels. The name of the X label must be the same as that used to label the graph. For example, in Kinetics, the graph label is Concentration (pM).
- Lines 3 and onward contain the X,Y data
- After the last line of X,Y pairs there must be a blank line separating the data from any text audit strings or the end of the file.

For example, if a concentration file was collected as:

<table>
<thead>
<tr>
<th>pM</th>
<th>intensity</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0083</td>
<td>0.1392</td>
</tr>
<tr>
<td>0.01667</td>
<td>0.1388</td>
</tr>
<tr>
<td>0.02450</td>
<td>0.1245</td>
</tr>
</tbody>
</table>

It would appear as follows in the ASCII file:

```
Concentration,
Concentration (pM); Intensity (a.u.)
0.0083, 0.1392,
0.01667, 0.1388,
0.02450, 0.1245,
Concentration,
```

Microsoft’s Excel generates a CSV file with the above format.

There is also an alternative format for CSV files. For example,

```
Concentration
Concentration (pM); Intensity (a.u.)
0.0083, 0.1392
0.01667, 0.1388
0.02450, 0.1245
Concentration,
```
Use the Auto store page to specify if and when data will be stored, and in what format. You can also use this page to set up the system to automatically send an email at the end of a read.

From the Setup menu, select Auto store to access the Auto store page. You can also access the page from the Setup dialog by selecting the Auto store tab. To access the Setup dialog, select the Setup button from the Concentration Application window.

The page comprises the following:

**Storage**

- **Off**
  - Select this if you do not wish to be automatically prompted to save collected data.
  - **Note**
    - If you wish to store your unsaved data manually, choose Save Data As… from the File Menu to save your data after the run is complete. You can save your data as a Batch or Data file.

- **On; Prompt At Start**
  - Select this to display the Windows Save As dialog at the start of the collect where you can enter the Batch filename for your data.

- **On; Prompt At End**
  - Select this to display the Windows Save As dialog at the end of a collect where you can enter the Batch filename for your data.

**Auto convert**

Use the Auto convert options to convert your data into an ASCII file format that can be opened in spreadsheet programs such as Microsoft Excel.

- **None**
  - Select None if you do not need to generate an ASCII file.

- **ASCII (csv)**
  - Select this option to save the data as a comma delimited ASCII Spreadsheet file (*.CSV) file. Only the currently collected data is converted.

  ASCII files are stored in the same folder as the data file from which they were generated and designated the same filename as the method used to collect the data plus the extension ‘CSV’.

- **ASCII (csv) with log**
  - Select this option to save the data as a comma delimited ASCII Spreadsheet file (*.CSV) file. Only the currently collected data is converted. The key method parameters used to collect the data are also saved in this file.
ASCII files are stored in the same folder as the data file from which they were generated and designated the same filename as the method used to collect the data plus the extension ‘CSV’.

**Note**
If you choose to auto-convert the data to an ASCII file but you have set the Storage option to Off, then the ASCII file will be saved with the following format:
DATE TIME. CSV

The date and time will appear in the same form that is set in the Windows Regional Settings Properties dialog that is accessed from the Windows Control Panel.

For example, 22_4_97 1;32;08.CSV

tell me more about ASCII Spreadsheet Files

**Email**
Select the Email check box and enter the Email details to send an email to a person once the read is completed. This useful facility can be used to alert you to the completion of the read, particularly if you are working away from the laboratory where the Cary Eclipse instrument is. You can also attach the collected data to the email.

**Note**
You must set up the email Host and Port options on the Email options page of the System Information application in order for the email facility to work correctly.

- **To** Enter the recipient's email address. You can enter more than one recipient in this field.
- **From** Enter your email address (or the email address associated with the computer on which the Cary Eclipse software is installed).
- **Subject** Enter the subject of your email (or leave this field blank if you do not wish to include a subject).
- **Cc** Enter the email address of any recipient you want to copy the email to.
- **Message** Enter your message.
- **Attached…** Use the Attached button include a file attachment in your email. To do this, select Attached and browse your computer to select a file, using the Windows® Open dialog that appears.
- **Attach current data** Select this option to attach the data that has been collected during the run to the email.

see more about ASCII Spreadsheet Files

**Status display**
Select this check box to display the Status Display window, where you can view the status of your instrument during the data collection.
How to export collected data—Concentration

If you want to export data from a Cary Eclipse application into a third-party application, then you simply need to save your data as an Spreadsheet ASCII (*.CSV) file or a Rich Text Format (*.RTF) file. Spreadsheet ASCII files are recommended if you are planning to use the data in a spreadsheet program such as Microsoft® Excel. Rich Text Format files are recommended if you are planning to open the data in a word processor program such as Microsoft® Word.

To export collected data:

Step 1

From the File menu, select Save As. The Save As dialog is displayed.

Step 2

Click on the down arrow next to the Files of type list box and select the Spreadsheet ASCII (*.CSV) or Rich Text Format (*.RTF) option.

Step 4

Type the name of your new file in the File name field.

Step 5

Select Save to export your data to this new file.
Software and Screen features—Concentration

- Status display
- Menu line
- Status line
- Buttons
- Function keys
- Right mouse menu
- How to remove or display the command buttons

Menu line—Concentration

- File menu
- Edit menu
- View menu
- Commands menu
- Setup menu
- Graph menu
- Help menu

Status line—Concentration

The Status Line is displayed at the very bottom of the Concentration Application window. The line will display the following information (from left to right):

- **Hardware Status**: This field displays status messages indicating the current instrument activity.
- **Sequence Information**: This field displays status messages indicating the current system activity.
- **Current Results**: This field displays information on the last reading.

Buttons—Concentration

The major commands in this application can be accessed both from the menu items at the top of the main window and from the application buttons. These buttons, which appear in the left side of the Concentration Application window, comprise the following:

**Hot Tip**

If you wish to access these commands from the main menus, rather than from these buttons, you can remove the Command buttons from your application window. To do this, clear the **Buttons** command in the **View** menu. This will enlarge the Graphics area, making viewing and selection of scans easier.

<table>
<thead>
<tr>
<th>Button</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Setup</td>
<td>Click this button to display the <strong>Setup</strong> dialog where you can set up the collection, storage and analysis parameters for the concentration run. You can also set up any accessories that you may have and define the reporting options.</td>
</tr>
<tr>
<td>Zero</td>
<td>Select this to zero the current ordinate value.</td>
</tr>
</tbody>
</table>
The function keys at the top of your keyboard perform basic Cary Eclipse operations simply and quickly.

<table>
<thead>
<tr>
<th>Function key</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>F1</strong></td>
<td>This displays the Help for the current dialog or window.</td>
</tr>
<tr>
<td><strong>F4</strong></td>
<td>Press F4 to display the <strong>Go To Wavelength</strong> dialog.</td>
</tr>
<tr>
<td><strong>F5</strong></td>
<td>Press F5 to perform an instrument Zero. The current intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the <strong>Zero</strong> command before selecting <strong>Start</strong> if you want your data to start at 0.</td>
</tr>
</tbody>
</table>
The following commands are accessible by clicking the right mouse button on editable fields in the Cary Eclipse software.

<table>
<thead>
<tr>
<th>Command</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Undo</strong></td>
<td>If available, this reverts to the previous value before the Edit on the field.</td>
</tr>
<tr>
<td><strong>Cut</strong></td>
<td>If available, this cuts the highlighted text to the clipboard.</td>
</tr>
<tr>
<td><strong>Copy</strong></td>
<td>If available, this copies the highlighted text to the clipboard.</td>
</tr>
<tr>
<td><strong>Paste</strong></td>
<td>If available, this pastes the text from the clipboard to the cursor position.</td>
</tr>
<tr>
<td><strong>Delete</strong></td>
<td>If available, this deletes the highlighted text/number.</td>
</tr>
<tr>
<td><strong>Select All</strong></td>
<td>If available, this selects all entries.</td>
</tr>
</tbody>
</table>

**Note**

Before you select **Zero**, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.

Select the **Clear Zero** command from the **Commands** menu to remove zero correction.

**F6**
Press F6 to display the **Rapid Result** dialog where you can read a blank and multiple sample readings without changing the application method. This command is also accessible from the **Commands** menu.

**F9**
If the application is online, press F9 to start a data collection. This is equivalent to selecting the **Start** button. If the application is not online, you will need to press F9 to first connect to the Cary Eclipse. Then press F9 to start the data collection. This command is also accessible from the **Commands** menu or by selecting the **Start** button.

**F12**
Select this to stop the current measurement. This is equivalent to selecting the **Stop** button. This command is also accessible from the **Commands** menu or by selecting the **Stop** button.

---

**Right mouse menu—Concentration**

The following menu commands are accessible by right clicking on the readable fields in the Cary Eclipse software.

See also:

- **Go To Wavelength**
- **Zero**
- **Rapid Result**
- **Start/Stop Instrument Buttons**
ADL—Concentration

- ADL Command Line
- ADL Hooks
- How to insert ADL commands

ADL Command Line—Concentration

Introduction to ADL

Use the ADL Command Line to run Single-line or Multi-line Applications Development Language (ADL) commands that enable you to perform functions over and above that of the existing program functionality.

ADL is not a general purpose programming language. It has been developed specifically by Varian to meet the needs of spectroscopists. It can be used to handle all the data collected from the Cary Eclipse—both photometric values and entire data collections. ADL programs that you develop should be stored with the extension 'ADL'.

To access the ADL Command Line dialog, select the ADL Command item from the Commands menu. You can also press ALT+Y to open the dialog.

**Note**

Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.

Software controls

**Single Line**

Selecting this lets you enter ADL commands on a single line into the Command Line entry box.

Q. Can I enter more than one ADL command on a single line?
A. Yes, provided that the commands are separated by a semicolon ‘;’.

**Multi-line**

The Multi-line capability enlarges the Command Line entry box. Use the Multi-line capability to create a more complex ADL program by adding a series of commands over multiple lines. When Multi-line mode is selected, the Quick Reference option becomes available.

**Quick Reference**

Select Quick Reference to open the Quick Reference dialog where you can easily add a command in the correct syntax to your ADL program. This option is only available for Multi-line commands.

Show me How to insert ADL commands using the Quick Reference.

A detailed description of all the commands in the Quick Reference is available by
The ADL Hooks command is accessible from the View menu.

Instead of writing a full ADL program you can use ADL hooks to link into an existing application. This is particularly useful if you want to make small changes to the way an application works, e.g. you want to pop up a dialog for the user to enter some specific data before the run starts. ADL hooks are links at specific points in an application at which you can run an ADL program. For example, ADL hooks have been supplied to run the following ADL programs:

<table>
<thead>
<tr>
<th>Hook</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>CN_Startup.ADL</td>
<td>This is run when the Concentration application starts. For this to work the application must be started from a batch or method file that has ADL Hooks on.</td>
</tr>
<tr>
<td>CN_PreSeq.ADL</td>
<td>This runs when the Start button is selected.</td>
</tr>
<tr>
<td>CN_PostSeq.ADL</td>
<td>This runs at the end of the Start button sequence.</td>
</tr>
<tr>
<td>CN_Online.ADL</td>
<td>This runs when Cary Eclipse is detected online.</td>
</tr>
</tbody>
</table>

Note
Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.
<table>
<thead>
<tr>
<th>ADL Name</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>CN_OffLine.ADL</td>
<td>This runs when Cary Eclipse goes offline.</td>
</tr>
<tr>
<td>CN_Zero.ADL</td>
<td>This runs when the Zero button is selected.</td>
</tr>
<tr>
<td>CN_Recalc.ADL</td>
<td>This runs when the Recalculate button is selected.</td>
</tr>
<tr>
<td>CN_Shutdown.ADL</td>
<td>This runs when the application is shutting down.</td>
</tr>
<tr>
<td>CN_PreRead.ADL</td>
<td>This runs just before a read is taken.</td>
</tr>
<tr>
<td>CN_PostRead.ADL</td>
<td>This runs just after a read is taken (It has the reading result stored in the UserResult variable.)</td>
</tr>
<tr>
<td>CN_PreSetup.ADL</td>
<td>This runs when the Setup button is selected.</td>
</tr>
<tr>
<td>CN_PostSetup.ADL</td>
<td>This runs when the OK button on the Setup dialog is selected.</td>
</tr>
<tr>
<td>CN_CellChange.ADL</td>
<td>This runs instead of the Cary Eclipse driving the Multicell Holder. It enables you to add your own instructions for the Cary Eclipse Multicell Holder or to use your own cell changing accessory.</td>
</tr>
<tr>
<td>CN_UserStartReport.ADL</td>
<td>This runs when the report is started. It is used to replace the normal report headings with customized headings.</td>
</tr>
<tr>
<td>CN_UserReportParams.ADL</td>
<td>This runs when the report parameters have been printed. It is used to add any extra user parameters to the parameter list.</td>
</tr>
<tr>
<td>CN_Userload.ADL</td>
<td>This runs when a sample is to be loaded. It is used to replace the normal sample loading procedures with a customized procedure.</td>
</tr>
<tr>
<td>CN_UserReportResults.ADL</td>
<td>This runs when results are to be reported. It is used to replace the normal report line for each sample with a customized one. This hook can be used if there is no difference in how standards and samples are reported. You can also use the ReportStandard and ReportSample hooks should be used.</td>
</tr>
<tr>
<td>CN_PreCalcStd.ADL</td>
<td>This runs prior to the standard calculations. It is used for any pre-treatment of the data prior to the normal calculations.</td>
</tr>
<tr>
<td>CN_ReportStandard.ADL</td>
<td>This runs when a standard is to be reported. It is used to replace the normal report line for each standard with a customized one.</td>
</tr>
<tr>
<td>CN_ReportSample.ADL</td>
<td>This runs when a sample is to be reported. It is used to replace the normal report line for each sample with a customized one.</td>
</tr>
<tr>
<td>CN_PreStdSeq.ADL</td>
<td>This runs just before the standards are measured. It is used for preliminary procedures for the standards.</td>
</tr>
<tr>
<td>CN_StartStandardsReport.ADL</td>
<td>This runs when the standards report is to be started. It is used to replace the normal report columns for the standards with customized columns.</td>
</tr>
<tr>
<td>CN_PostCal.ADL</td>
<td>This runs after the calibration of a sample is requested. It is used to perform any extra post calibration operations.</td>
</tr>
</tbody>
</table>
The following database nodes can be used to pass data between ADL and the Concentration application.

<table>
<thead>
<tr>
<th>Database Nodes</th>
<th>Data Type Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>'Current Solution Description';</td>
<td>String containing the solution name</td>
</tr>
<tr>
<td>'Current solution type';</td>
<td>Can have the following values:</td>
</tr>
<tr>
<td></td>
<td>CNC_STANDARD = 0;</td>
</tr>
<tr>
<td></td>
<td>CNC_SAMPLE = 1;</td>
</tr>
<tr>
<td></td>
<td>CNC_BLANK = 2;</td>
</tr>
<tr>
<td>'Current solution num';</td>
<td>Integer</td>
</tr>
<tr>
<td>'Current conc valid';</td>
<td>Boolean – used to determine if values are valid.</td>
</tr>
<tr>
<td>'Current mean valid';</td>
<td>Boolean – used to determine if values are valid.</td>
</tr>
<tr>
<td>'Current mean';</td>
<td>Real</td>
</tr>
<tr>
<td>'Current SD valid';</td>
<td>Boolean – used to determine if values are valid.</td>
</tr>
<tr>
<td>'Current SD';</td>
<td>Real</td>
</tr>
<tr>
<td>'Current RSD valid';</td>
<td>Boolean – used to determine if values are valid.</td>
</tr>
<tr>
<td>'Current RSD';</td>
<td>Real</td>
</tr>
<tr>
<td>'Current replicate num';</td>
<td>Integer</td>
</tr>
</tbody>
</table>
How to insert ADL commands using the Quick Reference—Concentration

The Quick Reference window is accessed from the ADL Command window. It enables you to easily add a command in the correct syntax to your ADL program. To do this:

Step 1

Press Alt-Y to open the ADL Command Line window.

Step 2

Select the Multi-line option. The Command Line entry box will expand to take multiple lines of ADL commands and the Quick Reference check box is displayed.

Step 3

Select the Quick Reference check box. The Quick Reference window is displayed.

Step 4

If necessary, use the Go To field to jump to the appropriate part of the Quick Reference listing.

Step 5

Highlight the command you require in the Quick Reference list.

Step 6

Click the Insert in ADL button. The selected ADL command will appear in the Command Line entry box.

Step 7

Where necessary, alter the defaults supplied by the inserted Quick Reference command in the Command Line entry box to suit your program.

Step 8

Repeat steps 4–7 to add more commands as required.

See also:

ADL Command Line
Help and Safety information—Concentration

- Help menu
- Safety home page

Help menu—Concentration

Use the Help menu to view the online Help and software information for this application.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does...</th>
</tr>
</thead>
<tbody>
<tr>
<td>Concentration Help</td>
<td>Select this to display the Concentration Home Page. This contains a list of online Help topics specific to the Concentration Application.</td>
</tr>
<tr>
<td>Help Topics</td>
<td>Select this to display the Cary Eclipse Windows Online Help Home Page.</td>
</tr>
<tr>
<td>Tip of the day</td>
<td>View useful tips about using your Cary Eclipse instrument and software.</td>
</tr>
<tr>
<td>Eclipse Tour</td>
<td>Select this to display the Cary Eclipse Multimedia Online Help. This will give you an animated introduction to the Cary Eclipse software and hardware.</td>
</tr>
<tr>
<td>About...</td>
<td>Select About to access a dialog that displays information about the application as well as Varian email and Internet contact information. To clear this About dialog, select OK.</td>
</tr>
</tbody>
</table>

Safety Home Page

Click on the desired index entry to view the Help.

- About Safety
- Lamp module
- Ultraviolet radiation
- Electrical hazards — Cary Eclipse
- Panels, covers and modules
- Other precautions
- Warnings and cautions
- Warning symbols
- Color coding
- Information symbols
- FCC advisory
- CE compliant products
Concentration How To

Select the procedure you require below for a step-by-step informative guide on running the application software.

Standard Calibrations

- How to perform a calibration
- How to delete a standard from the calibration
- How to change the curve of best fit used in the calibration

Measuring Samples

- How to perform a calibration and manually measure concentrations
- How to perform a calibration and measure concentrations with the Multicell Holder
- How to measure concentrations manually

Other How To

- How to email your results
- How to determine absorptivity
- How to import sample labels, weights and volumes
- How to export collected data
- How to remove or display the command buttons
- How to generate a report from previously calculated data
- How to use advanced reporting options
- How to insert ADL commands using the Quick Reference

See also:
Concentration Home Page
Graphics How To

How to perform a calibration—Concentration

This demonstrates how to perform and store a fluorescence multi-standard calibration, without measuring any sample concentrations.

Follow the steps through, adjusting them to meet the requirements of your particular data collection.

- Start the Concentration application
- Set up data collection parameters
- Set up instrument parameters
- Set up the display options
- Set up accessories
- Set up the calibration
- Set up your samples
- Set up reporting and printing requirements
- Set up storage of collected data
- Set up visual system monitoring
- Finish Setup
- Zero the instrument
- Perform the calibration
Start the Concentration application

Step 1

Click the Windows Start button, point to Programs, Cary Eclipse and then select Scan. The Scan Application is displayed.

Note
If you are running a GLP system, you may be prompted to enter a password before accessing the Concentration application.

Set up data collection parameters

Step 2 [Setup dialog]

Select Setup to display the Setup dialog and specify the method parameters for a new method.

To do this:

Set up instrument parameters

Step 3 [Setup dialog | Cary page]

a. In the Data mode field set the mode to Fluorescence.

b. In the Ex. Wavelength field, set the wavelength of the excitation monochromator, which is the wavelength value at which you want to excite your sample.

c. In the Em. Wavelength field, set the wavelength of the emission monochromator, which is the wavelength value at which your sample emits.

d. Set the slit width of the excitation monochromator.
   Note that as a general rule, at a fixed emission slit width, increasing the excitation slit width will result in an increase in the fluorescence excitation intensity.

e. Set the slit width of the emission monochromator.
   Note that as a general rule, at a fixed excitation slit width, a two-fold increase in the emission slit width will result in a four-fold increase in the emission intensity. Accordingly, a two-fold decrease in the emission slit width will result in a four-fold decrease in the emission intensity.

f. In the Ave Time field enter the required value. A good starting point is 0.1 sec.

Set up the display options

Step 4 [Setup dialog | Options page]

a. Select the Options tab. The Options page is displayed.

b. Enter the minimum and maximum Y values to be displayed on the graph axis during the data collection.

c. Set the Excitation filter for the concentration measurement. It is recommended that you set the filter to Auto (the default setting). This will automatically move the filter wheel to the appropriate position for the selected excitation wavelength.
d. Set the **Excitation filter** and the **Emission filter** for the concentration measurement. In order to minimize steps in the spectra, it is recommended that you set the filter to **Open** (the default setting).

e. Set the EHT or sample detector PMT. The setting you choose will depend upon your sample. For example, a **Low** setting is recommended for highly emitting samples, while a **High** setting is recommended for low emitting samples.

**Set up accessories**

Step 5 [Setup dialog | Accessories page]

Select the **Accessories** tab and ensure that no accessories are selected.

**Set up the calibration**

Step 6 [Setup dialog | Standards page]

a. Select the **Standards** tab to access the **Standards** page where you can set up the standards and their parameters associated with the data collection.

b. Set the appropriate **Units** for your standards for reporting purposes.

c. In the **Standards** field, enter the number of standards that you are using in your calibration. The table below this field will expand or contract to match your choice.

d. In the **Standards** table, enter the concentration of each standard in the **Conc** column.

e. Select the type of curve fitting required for your calibration in the **Fit Type** group.

f. Enter the required $R^2$ value or correlation coefficient in the **Min $R^2$** field. The closer the number is to 1.000 the better the fit. Typically, 0.95 is used.

**Set up your samples**

Step 7 [Setup dialog | Samples page]

As this is a calibration only, set the **No. of Samples** field to ‘0’.

**Set up reporting and printing requirements**

Step 9 [Setup dialog | Reports page]

a. Select the **Reports** tab to display the **Reports** page where you can specify your reporting requirements for this method.

b. Enter your name in the **Name** field.

c. Enter a comment relating to your experiment in the **Comment** field (optional).

d. Set up your report style by selecting the appropriate check boxes in the **Options** group.

For example, select the **Auto Print** check box to obtain a printout of your report automatically. Select the **Parameters** check box to include your experimental parameters in the report.
Select the **Graph** check box to include a calibration graph in the generated report.

### Note

If **Auto Print** is selected, then the system will send the report information to the specified printer as well as the Report area. However if **Auto Print** is not selected, the report will only be sent to the Report area and can be viewed by selecting the **Report** command from the **View** menu.

### Set up storage of collected data

**Step 10 [Setup dialog | Auto Store page]**

a. Select the **Auto Store** tab to display the **Auto Store** page.

b. Set the Storage option to **Off**. The method (with calibration), collected data and report will not be automatically saved. However, you can manually save it all at the end of the collection.

c. Select the **Auto Convert** option you require. If you select **ASCII (csv)** or **ASCII (csv) with log**, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format. The data file will be saved in the current folder.

### Set up visual system monitoring

**Step 11 [Setup dialog]**

Select the **Status Display** check box on any of the Setup pages to display various information fields on your current reaction.

### Finish Setup

**Step 12**

Once you are satisfied with your method setup select **OK** to confirm any changes you have made and clear the **Setup** dialog.

### Zero the instrument

**Step 13**

a. From the Concentration Application window, select the **Zero** button to zero the system. Alternatively, select the **Zero** command from the **Commands** menu. The **Zero** dialog will be displayed.

b. Place a blank in the sample compartment and select **OK**.

### Perform the calibration

**Step 14**

a. Select the **Start** button or select **Start** from the Commands menu. The **Standard/Sample Selection** dialog will be displayed.

b. Select the standards and samples to be used in the analysis. By default, all standards are selected.
c. Select **OK** to clear the **Standard/Sample Selection** dialog.

d. The **Present Standard** dialog will be displayed to prompt you to place the appropriate standard in the sample compartment. Press **OK** to measure the standard.

e. Repeat Step 14 d until you have measured all the standards. The Cary Eclipse will calculate the calibration and the correlation coefficient.

**Note**

If the set correlation coefficient \((R^2)\) value is not met, the Cary Eclipse will prompt you with ‘Min \(R^2\) test failed’.

Step 15

a. Once the run is finished, select the **Save As** command from the **File** menu.

b. In the **File name** field, enter a file name for this Concentration run.

c. Select **Save** and save the method with the calibration as a Method (*.FMCN) file.

**See also:**
- How to change the curve of best fit used in the calibration
- How to delete a standard from the calibration
- How to import samples labels, weights and volumes
- How to export collected data
- How to generate a report from previously calculated data

**How to perform a calibration and manually measure concentrations—Concentration**

This demonstrates how to perform a fluorescence multi-standard calibration and measure sample concentrations using no accessories.

Follow the steps through, adjusting them to meet the requirements of your particular data collection.

- Start the Concentration application
- Set up data collection parameters
- Set up instrument parameters
- Set up display options
- Set up accessories
- Set up the calibration
- Set up your samples and increment names
- Set up your samples for weight and volume correction
- Set up reporting and printing requirements
- Set up storage of collected data
- Set up visual system monitoring
- Finish Setup
- Zero the instrument
- Perform the calibration
- Measure sample concentration
- Save your data

Start the Concentration application

Step 1

Click the Windows Start button, point to Programs, Cary Eclipse and then select Scan. The Scan Application is displayed.

**Note**
If you are running a GLP system you may be prompted to enter a password before accessing the Scan Application.

Set up data collection parameters

Step 2 [Setup dialog]

Select Setup to display the Setup dialog and specify the method parameters for a new method.

To do this:

Set up instrument parameters

Step 3 [Setup dialog | Cary page]

a. In the **Data mode** field set the mode to **Fluorescence**.

b. In the **Ex. Wavelength** field, set the wavelength of the excitation monochromator, which is the wavelength value at which you want to excite your sample.

c. In the **Em. Wavelength** field, set the wavelength of the emission monochromator, which is the wavelength value at which your sample emits.

d. Set the slit width of the excitation monochromator.
   Note that as a general rule, at a fixed emission slit width, increasing the excitation slit width will result in an increase in the fluorescence excitation intensity.

e. Set the slit width of the emission monochromator.
   Note that as a general rule, at a fixed excitation slit width, a two-fold increase in the emission slit width will result in a four-fold increase in the emission intensity. Accordingly, a two-fold decrease in the emission slit width will result in a four-fold decrease in the emission intensity.

f. In the **Ave Time** field enter the required value. A good starting point is 0.1 sec.

Set up the options

Step 4 [Setup dialog | Options page]

a. Select the Options tab. The Options page is displayed.
b. Enter the minimum and maximum Y values to be displayed on the graph axis during the data collection.

c. Set the **Excitation filter** for the concentration measurement. It is recommended that you set the filter to **Auto** (the default setting). This will automatically move the filter wheel to the appropriate position for the selected excitation wavelength.

d. Set the **Excitation filter** and the **Emission filter** for the concentration measurement. In order to minimize steps in the spectra, it is recommended that you set the filter to **Open** (the default setting).

e. Set the EHT or sample detector PMT. The setting you choose will depend upon your sample. For example, a **Low** setting is recommended for highly emitting samples, while a **High** setting is recommended for low emitting samples.

### Set up accessories

Step 5 [Setup dialog | Accessories page]

Select the **Accessories** tab and ensure that no accessories are selected.

### Set up the calibration

Step 6 [Setup dialog | Standards page]

a. Select the **Standards** tab to access the **Standards** page where you can set up the standards and their parameters associated with the data collection.

b. Set the appropriate **Units** for your standards for reporting purposes.

c. In the **Standards** field, enter the number of standards that you are using in your calibration. The table below this field will expand or contract to match your choice.

d. In the **Standards** table, enter the concentration of each standard in the **Conc** column.

e. Select the type of curve fitting required for your calibration in the **Fit Type** group.

f. Enter the required $R^2$ value or correlation coefficient in the **Min $R^2$** field. The closer the number is to 1.000 the better the fit. Typically, 0.95 is used.

### Set up your samples and increment names

Step 7 [Setup dialog | Samples page]

a. Select the **Samples** tab to display the **Samples** page. This page enables you to enter a list of sample names that will be used during your analysis.

b. Enter the number of samples that you are going to use in the **No. of Samples** field. The table below this field will expand or contract to match your choice.

c. In the **Sample names** table enter the name of each sample. You can enter up to 20 characters for each name.

d. If you would like the samples to have the same name with a different numeric extension, enter the name in the first sample position and then select the Increment button.
**Set up weight and volume correction**

Step 8 [Setup dialog | Samples page]

a. Select the **Weight/Volume Corrections** check box.

b. Enter the theoretical sample weight in the **Method Weight** field. This is the weight of the sample specified in your method. (The **Actual Weight** is entered for each sample in the **Sample Names** list.)

c. Enter the units in the **Weight Units** field.

d. Enter the theoretical sample volume in the **Method Volume** field. This is the volume to which the method tells you to make the sample. (The **Actual Volume** is entered for each sample in the **Sample Names** list.)

e. Enter the units in the **Volume Units** field.

f. In the **Sample Names** table, enter the **Actual Weight** and **Actual Volume** for each sample.

**Set up reporting and printing requirements**

Step 9 [Setup dialog | Reports page]

a. Select the **Reports** tab to display the **Reports** page where you can specify your reporting requirements for this method.

b. Enter your name in the **Name** field.

c. Enter a comment relating to your experiment in the **Comment** field (optional).

d. Set up your report style by selecting the appropriate check boxes in the **Options** group.

For example, select the **Auto Print** check box to obtain a printout of your report automatically. Select the **Parameters** check box to include your experimental parameters in the report. Select the **Graph** check box to include a calibration graph in the generated report.

**Note**

If **Auto Print** is selected, then the system will send the report information to the specified printer as well as the Report area. However if **Auto Print** is not selected, the report will only be sent to the Report area and can be viewed by selecting the **Report** command from the **View** menu.

**Set up storage of collected data**

Step 10 [Setup dialog | Auto Store page]

a. Select the **Auto Store** tab to display the **Auto Store** page.

b. Set the Storage option to **Off**. The method (with calibration), collected data and report will not be automatically saved. However, you can manually save it all at the end of the collection.

c. Select the **Auto Convert** option you require. If you select **ASCII (csv)** or **ASCII (csv) with log**, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format. The data file will be saved in the current
folder.

**Set up visual system monitoring**

Step 11 [Setup dialog]

Select the **Status Display** check box on any of the Setup pages to display various information fields on your current reaction.

**Finish Setup**

Step 12

Once you are satisfied with your method setup select **OK** to confirm any changes you have made and clear the **Setup** dialog.

**Zero the instrument**

Step 13

a. From the Concentration Application window, select the **Zero** button to zero the system. Alternatively, select the **Zero** command from the **Commands** menu. The **Zero** dialog will be displayed.

b. Place a blank in the sample compartment and select **OK**.

**Perform the calibration**

Step 14

a. Select the **Start** button or select **Start** from the Commands menu. The **Standard/Sample Selection** dialog will be displayed.

b. Select the standards and samples to be used in the analysis. By default, all standards and samples are selected.

c. Select **OK** to clear the **Standard/Sample Selection** dialog.

d. The **Present Standard** dialog will be displayed to prompt you to place the appropriate standard in the sample compartment. Press **OK** to measure the standard.

e. Repeat Step 14 d until you have measured all the standards. The Cary Eclipse will calculate the calibration and the correlation coefficient.

**Note**

If the set correlation coefficient (R^2) value is not met, the Cary Eclipse will prompt you with ‘Min R^2 test failed’. When you select **OK**, the Cary Eclipse will then prompt you with ‘There is no valid calibration. Proceed in Intensity (a.u.)?’. If you select **Cancel**, the Concentration run will finish. If you select **OK**, the Cary Eclipse will measure the intensity of any presented samples, but will not generate a concentration.

**Measure sample concentration**

Step 15
a. Once all the standards have been read, the **Present Sample** dialog will be displayed to prompt you to place the appropriate sample in the sample compartment. Press **OK** to measure the sample and calculate its concentration. (If replicates have been nominated, then the concentration is calculated after the final sample replicate is read.)

b. Repeat Step 15 b for the remaining samples.

**Save your data**

**Step 16**

a. Once the run is finished, select the **Save As** command from the **File** menu.

b. In the **File name** field, enter a file name for this Concentration run.

c. Select **Save**. The data will be stored as a Batch file.

See also:

- How to change the curve of best fit used in the calibration
- How to delete a standard from the calibration
- How to import samples labels, weights and volumes
- How to perform a calibration and measure concentrations using the Multicell Holder accessory

**How to perform a calibration and measure concentrations using the Multicell Holder—Concentration**

This demonstrates how to perform a fluorescence multi-standard calibration and measure sample concentrations using the Multicell Holder.

Follow the steps through, adjusting them to meet the requirements of your particular data collection.

- Start the Concentration application
- Set up data collection parameters
- Set up instrument parameters
- Set up display options
- Set up Multicell Holder accessory
- Set up the calibration
- Set up your samples and increment names
- Set up weight and volume correction
- Set up reporting and printing requirements
- Set up storage of collected data before run
- Set up visual system monitoring
- Finish Setup
- Zero the instrument
- Perform the calibration and measure samples
- Save your data

**Start the Concentration application**

**Step 1**

Click the Windows **Start** button, point to **Programs, Cary Eclipse** and then select **Scan**. The Scan Application is displayed.
Set up data collection parameters

Step 2 [Setup dialog]

Select Setup to display the Setup dialog and specify the method parameters for a new method.

To do this:

Set up instrument parameters

Step 3 [Setup dialog | Cary page]

a. In the Data mode field set the mode to Fluorescence.

b. In the Ex. Wavelength field, set the wavelength of the excitation monochromator, which is the wavelength value at which you want to excite your sample.

c. In the Em. Wavelength field, set the wavelength of the emission monochromator, which is the wavelength value at which your sample emits.

d. Set the slit width of the excitation monochromator.
   Note that as a general rule, at a fixed emission slit width, increasing the excitation slit width will result in an increase in the fluorescence excitation intensity.

e. Set the slit width of the emission monochromator.
   Note that as a general rule, at a fixed excitation slit width, a two-fold increase in the emission slit width will result in a four-fold increase in the emission intensity. Accordingly, a two-fold decrease in the emission slit width will result in a four-fold decrease in the emission intensity.

f. In the Ave Time field enter the required value. A good starting point is 0.1 sec.

Set up the display options

Step 4 [Setup dialog | Options page]

a. Select the Options tab. The Options page is displayed.

b. Enter the minimum and maximum Y values to be displayed on the graph axis during the data collection.

c. Set the Excitation filter for the concentration measurement. It is recommended that you set the filter to Auto (the default setting). This will automatically move the filter wheel to the appropriate position for the selected excitation wavelength.

d. Set the Excitation filter and the Emission filter for the concentration measurement. In order to minimize steps in the spectra, it is recommended that you set the filter to Open (the default setting).

e. Set the EHT or sample detector PMT. The setting you choose will depend upon your sample. For example, a Low setting is recommended for highly emitting samples, while a High setting is recommended for low emitting samples.

Note
If you are running a GLP system, you may be prompted to enter a password before accessing the Concentration application.
Set up Multicell Holder accessory

Step 5 [Setup dialog | Accessories page]

a. Select the Accessories tab to display the Accessories page.

b. Select the Multicell Holder check box to enable the accessory.

c. Ensure that you have this accessory installed prior to commencing the run.

d. On the Cell Holder diagram select the cells you require.

e. Select the Multi Zero check box to enable the Multi Zero facility.

f. Make sure that no other accessories are selected on this page.

Set up the calibration

Step 6 [Setup dialog | Standards page]

a. Select the Standards tab to access the Standards page where you can set up the standards and their parameters associated with the data collection.

b. Set the appropriate Units for your standards for reporting purposes.

c. Select Replicates or Std Averaging if required, and enter the appropriate number.

d. In the Standards field, enter the number of standards that you are using in your calibration. The table below this field will expand or contract to match your choice.

e. In the Standards table, enter the concentration of each standard in the Conc column.

f. Select the type of curve fitting required for your calibration in the Fit Type group.

g. Enter the required R² value or correlation coefficient in the Min R² field. The closer the number is to 1.000 the better the fit. Typically, 0.95 is used.

Set up your samples and increment names

Step 7 [Setup dialog | Samples page]

a. Select the Samples tab to display the Samples page. This page enables you to enter a list of sample names that will be used during your analysis.

b. Enter the number of samples that you are going to use in the No. of Samples field. The table below this field will expand or contract to match your choice.

c. In the Sample names table enter the name of each sample. You can enter up to 20 characters for each name.

d. If you would like the samples to have the same name with a different numeric extension, enter the name in the first sample position and then select the Increment button.
**Set up weight and volume correction**

Step 8 [Setup dialog | Samples page]

a. Select the **Weight/Volume Corrections** check box.

b. Enter the theoretical sample weight in the **Method Weight** field. This is the weight of the sample specified in your method. (The **Actual Weight** is entered for each sample in the **Sample Names** list.)

c. Enter the units in the **Weight Units** field.

d. Enter the theoretical sample volume in the **Method Volume** field. This is the volume to which the method tells you to make the sample. (The **Actual Volume** is entered for each sample in the **Sample Names** list.)

e. Enter the units in the **Volume Units** field.

f. In the **Sample Names** table, enter the **Actual Weight** and **Actual Volume** for each sample.

**Set up reporting and printing requirements**

Step 9 [Setup dialog | Reports page]

a. Select the **Reports** tab to display the **Reports** page where you can specify your reporting requirements for this method.

b. Enter your name in the **Name** field.

c. Enter a comment relating to your experiment in the **Comment** field *(optional)*.

d. Set up your report style by selecting the appropriate check boxes in the **Options** group.

For example, select the **Auto Print** check box to obtain a printout of your report automatically. Select the **Parameters** check box to include your experimental parameters in the report. Select the **Graph** check box to include a calibration graph in the generated report.

**Note**
If **Auto Print** is selected, then the system will send the report information to the specified printer as well as the Report area. However if **Auto Print** is not selected, the report will only be sent to the Report area and can be viewed by selecting the **Report** command from the **View** menu.

**Set up storage of collected data before run**

Step 10 [Setup dialog | Auto Store page]

a. Select the **Auto Store** tab to display the **Auto Store** page.

b. Set the Storage option to **Off**. The method (with calibration), collected data and report will not be automatically saved. However, you can manually save it all at the end of the collection.

c. Select the **Auto Convert** option you require. If you select **ASCII (csv)** or **ASCII (csv) with log**, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format. The data file will be saved in the current
Set up visual system monitoring

Step 11 [Setup dialog]

Select the Status Display check box on any of the Setup pages to display various information fields on your current reaction.

Finish Setup

Step 12

Once you are satisfied with your method setup select OK to confirm any changes you have made and clear the Setup dialog.

Zero the instrument

Step 13

a. From the Concentration Application window, select the Zero button to zero the system. Alternatively, select the Zero command from the Commands menu. The Zero dialog will be displayed.

b. Place a blank in the sample compartment and select OK.

Perform the calibration and measure samples

Step 14

a. Select the Start button or select Start from the Commands menu. The Standard/Sample Selection dialog will be displayed.

b. Select the standards and samples to be used in the analysis. By default, all standards and samples are selected.

c. Select OK to clear the Standard/Sample Selection dialog.

d. The Cell Loading Guide dialog will be displayed. Load your standards as indicated, then select OK to start reading the standards. If additional Cell Loading Guide dialogs are displayed, continue to load your standards as indicated and select OK to continue loading the standards. Cary Eclipse will measure the standards and calculate the calibration curve.

e. A Cell Loading Guide will be displayed for the samples. Load your samples as indicated and select OK. The Cary Eclipse will then measure the samples and calculate their concentration.

Note

If the set correlation coefficient ($R^2$) value is not met, the Cary Eclipse will prompt you with ‘Min $R^2$ test failed’. When you select OK, the Cary Eclipse will then prompt you with ‘There is no valid calibration. Proceed in Intensity (a.u.)?’. If you select Cancel, the Concentration run will finish. If you select OK, the Cary Eclipse will measure the intensity of any presented samples, but will not generate a concentration.

Save your data
Step 15

a. Once the run is finished, select the **Save As** command from the **File** menu.

b. In the **File name** field, enter a file name for this Concentration run.

c. Select **Save**. The data will be stored as a Batch file.

See also:
- How to change the curve of best fit used in the calibration
- How to delete a standard from the calibration
- How to import samples labels, weights and volumes

### How to measure concentrations manually—Concentration

You can use the Concentration application without calibrating the system to measure samples in intensity mode. You can also use the duplicate/triplicate or replicate feature of the Concentration software to allow simple statistical calculations to be applied to your measurements. These measurements modes can be used in conjunction with the accessories.

- Start the Concentration application
- Set up data collection parameters
- Set up instrument parameters
- Set up the options
- Set up accessories
- Set up the calibration
- Set up your samples and increment names
- Set up reporting and printing requirements
- Set up storage of collected data
- Set up visual system monitoring
- Finish Setup
- Zero the instrument
- Measure sample intensity
- Save your data

### Start the Concentration application

Step 1

Click the Windows **Start** button, point to **Programs, Cary Eclipse** and then select **Scan**. The Scan Application is displayed.

**Note**
If you are running a GLP system you may be prompted to enter a password before accessing the Scan Application.

### Set up data collection parameters

Step 2 [Setup dialog]

Select **Setup** to display the **Setup** dialog and specify the method parameters for a new method.

To do this:
Set up instrument parameters

Step 3 [Setup dialog | Cary page]

a. In the **Data mode** field set the mode to **Fluorescence**.

b. In the **Ex. Wavelength** field, set the wavelength of the excitation monochromator, which is the wavelength value at which you want to excite your sample.

c. In the **Em. Wavelength** field, set the wavelength of the emission monochromator, which is the wavelength value at which your sample emits.

d. Set the slit width of the excitation monochromator.
   Note that as a general rule, at a fixed emission slit width, increasing the excitation slit width will result in an increase in the fluorescence excitation intensity.

e. Set the slit width of the emission monochromator.
   Note that as a general rule, at a fixed excitation slit width, a two-fold increase in the emission slit width will result in a four-fold increase in the emission intensity. Accordingly, a two-fold decrease in the emission slit width will result in a four-fold decrease in the emission intensity.

f. In the **Ave Time** field enter the required value. A good starting point is 0.1 sec.

Set up the options

Step 4 [Setup dialog | Options page]

a. Select the **Options** tab. The **Options** page is displayed.

b. Enter the minimum and maximum Y values to be displayed on the graph axis during the data collection.

c. Set the **Excitation filter** for the concentration measurement. It is recommended that you set the filter to **Auto** (the default setting). This will automatically move the filter wheel to the appropriate position for the selected excitation wavelength.

d. Set the **Excitation filter** and the **Emission filter** for the concentration measurement. In order to minimize steps in the spectra, it is recommended that you set the filter to **Open** (the default setting).

e. Set the EHT or sample detector PMT. The setting you choose will depend upon your sample. For example, a **Low** setting is recommended for highly emitting samples, while a **High** setting is recommended for low emitting samples.

Set up accessories

Step 5 [Setup dialog | Accessories page]

Select the **Accessories** tab and ensure that no accessories are selected.

Set up the calibration

Step 7 [Setup dialog | Standards tab]

a. Select the **Standards** tab. The **Standards** page is displayed.
b. Set the **Standards** field to '1', (It is not possible to set this field to '0').

**Set up your samples and increment names**

**Step 7 [Setup dialog | Samples page]**

a. Select the **Samples** tab to display the **Samples** page. This page enables you to enter a list of sample names that will be used during your analysis.

b. Enter the number of samples that you are going to use in the **No. of Samples** field. The table below this field will expand or contract to match your choice.

c. In the **Sample names** table enter the name of each sample. You can enter up to 20 characters for each name.

d. If you would like the samples to have the same name with a different numeric extension, enter the name in the first sample position and then select the Increment button.

**Set up storage of collected data**

**Step 8 [Setup dialog | Auto Store page]**

a. Select the **Auto Store** tab to display the **Auto Store** page.

b. Set the Storage option to **Off**. The method (with calibration), collected data and report will not be automatically saved. However, you can manually save it all at the end of the collection.

c. Select the **Auto Convert** option you require. If you select **ASCII (csv)** or **ASCII (csv) with log**, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format. The data file will be saved in the current folder.

**Set up reporting and printing requirements**

**Step 9 [Setup dialog | Reports page]**

a. Select the **Reports** tab to display the **Reports** page where you can specify your reporting requirements for this method.

b. Enter your name in the **Name** field.

c. Enter a comment relating to your experiment in the **Comment** field *(optional)*.

d. Set up your report style by selecting the appropriate check boxes in the **Options** group.

For example, select the **Auto Print** check box to obtain a printout of your report automatically. Select the **Parameters** check box to include your experimental parameters in the report. Select the **Graph** check box to include a calibration graph in the generated report.

**Note**

If **Auto Print** is selected, then the system will send the report information to the specified printer as well as the Report area. However if **Auto Print** is not selected, the report will only be sent to the Report area.
and can be viewed by selecting the Report command from the View menu.

Set up visual system monitoring

Step 10 [Setup dialog]

Select the Status Display check box on any of the Setup pages to display various information fields on your current reaction.

Finish Setup

Step 11

Once you are satisfied with your method setup select OK to confirm any changes you have made and clear the Setup dialog.

Note
As a calibration is not to be performed it is recommended the display of the graphics area is turned off. To do this, clear the Graphics option in the View. At this stage you should consider saving the method from the File menu, Save method as, for future use.

Zero the instrument

Step 12

a. From the Concentration Application window, select the Zero button to zero the system. Alternatively, select the Zero command from the Commands menu. The Zero dialog will be displayed.

b. Place a blank in the sample compartment and select OK.

Measure sample intensity

Step 13

a. Select the Start button or select Start from the Commands menu. The Standard/Sample Selection dialog will appear.

b. In the Selected for Analysis list, highlight the standard and select \[\text{\textless}\] to transfer it to the Solutions Available list. The standard will not be included in the analysis.

c. Select the samples to be used in the analysis. By default, all samples are selected.

d. Select OK to clear the Standard/Sample Selection dialog. As no standards have been selected the message ‘There is no valid calibration. Proceed in Intensity (a.u.)?’ will be displayed.

e. Select OK to continue.

f. The Present Sample dialog will be displayed to prompt you to place the appropriate sample in the sample compartment.

Note
If you are using an accessory, a dialog specific to that accessory will appear in place of the Present
Sample dialog.

g. Select **OK** to measure the sample and display the intensity. (If replicates have been nominated, then the mean intensity is calculated after the final sample replicate is read.)

h. Repeat Steps 13 f and 13 g for the remaining samples.

Save your data

Step 14

a. Once the run is finished, select the **Save As** command from the **File** menu.

b. In the **File name** field, enter a file name for this Concentration run.

c. Select **Save**. The data will be stored as a Batch file.

See also:
- How to import samples labels, weights and volumes
- How to export collected data
- How to generate a report from previously calculated data

How to change the curve of best fit used in the calibration—Concentration

- Open stored calibration and sample data
- Change calibration curve type
- Start recalculation

Once the standards have been measured and the calibration curve plotted on the screen, you may wish to change the type of curve used to fit the standard points.

Open stored calibration and sample data

Step 1

From the **File** menu, select **Open** to display the Windows **Open** dialog.

A list of previously stored batch files will appear. Select the batch file you require then select the **Open** button.

The system will automatically retrieve the method and calibration that was used to measure the samples as well as the sample data.

Change calibration curve type

Step 2

Select the **Recalculate** button to display the **Recalculate** dialog. This dialog contains the **Standards** and **Reports** pages which enable you to set up the recalculation parameters.

Step 3
Select the Standards tab. The Standards page is displayed.

Step 4

Select the Fit Type algorithm by selecting the button that corresponds to the fit type you require.

**Start recalculation**

Step 5

Select **OK** to perform the calibration using the selected Fit Type.

The Cary Eclipse will create a calibration curve by fitting the selected fit curve to the data results. The calibration equation and correlation coefficient will also be sent to the Report area along with the recalculated sample results.

See also:
- How to delete a standard from the calibration
- How to export collected data
- How to generate a report from previously calculated data
- How to remove or display the command buttons

How to delete a standard from the calibration—Concentration

- Open stored calibration and sample data
- Change the calibration parameters
- Start recalculation

Once the standards have been measured and the calibration curve plotted on the screen, you may wish to delete one or more of the standards from the calibration curve if they are obviously in error.

**Open stored calibration and sample data**

Step 1

From the File menu, select **Open** to display the Windows **Open** dialog.

A list of previously stored batch files will appear. Select the batch file you require then select the **Open** button.

The system will automatically retrieve the method and calibration that was used to measure the samples as well as the sample data.

**Change the calibration parameters**

Step 2

Select the Recalculate button to display the Recalculate dialog. This dialog contains the Standards and Reports pages which enable you to set up the recalculation parameters.

Step 3

Select the Standards tab. The Standards page is displayed.
Step 4

In the **Use** column of the **Standards** table, clear the standard/s you do not want to be read. To do this, double-click on the relevant cell to toggle to **No**. (You can also highlight the cell and select any key on the keyboard). The standards that you require for the calibration should be set to **Yes**.

**Start recalculation**

Step 5

Select **OK** to perform the calibration of the selected standards.

The Cary Eclipse will create a calibration curve by fitting the selected fit curve to the data results. The calibration equation and correlation coefficient will also be sent to the Report area along with the recalculated sample results.

See also:

- How to change the curve of best fit used in the calibration
- How to export collected data
- How to generate a report from previously calculated data
- How to remove or display the command buttons

**How to determine the absorptivity of a sample—Concentration**

Step 1

Using the Concentration application, prepare a calibration curve using the appropriate standards.

Step 2

Create a calibration curve using **Linear Direct** as the **Fit Type**.

Step 3

Now look at the calibration equation that is reported in the calibration report. e.g. \( \text{Abs} = 0.05692 \times \text{Conc} \).

Using the Beer-Lambert law, which states that:

\[
A = \varepsilon l c
\]

where:
- \( A \) = absorbance
- \( \varepsilon \) = absorptivity, \( \text{cm}^{-1} \text{M}^{-1} \)
- \( l \) = pathlength, cm
- \( C \) = concentration of the solute, M

You can see that, in this example, 0.05692 = \( \varepsilon l \), or the slope of the calibration curve.
Knowing the pathlength of your cell, you can therefore calculate the absorptivity.

For example:
If cell pathlength \( l \) = 1 cm, then Absorptivity \( (\varepsilon) \) = \( 0.05692 \text{ cm}^{-1}\text{M}^{-1} \)
If cell pathlength \( l \) = 0.2 cm, then Absorptivity \( (\varepsilon) \) = \( 0.2846 \text{ cm}^{-1}\text{M}^{-1} \)

See also:
- How to perform a calibration
- How to perform a calibration and manually measure concentrations
- How to perform a calibration and measure concentrations using the Multicell Holder accessory

**How to import sample labels, weights and volumes—Concentration**

If you want to import sample data into the Concentration application, then you simply need to enter your data in a *.txt file and then open this in the Concentration application. The data should be entered in the following manner:

Each sample name in the imported file must be on a separate line, and any tabs in the file are ignored. If you want to include duplicates or triplicates, then they must be on a separate line immediately after the initial sample name and called ‘Dup’ and ‘trip’ (maintain the case as shown). Place the sample number immediately after the Dup or trip identifier.

Weight correction and volume correction can be added immediately after the sample name in that order. (i.e. sample_name, weight_correction, volume_correction). If no entry is added for weight and volume, the Cary Eclipse will assign a value of '1'.

An example file is given below.

```
Sample_name1, 1.0001, 1.01,
Dup1, 1.0002, 1.02,
trip1,1.0003, 1.03,
Sample_name2, 1.0004, 1.04,
Dup2, 1.0005, 1.05,
trip2,1.0006, 1.06,
Sample_name3, 1.0007, 1.07,
Dup3, 1.0008, 1.08,
trip3,1.0009, 1.09,
Sample_name4, 1.0010, 1.10,
Dup4, 1.0011, 1.11,
trip4,1.0012, 1.12,
Sample_name5, 1.0013, 1.13,
Dup5, 1.0014, 1.14,
trip5,1.0015, 1.15,
```

Slope = 4
Step 1
Enter your data into a text editor and save it as a text file.

Step 2
From the Concentration Application window, select the **Setup** button to display the **Setup** dialog.

Step 3
Select the **Samples** tab to display the **Samples** page. Set the **Sample Averaging** field to that the number of repeat samples specified in your sample file (the example above, would be 3).

Step 5
If you are using weight or volume corrections in your sample file, select **Weight/volume corrections**.

Step 6
Select the **Import Names** button. The **Open** dialog is displayed.

Step 7
Browse for your text file and select **Open** to open the file. The sample names will appear in the **Sample Names** table.

See also:
- How to export collected data
- How to generate a report from previously calculated data
- How to remove the command buttons from the application window

**How to email your results—Concentration**

You may wish to send an email to a person once the read is completed. This useful facility can be used to alert you to the completion of the read, particularly if you are working away from the laboratory where the Cary Eclipse instrument is.

To do this:

Step 1
Ensure that you have set up the **Host** and **Port** options on the **Email options** page of the System Information application in order for the email facility to work correctly.

Step 2
From the **Setup** menu, select **Auto Store**. The **Auto Store** page is displayed.

Step 3
Select the Email check box to enable the email options.

Step 4

In the To field, enter the recipient's email address (for example, rchandle@varianinc.com). To enter multiple recipients, simply type a comma ',' between each email address.

Step 5

In the From field, enter the email address of the computer on which the Cary Eclipse software is installed. This may be your email address, or a communal email address, if the Cary Eclipse software is installed on a computer that has multiple users.

Step 6

Enter information in the Subject and Message fields as required.

Step 7

Select Attach current data. The data that has been collected during the run will be attached to the email.

**Hot Tip**
You can attach other documents to your email. To do this, select the Attached button and browse your computer to select a file, using the Windows® Open dialog that is displayed.

Step 8

Select OK to clear the Auto Store dialog. Your email will be sent once the run has been completed.

See also: Auto Store

How to generate a report from previously calculated data—Concentration

Step 1

Select Open… from the File menu to display the Windows Open dialog.

Step 2

A list of previously stored files will appear. If this is not a list of batch files, select the down arrow at the right of the Files of type list box and select 'Batch' to list all the batch files.

Step 3

Select the Batch file you require then select Open.

The system will automatically retrieve the collected data into the Graphics area. The method that was used to collect the data will also be retrieved.
Step 4

Select the **Recalculate** button to display the **Setup** dialog.

Step 5

Select the **Reports** tab and on the **Reports** page specify the information you would like to include in the report. Select **Auto Print** if you require the new report to print to the Reports area and to a printer.

Step 6

Click **OK** to display the results in the Report area (and send to the printer if **Auto Print** is selected).

**Note**

If you require a printout of only the current graphics in the Graphics area of a previously stored data file:

a. Select the **Clear Report** option in the Commands menu to clear any text that may be displayed in the Report area.

b. Press the **Print** button to obtain a printout of the graphics.

Alternatively tick only the **Graph** check box in the **Reports** page to obtain a printout of graphics only.

**See also:**

[How to use advanced reporting options](#)

**How to use advanced reporting options—Concentration**

This outlines ways to fully customize your reports. Options available include:

- Including header and footer information
- Including a logo
- Editing the text in the report
- Viewing the report on screen before printing
- Cutting and pasting to a word processor

**Including header and footer information**

You can have text appearing at the top and bottom of every page in your report. To specify what the text should be, do the following:

Step 1

On the Windows Taskbar, click the Windows **Start** button, point to **Programs, Cary Eclipse** and then select **System Information**. The System Information application appears with the Company page displayed.

Step 2

Enter the header and footer information in the **Header and Footer** fields.
Step 3

Click OK to implement your changes.

Including a logo

You can include a logo on the first page of your report. To specify what the logo should be, do the following:

Step 1

On the Windows Taskbar, click the Windows Start button, point to Programs, Cary Eclipse and then select System Information. The System Information application appears with the Company page displayed.

Step 2

Enter a logo using the Add Logo button. This logo will always be included on the front page of reports.

Step 3

Click OK to implement your changes.

Q. Why isn't the logo I selected included in my report?
A. If you are running the Cary Eclipse software under Windows NT you need to be logged on as an Administrator in order to specify the logo in the System Information application.

Editing the text in the report

You can edit any of the text in the Report area. Just select Edit Report from the Edit menu. You can then add, delete and change text in the Report area.

Q. The Edit Report option is not available, why not?
A. You do not have reporting editing privileges set in the GLP Administration application. The Administrator of the system will have to change your privileges before you can edit reports.

Viewing the report on the screen before printing

You can view a report before printing it. To do this, from the File menu select Print Preview. You can then ensure that the report format is correct before you print it.

Hot Tip
Use the arrow keys at the bottom of the Print Preview screen to change pages.

Cutting and pasting to a word processor

To have complete control over how your report looks you may wish to copy it to a word processor program, such as Microsoft Word. To do this:

Step 1

Open a report.

Step 2
From the **Edit** menu, choose **Edit Report**.

**Step 3**

Click in the Report area and highlight the text you want to export to your word processor.

**Step 4**

Right-click on the Report area and select **Copy** from the menu.

**Step 5**

Start up your word processor and use the Paste function (usually under the Edit menu) to place the text into a document.

See also:

- How to add a label to a graph
- How to add a picture to a graph
- How to alter the look of your graphs
- How to display a trace that is not appearing

**How to export collected data—Concentration**

If you want to export data from a Cary Eclipse application into a third-party application, then you simply need to save your data as a Spreadsheet ASCII (*.CSV) file or a Rich Text Format (*.RTF) file. Spreadsheet ASCII files are recommended if you are planning to use the data in a spreadsheet program such as Microsoft® Excel. Rich Text Format files are recommended if you are planning to open the data in a word processor program such as Microsoft® Word.

To export collected data:

**Step 1**

From the **File** menu, select **Save As**. The **Save As** dialog is displayed.

**Step 2**

Click on the down arrow next to the **Files of type** list box and select the **Spreadsheet ASCII (*.CSV)** or **Rich Text Format (*.RTF)** option.

**Step 4**

Type the name of your new file in the **File name** field.

**Step 5**

Select **Save** to export your data to this new file.

**How to remove or display the command buttons—Concentration**

The command buttons (**Setup, Zero, Reread, Pre-scan, Clear Report, Recalculate** and **Print**) appear on the left side of the Concentration Application window.
To remove the command buttons

Step 1

From the Concentration Application window, select the View menu.

Step 2

Clear the Buttons menu item.

The buttons will disappear from the application screen, giving you more room to display reports.

To show the command buttons

Step 1

From the Concentration Application window, select the View menu.

Step 2

Select the Buttons menu item.

The buttons will re-appear in the application screen.

Graphics How to—Concentration

Traces

- How to track the focused trace
- How to select the focused trace
- How to display a trace that is not appearing in a graph box
- How to remove a trace from a graph
- How to remove a trace from the trace List

Sizing and scale

- How to resize graph boxes
- How to scale your graph manually
- How to scale your graph automatically
- How to alter the look of your graphs
- How to zoom a graph

Labels

- How to add a label to a graph
- How to edit a label
- How to delete a label
- How to move a label

Pictures
How to add a label to a graph—Concentration

You can add a label to your graph or Graphics area background. To do this:

Step 1

Click on the graph box you require or click outside the graph box(es) to choose the Graphics area background.

Note
There must be at least one trace Visible in the graph box before you can add a label to it.

Step 2

Click the Add Label button on the toolbar or right-click on the graph and select Add Label from the menu. The Add Label dialog will appear. The Add Label command is also accessible from the Graph menu.

Step 3

Click the Font button. The Windows Font dialog appears. Select the font type, size and style you require for your label. A sample of the font style appears in the Sample field. When you are satisfied with your selections, select OK to return to the Add Label dialog.

Step 4

Enter the label in the Label field and select OK. The label will be displayed on the selected graph. If necessary, you can move a label so that it is more appropriately placed on your graph.

See also:
How to move a label
How to edit a label
How to resize a label

How to edit a label—Concentration

Step 1

Place the cursor on the label and click the left mouse button to select the label.

Hot Tip
If you are having trouble selecting the label it may be that a trace is interfering with the label selection. Try zooming in and clicking in a different part of the label.

Step 2
Click the right mouse button and select **Edit Annotation** from the drop-down menu. Alternatively you can select the **Edit Annotation** command from the **Graph** menu. The **Edit Label** dialog is displayed.

**Step 3**

Use the **Edit Label** dialog to change the label.

For example, if you want to change the font size of the label, select the **Font** button. In the **Size** field, scroll through the available sizes for the font and select the size you require. Alternatively, highlight the currently selected size in the **Size** entry field and enter the size you require. Click the **OK** button in this dialog to return to the **Edit Label** dialog.

**Step 4**

Click **OK** to clear the **Edit Label** dialog. Your label will appear with the new font size.

See also:
- [How to add a label to a graph](#)
- [How to move a label](#)
- [How to delete a label](#)

**How to move a label—Concentration**

To move a label so that it is more appropriately placed on your graph or Graphics area background, do the following:

**Step 1**

Place the cursor on the label.

**Step 2**

Click and hold down the left mouse button.

**Hot Tip**

If you are having trouble selecting the label, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

**Step 3**

Drag the label to the correct position and release the mouse button. The label will be displayed at the new position.

**Note**

Labels are transparent so as not to interfere with any traces on graphs.

See also:
- [How to add a label to a graph](#)
- [How to edit a label](#)
- [How to resize a label](#)
- [How to delete a label](#)

**How to resize a label—Concentration**
To resize a label so that it is more appropriately sized on your graph or Graphics area background, do the following:

Step 1

Select the label by clicking on it with the mouse. A border will be displayed around the label to indicate it is selected.

![Hot Tip]
If you are having trouble selecting the label, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Position the cursor over one of the corner edges of the label until the cursor becomes a diagonal double-headed arrow. Click and hold down the left mouse button.

![Example of label showing the border that indicates it is selected and the arrow to be used for re-sizing]

Step 3

Increase or decrease the size of the label by dragging the mouse. The label text will also be resized. Release the mouse button when the label and text are the correct size.

![Note]
Labels are transparent so as not to interfere with any traces on graphs.

See also:
- How to add a label to a graph
- How to edit a label
- How to delete a label

**How to delete a label—Concentration**

To delete a label from a graph or Graphics area background, do the following:

Step 1

Position the cursor on the label and click the left mouse button to select it.

![Hot Tip]
If you are having trouble selecting the label it may be that a trace is interfering with the label selection. Try zooming in and clicking in a different part of the label.

Step 2

Click with the right mouse button and select **Delete Annotation** from the drop-down menu displayed. Alternatively, you can select the **Delete Annotation** command from the **Graph** menu. The label will be deleted from the graph or Graphics area.
How to add a picture to a graph—Concentration

You can add a picture to your graph or the Graphics area background. To do this:

Step 1

Click on the graph box you require or click outside the graph box(es) to choose the Graphics area background.

Note
There must be at least one trace Visible in the graph box before you can add a picture to it.

Step 2

Click the Add Picture button on the toolbar or right-click on the graph and select Add Picture from the menu. The Add Picture dialog will appear. The Add Picture command is also accessible from the Graph menu.

Step 3

Click the Load button. The Windows Open dialog appears.

Step 4

Browse through your directories to find the picture you want to insert. All pictures must be in the bitmap format and will have the extension .BMP.

Step 5

Highlight the .BMP that you want to insert and select Open.

Step 6

The Add Picture dialog will appear showing a preview of the bitmap. If the bitmap is the correct one select the OK button. The bitmap will be placed onto the graph.

Note
You should take care not to add a bitmap that is too big as it will obscure part or all of your graph.

You can move a picture so that it is more appropriately placed on your graph.

See also:
How to move a picture to a graph
How to delete a picture
How to replace a picture
How to resize a picture

How to replace a picture—Concentration
To change a picture that you have added to a graph or Graphics area background do the following:

Step 1

Move the cursor to the picture and select the left mouse button to select it.

**Hot Tip**
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Click the right mouse button and select *Edit Annotation* from the drop-down menu or select *Edit Annotation* from the *Graph* menu. The *Edit Picture* dialog is displayed.

Step 3

From the *Edit Picture* dialog, select the *Load* button. The Windows *Open* dialog is displayed.

Step 4

Browse through your directories to find the picture to replace the current bitmap. Picture files must be in one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.

Step 5

Highlight the file that you want to insert and select *Open* to return to the *Edit Picture* dialog.

Step 6

The *Edit Picture* dialog will display a preview of the bitmap. If the bitmap is the correct one select the OK button. The bitmap will be placed onto the graph. If you need to, move the picture so that it is more appropriately placed on your graph.

**Note**
Do not to add a bitmap that is too big as it will obscure part or all of your graph.

See also:
- How to add a picture to a graph
- How to move a picture
- How to delete a picture
- How to resize a picture

**How to move a picture—Concentration**

To move a picture so that it is more appropriately placed on your graph or Graphics area background, do the following:

Step 1

Place the cursor on the picture.

Step 2
Click and hold down the left mouse button.

**Hot Tip**
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 3

Drag the picture to the correct position and release the mouse button. The picture will be displayed at the new position.

**See also:**
How to add a picture to a graph
How to delete a picture
How to replace a picture
How to resize a picture

**How to resize a picture—Concentration**

To resize a picture so that it is more appropriately sized on your graph or Graphics area background, do the following:

Step 1

Select the picture by clicking on it with the mouse. A border will be displayed around the picture to indicate it is selected.

**Hot Tip**
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Position the cursor over one of the corner edges of the picture until the cursor becomes a diagonal double-headed arrow. Click and hold down the left mouse button.

*Example of picture showing the border that indicates it is selected and the arrow to be used for re-sizing*

Step 3

Increase or decrease the size of the picture by dragging the mouse. The picture will also be resized. Release the mouse button when the picture is the correct size.

**See also:**
How to add a picture to a graph
How to delete a picture—Concentration

To delete a picture from a graph or Graphics area background, do the following:

Step 1

Position the cursor on the picture and click the left mouse button to select it.

**Hot Tip**
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Click with the right mouse button and select **Delete Annotation** from the drop-down menu displayed. Alternatively, you can select the **Delete Annotation** command from the **Graph** menu. The picture will be deleted from the graph or Graphics area.

See also:
- How to add a picture to a graph
- How to move a picture
- How to zoom a graph
- How to resize a picture

How to select the focused trace—Concentration

To select the scan that you want to be the focused (red) trace, do one the following:

- From the Graph, click the left mouse button on the trace that you want to make the focused trace.
- From the Graph Legend (if displayed), click the left mouse button on the trace that you want to make the focused trace.
- From the **Trace Preferences** dialog, click in the **Color** column of the trace that you want to make the focused trace.

**Note**
Select the **Show Legend** check box in the **Graph Preferences** dialog to display the Graph Legend.

See also:
- How to track the focused trace
- How to view a particular trace

How to display a trace that is not appearing in the graph box—Concentration

Step 1

Click on the graph in the Graphics area.
Step 2

Select the **Trace Preferences** button  from the Graphics toolbar, or select **Trace Preferences** from the **Graph** menu. The **Trace Preferences** dialog is displayed.

Step 3

Find that trace that you wish to see and select the **Visible** option. A tick will appear in the **Visible** column to indicate that it will now be displayed.

If you wish to make multiple traces visible, do the following:

a. Hold down the CTRL key select all the traces that you want to display.

b. Click the right mouse button and select the **Show Traces** command from the drop-down menu. Ticks will appear next to the selected traces to indicate that they will now be displayed.

Step 4

Press **OK** to clear the **Trace Preferences** dialog. The selected trace(s) will be displayed.

---

See also:

- [How to remove a trace from a graph](#)
- [How to remove a trace from the trace list](#)

**How to remove a trace from a graph—Concentration**

Step 1

Select a graph in the Graphics area.

Step 2

Click the **Trace Preferences** button  on the Graphics toolbar or select **Trace Preferences** from the **Graph** menu. The Trace Preferences dialog is displayed.

Step 3

Find that trace that you no longer wish to see and select the **Visible** option. The tick will be removed from the **Visible** column to indicate that it will not be displayed.

If you wish to make multiple traces visible, do the following:

a. Hold down the CTRL key select all the traces that you no longer want to display.

b. Click the right mouse button and select the **Hide Traces** command from the drop-down menu. The ticks will be removed from the selected traces to indicate that they will not be displayed.

Step 4

Select **OK** to clear the **Trace Preferences** dialog. The trace will be removed from the graph.

---

See also:

- [How to display a trace that is not appearing in the graph box](#)
How to remove a trace from the trace list

How to remove a trace from the trace list—Concentration

If your graphs contain unwanted traces, you may wish to delete them (rather than simply making them invisible) in order to free up application memory and aid system performance.

Step 1

Select a graph in the Graphics area.

Step 2

Click the **Trace Preferences** button on the Graphics toolbar or select **Trace Preferences** from the Graph menu. The Trace Preferences dialog is displayed.

Step 3

Select the trace to be removed by clicking on it.

Step 4

Click the right mouse button to display the **Trace Preferences** menu.

Step 5

Select **Delete Traces** to delete the sample from the trace list. You will be prompted to confirm this action.

Step 6

Select **OK** to clear the **Trace Preferences** dialog. The trace will be removed from all graphs.

See also:

- How to remove a trace from a graph
- How to display a trace that is not appearing in the graph box

How to alter the look of your graphs—Concentration

Step 1

Click the **Graph Preferences** button on the Graphics toolbar, or select **Graph Preferences** from the **Graph** menu. The **Graph Preferences** dialog is displayed.

Step 2

Use the available options available to choose your preferred appearance for the graphs. A preview box in the top right-hand corner will demonstrate how the graphs will appear.

Step 3

Select **OK** to clear the **Graph Preferences** dialog and update all graphs to reflect your selections.
Step 4

To re-use your graphics setup at a later date, save the setup as a graphics template.

See also:
- How to add a label to a graph
- How to add a picture to a graph
- How to move a label
- How to move a picture
- How to resize graph boxes
- How to make a graphics template

How to track the focused trace—Concentration

To track the focused (red) trace in the graph:

Step 1

Click the **Cursor Modes** button ![Cursor Modes](cursor_modes.png) from the Graphics toolbar, or select **Cursor Mode** from the **Graph** menu. The **Cursor Modes** dialog will appear.

Step 2

Select **Track** and select the **OK** button.

Step 3

Place the blue tracking cursor in the graph box you require.

Step 4

Move the cursor left and right across the graph box to trace the focused (red) scan. The cursor coordinates change (in the X, Y field at the bottom left hand corner of the Graphics area) as you move along the curve.

**Hot Tip**

If you wish to fix the cursor on a particular position on the curve, move the cursor to the required position. Press `ALT` on the keyboard, then release the mouse button.

See also:
- How to select the focused trace
- How to view a particular trace

How to scale your graph manually—Concentration

Step 1
Click on the graph whose axes you want to change to make it the focused trace.

Step 2

Click the Axes Scales button \(\text{_axes_scales}\) on the toolbar, or select **Axes Scales** from the **Graph** menu. The Axes Scales dialog is displayed.

Step 3

Type in the new ranges for the X and Y axes in the format ‘n-n’, or choose an option for each from the drop-down list.

Step 4

Select **OK**. The axes on the selected graph will change to reflect your chosen settings.

**Hot Tip**
Select the **Zoom Out** command to resize the graph to its original settings.

**See also:**
- How to zoom a graph
- How to scale a graph automatically

**How to scale your graph automatically—Concentration**

Step 1

Click on the graph whose axes you want to change to make it the focused trace.

Step 2

To autoscale the graph, select one of the following:

Click the Autoscale XY button \(\text{autoscale_xy}\) on the toolbar to autoscale both X and Y axes.

Click the Autoscale X button \(\text{autoscale_x}\) on the toolbar to autoscale the X axis only.

Click the Autoscale Y button \(\text{autoscale_y}\) on the toolbar to autoscale the Y axis only.

The focused trace will be scaled according to the option selected.

**Hot Tip**
Select the **Zoom Out** command to resize the graph to its original settings.

**See also:**
- How to zoom a graph
- How to scale your graph manually

**How to make a graphics template—Concentration**
A graphics template lets you set up and store the style of a graphics report for future use. You can do this by adding labels and pictures to the background of the Graphics area and altering any axes styles etc. One advantage of this over the application's built-in reporting features is that you have more flexibility as to where you can position graph boxes and any text that you may want to appear (e.g. operator name, batch number etc.) and also where you add pictures, e.g. company logos etc.

Step 1

Start the application and open the data file from which you want to make a graphics template. Alternatively, collect some new data.

Step 2

Make as many graph boxes as you require in your template by using the Add Graph function.

Step 3

Adjust the graph boxes to the correct size and position them as you want them to appear in the template.

Step 4

Do any of the following, if required:

- Change the appearance of the graph boxes in the Graph Preferences dialog
- Use the Add Label function to append text to the Graphics area
- Use the Add Picture function to add pictures, such as your company's logo to the Graphics area

Step 5

From the File menu, select Print Preview. Use the Print Preview dialog displayed to check the template that you have created.

Step 6

When you are satisfied with the appearance of the graphics, select the Save As… command from the File menu. The Save As dialog is displayed.

Step 7

A list of stored data files will appear. Select the down arrow at the right of the Files of type field and select 'Graph Template' to list all the template files. Ensure that the Save only focused trace check box is cleared.

Step 8

Enter a name for the template in the File name field.

Step 9

Select OK to save the graphic template. The template will be used each time you create a new file.

Note

You can also make a graphic template in a similar manner from existing data. No data will be saved in the template.
How to resize graph boxes—Concentration

Step 1

Click on the graph that you want to resize so that the highlight box appears around it.

Step 2

Move the cursor to the edge of the highlight box and place it over one of the square marks (e.g. ■) in the box until it turns into a double headed arrow (↔).

Step 3

Click and hold the left mouse button and drag the box to the desired size.

Step 4

Release the left mouse button.

See also:
- Example of a Graphics Template
- How to add a label to a graph
- How to add a picture to a graph
- How to move a label
- How to move a picture

How to zoom a graph—Concentration

To zoom a graph, do the following:

Step 1

Click on the button on the toolbar, or select Cursor Mode from the Graph menu. The Cursor Modes dialog will appear.

Step 2

Select Free and select the OK button.

Step 3

Click and hold down left mouse button in the highlighted graph box.

Step 4

Drag the mouse sideways and up or down. You will see a zoom box made up of a dotted line appear on the screen. (See picture below.)
Step 5

Release the left mouse button. The area defined by the zoom box will be redrawn to fill the entire graph box, and the scans within the box will be enlarged accordingly. The abscissa (X) and ordinate (Y) coordinates displayed below the graph will also be changed to reflect the positions of the corners of the zoom box.

You can repeat this process if you need to zoom in even further.

**Note**
The system may limit the extent to which you can Zoom.

**See also:**
- **How to scale your graph manually**
- **How to scale your graph automatically**
GLP Administration Home Page

Introduction

The GLP (Good Laboratory Practice) Administration application is used to protect the system from unauthorized use, enabling application-specific privileges to be turned on or off by the system administrator.

The GLP Administration application controls the access privileges of user groups operating the Cary Eclipse. The method of control includes the access of workgroups (which can be groups of one user) to the Cary Eclipse software, including the ability to reset the instrument or modify reports and methods, and the establishment of passwords. If this application is operational, users will need to be registered, have a user name and a valid password before they can access the various privileges.

At installation, select Custom and nominate GLP to install the GLP Administration application. Your system administrator will then need to set up the application to control the other Cary Eclipse applications.

Note

Selecting Typical (the default setting) at installation will not install the GLP Administration application and all applications will have unrestricted access. You can install only the GLP Administration application at any stage by using the Custom installation option.

After installation, the system administrator must assign a user name, workgroup and password to every person that will be running a Cary Eclipse application. The system administrator must also set up the various options available for each workgroup for every nominated Cary Eclipse application.
To access the GLP Administration Application, select the Windows Start button, point to Programs, Cary Eclipse and then select GLP Administration.

Software controls

The GLP Administration Application comprises the following tabbed pages:

- Users
- Privileges
- Groups
- Applications

The System Information Application also contains the following menus:

- View
- Help

See also:
GLP Administration Home Page
GLP Administration How to

GLP Login—GLP Administration

This dialog requires the administrator to enter an administration name and password to start the GLP Administration application.

Application Name
The application into which you are logging is listed beside here.

User Name
Enter your Administrator user name to login to the GLP Administration application. The default password is 'Admin', and must be typed exactly as shown.

Password
Enter your current Administrator password. The default password is 'Admin', and must be typed exactly as shown.

Note
You are required to enter the correct password before you can designate a new password. If you forget the password, please send us an email at fluorescence@osi.varianinc.com and we will try to help you.

New Password
Check this box to nominate a new password. You will be required to enter your new password.

(Confirmation)
Re-enter your new password to confirm its nomination.

Note
GLP Log—GLP Administration

This dialog lists all the changes made to GLP privileges since the application was installed. You cannot reset this list.

Print
Press this button to display the Windows Print Setup dialog which allows you to print the list.

Menu line—GLP Administration

- View menu
- Help menu

View menu—GLP Administration

Use the View menu to display the following.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hints</td>
<td>Select this option to toggle the Hints bubble text on and off. The bubble text appears when you place the arrow cursor over a field, button or text entry field for a short period.</td>
</tr>
<tr>
<td>Log</td>
<td>Select Log to display a read-only information dialog that tracks the changes made to GLP Privileges. The log will display the current privilege status of each User and Group for each nominated Cary Eclipse application.</td>
</tr>
</tbody>
</table>

Help menu—GLP Administration

Use the Help menu to view the online Help and software information for this application.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>GLP Administration Help</td>
<td>Select this to display the GLP Administration Home Page. This contains a list of online Help topics specific to the GLP Administration Application.</td>
</tr>
<tr>
<td>Help Topics</td>
<td>Select this to display the Cary Eclipse Windows Online Help Home Page.</td>
</tr>
<tr>
<td>About…</td>
<td>Select About to access a dialog that displays information about the application as well as Varian email and Internet contact information. To clear this About dialog, select OK.</td>
</tr>
</tbody>
</table>

Users—GLP Administration

The Users page enables you to nominate or alter the identity and privileges of a Cary Eclipse instrument operator.

The Users page is displayed when you open the GLP Administration Application. To access the GLP
Administration Application, click the Windows **Start** button, point to **Programs, Cary Eclipse** and then select **GLP Administration**.

Once you are in the application, you can access the Users page at any time by selecting the **Users** tab.

The page comprises the following:

**Status table**

This table identifies the current people allowed to use the Cary Eclipse applications. You must select the appropriate user before making any changes.

- **Note**
  - A □ in the first column of the table indicates the currently nominated user. Any modifications you make to the User Name, Group Name or Suspended status will be made to this nominated user only.

  - **User Name**: This field displays the names of all current operators or users. Use the **Add** or **Modify** buttons to add to or modify this list of users.
  - **Group Name**: This field displays a workgroup to which each user is assigned. Use the **Modify** button to change the workgroup that a user belongs to.
  - **Suspended**: This field displays the access rights of the user. The suspended status is either True or False. If a user has a Suspended status of True, then that user cannot run any Cary Eclipse applications. Use the **Modify** button to change the access rights of users.

**Software controls**

- **Add…**: Select the Add button to display the **Add User dialog** where you can add a new user.
  - **Note**: You can only add one new user at a time.

- **Modify…**: Select the Modify button to display the **Modify User dialog** where you can modify the nominated group or password of an existing user.

- **Delete…**: Select **Delete** to remove the nominated user in the status table. You will be prompted to confirm your action.

- **Bypass GLP checks**: Select this box to turn off the GLP password protection, access and privilege status requirements for operation of all Cary Eclipse applications.

**Add User—GLP Administration**

Use the Add User dialog to do the following:

- Add a new user
- Assign a new user to a group
- Nominate a new user password
- Set the user privilege status

The Add User dialog is accessible from the Users page by selecting **Add…**
The dialog comprises the following:

**User Name**  
Enter the name of the new user in this field. Note that you can have groups consisting of only one user. This enables you to set privileges for each individual user rather than groups of users.

**Group Name**  
Assign a user to a workgroup by selecting one of the following options:

- **Existing**  
  Select *Existing* to add a new user to an existing workgroup. To do this, simply select the workgroup from the existing list.

- **New**  
  Select *New* to create a new workgroup name.

*Note*  
A new workgroup can also be added by selecting *Add…* from the [Groups page](#).

- **Same As User Name**  
  Select this option to add a new workgroup that will have the same name as the new user name.

**Password**  
Enter a password for the new user. The password may be up to 16 characters long.

*Q.* Why does the Cary Eclipse gives me an error message when I select OK on the Add User dialog?  
*A.* You may not have entered and confirmed a password for the new user. You **must** set a password for a new user or the Cary Eclipse will not accept the new user details.

*(Confirmation)*  
You must re-enter the new password in this field to confirm your nomination.

**Suspend**  
Select *Suspend* to suspend the access rights of the nominated user. Once suspended, the user will be unable to run any Cary Eclipse application. To restore a user’s privileges, select *Modify* ([Modify User](#)) from the Users page and clear the Suspend check box.

### Modify User—GLP Administration

Use the Modify User dialog to modify the following:

- The name of a current user
- The group to which the user is assigned
- The user password
- The user privilege status

The dialog comprises the following:

**User Name**  
This read-only field displays the name of the user whose details are to be modified.

**Group Name**  
You can assign the user to a new group simply by selecting a Group Name from the list.

**Password**  
If required, enter a new password for the user in this field.

*(Confirmation)*  
Re-enter the new password in this field to confirm it.
Suspend

Select Suspend to suspend access to Cary Eclipse applications, or clear the check box to restore access.

Privileges—GLP Administration

Use the Privileges page to add or restrict a workgroup’s access to Cary Eclipse applications.

The Privileges page is accessible from the GLP Administration Application by selecting the Privileges tab. To access the GLP Administration Application, click the Windows Start button, point to Programs, Cary Eclipse and then select GLP Administration.

The page comprises the following:

Note

A □ in the first column of the table indicates the currently nominated Group Name. Any modifications you make to the User Name or Application Name status will be made to this nominated user only.

Status table

This table identifies the current groups nominated to use each Cary Eclipse application. Select the appropriate group to make changes to their privileges.

Note

The GLP Administration application automatically identifies the software applications that have been installed.

Group Name

The Group Name field displays a list of the current workgroups. Group names are repeated when more than one application is available to a group.

App Name

This field identifies the applications that are available to members of that workgroup.

Privilege check boxes

Select these check boxes to nominate the privilege level for each selected group and application. The available check boxes will vary depending upon the application. Select the application you are interested in to view its privileges.

- ADL Shell
- Advanced Reads
- Align
- Concentration
- Kinetics
- Lifetimes
Commands

- **Cancel Changes (Esc)**: Click **Cancel Changes** to cancel any modifications you have made to the privilege nominations or a group.
- **Apply Changes Now**
- **Add…**
- **Delete…**
- **Bypass GLP Checks**: Select **Bypass GLP Checks** to turn off the GLP password protection, access and privilege status requirements for operation of all Cary Eclipse applications.

See also: [Add Privileges](#)

**Add Privilege—GLP Administration**

Use the Add Privileges dialog to add your privileges to an additional application.

**Note**
Each application has to be individually assigned to each group.

The dialog comprises the following:

- **Group Name**: Select the required group. If the group you want does not already exist, clear this dialog and add a new User to the [Users page](#) or add a new Group to the [Groups page](#).
- **Application Name**: Select the Cary Eclipse application and select **OK** to assign the application to the selected group.

**Groups—GLP Administration**

Use the Groups page to nominate or alter the identity and privileges of a workgroup.

The Groups page is accessible from the [GLP Administration Application](#) by selecting the **Groups** tab. To access the GLP Administration Application, click the Windows **Start** button, point to **Programs**, **Cary Eclipse** and then select **GLP Administration**.

The page comprises the following:

**Note**
A in the first column of the table indicates the currently nominated Group Name. Any
modifications you make to the Group Name or Suspended status will be made to this nominated user only.

**Status table**

This table identifies the current groups and their access rights. You can select the appropriate group before making any changes to their privileges.

**Group Name** This field identifies all currently defined workgroups.

**Suspended** This field identifies the access rights for each group. The suspended status is either True or False. If a group has a Suspended status of True, then that group cannot run any Cary Eclipse applications.

**Commands**

- **Add…** Click **Add** to display the Add Group dialog where you can add a new group.
- **Modify…** Click **Modify** to display the Modify Group dialog where you can alter the access rights of the selected group.
- **Delete…** This button deletes the group highlighted in the status table.

**Note**

To avoid deleting a required group, you must first delete all Users from that group (on the Users tab), before deleting it.

**Bypass GLP Checks** Select **Bypass GLP Checks** to turn off the GLP password protection, access and privilege status requirements for operating all Cary Eclipse applications.

**Add Group—GLP Administration**

Use the Add User dialog to do the following:

- Add a new group
- Change the group's privilege status

The Add Group dialog is accessible from the Groups page by selecting **Add…**.

The dialog comprises the following:

**Group Name** Enter the name of the new workgroup.

**Suspend** Select **Suspend** to suspend the access rights of the nominated group. Once suspended, any users in the group will be unable to run any Cary Eclipse application. To restore a group’s privileges, select **Modify** ([Modify Group](#)) from the Groups page and clear the Suspend check box.
Modify Group—GLP Administration

Use the Modify Group dialog to modify the nominated group’s privilege status.

The dialog comprises the following:

**Group Name**  This read-only field displays the name of the group whose details are to be modified.

**Suspend**  Select **Suspend** to suspend access to Cary Eclipse applications, or clear the check box to restore access.

Applications—GLP Administration

Use the Applications page to nominate or alter the list of Cary Eclipse applications under the control of the GLP Administration application.

**Note**

All Cary Eclipse applications are nominated for control by the GLP Administration application at the time of installation.

The Applications page is accessible from the GLP Administration Application by selecting the Applications tab. To access the GLP Administration Application, click the Windows **Start** button, point to **Programs, Cary Eclipse** and then select **GLP Administration**.

The page comprises the following:

**Note**

A in the first column of the table indicates the currently nominated Group Name. Any modifications you make to the User Name or Application Name status will be made to this nominated user only.

Status table

This table identifies the current applications nominated at administration level. Select the appropriate application before making any changes. The table contains the following:

**App Name**  This field identifies all available Cary Eclipse applications.

Commands

**Add…**  Click **Add** to display the Add Application dialog where you can add a new application to GLP Administration.

**Delete…**  Click **Delete** to remove the highlighted application from GLP Administration (after requesting confirmation).

**Note**

To avoid deleting a required application, you must first delete the application anywhere it appears in the Privileges table.

**Bypass GLP**  Select **Bypass GLP Checks** to turn off the GLP password protection, access and
Checks privilege status requirements for operation of all Cary Eclipse applications.

See also: Privileges

Add Application—GLP Administration

Use the Add Application dialog to control the addition of a Cary Eclipse Application to GLP Administration.

The Add Application dialog is accessible from the Groups page by selecting Add…

The dialog comprises the following:

<table>
<thead>
<tr>
<th>Application Name</th>
<th>Insert the application name in this field.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>The Cary Eclipse application names that you can enter are:</td>
</tr>
<tr>
<td></td>
<td>• ADL Shell</td>
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<tr>
<td></td>
<td>• Advanced Reads</td>
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<td>• Kinetics</td>
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<td></td>
<td>• System Information</td>
</tr>
<tr>
<td></td>
<td>• Validate</td>
</tr>
</tbody>
</table>

GLP Administration How to

Select the procedure you require below for a step-by-step informative guide on running the GLP Administration software:

- How to run GLP for the first time and set up an Administrator password
- How to create a workgroup
- How to remove or restore a workgroup's access to the Cary Eclipse software
- How to assign User Names and Passwords
- How to modify User Names and Passwords
- How to change the privileges of a group for a Cary Eclipse application

How to run GLP for the first time and set up an Administrator password—GLP Administration

A default password is provided for the GLP Administration Application. However, you should change this password to prevent unauthorized access to the application's settings.

Step 1
GLP Administration Home Page

Click the Windows Start button, point to Programs, Cary Eclipse and then select GLP Administration. The GLP Administration Application and the GLP Login dialog are displayed.

Step 2

As this is the first time you have run GLP, the User Name field will display the word 'Admin'. Enter Admin in the Password field.

Note
Ensure that you enter the password correctly, as it is case-sensitive.

Step 3

Select the New Password check box. A New Password and Confirmation field will be displayed to enable you to change the administrator access to GLP from the default settings.

Step 4

Enter a new password in the field next to the New Password check box.

Step 5

Confirm the new password in the (Confirmation) field.

Step 6

Select OK to change the administrator password for the GLP application and open the main window of the application.

Note
If you forget Administrator password, please send us an email at fluorescence@osi.varianinc.com and we will try to help you.

See also:
How to create a workgroup
How to remove or restore a workgroup's access to the Cary Eclipse software
How to assign User Names and Passwords
How to modify User Names and Passwords
How to change the privileges of a group for a Cary Eclipse application

How to create a workgroup—GLP Administration

Step 1

From the GLP Administration Application, select the Groups tab. The Groups page is displayed.

Step 2

Select Add…. The Add Group dialog is displayed.
Step 3

Enter a new group name in the **Group Name** field. Select **Suspend** if you want to suspend the access privileges of the new group.

Step 4

Select **OK** to add the new group.

**Note**

You should now assign the group's privileges. See [How to change the privileges of a group for a Cary Eclipse application](#).

**How to remove or restore a workgroup's access to the Cary Eclipse software—GLP Administration**

Step 1

From the GLP Administration Application, select the **Groups** tab. The **Groups** page is displayed.

Step 2

Highlight the required group in the table. A □ in the first column of the table indicates the currently nominated group.

Step 3

Select **Modify**. The **Modify Group** dialog is displayed.

Step 4

Select the **Suspend** check box to suspend the access privileges of a group, or clear the check box to restore a group’s access.

Step 5

Select **OK** to implement the change you have made to the group's access.

See also:

- [How to create a workgroup](#)
- [How to assign User Names and Passwords](#)
- [How to modify User Names and Passwords](#)
- [How to change the privileges of a group for a Cary Eclipse application](#)

**How to assign user names and passwords—GLP Administration**

Step 1

From the GLP Administration Application window, select the **Users** tab. The **Users** page is displayed.
Step 2

Select **Add**. The **Add User** dialog is displayed.

Step 3

In the **User Name** field, enter the new user name.

Step 4

From the **Group Name** field, select the group that the name will be assigned to by doing one of the following:

a. Select **Existing** to add User Name to an existing group. You then need to highlight one of the existing group names in the list box.

b. Select **New** to create a new group. You then need to enter the name of the group in the field.

c. Select **Same As User Name** to create a new group. In this case, the group will have the same name as the entered User Name.

**Note**

A new group can also be added by selecting the **Groups** tab and selecting **Add** from the **Groups** page.

Step 5

Enter the password for the new user in the **Password** field.

Step 6

Confirm the new password in the **(Confirmation)** field.

Step 7

Select **Suspend** if you want to suspend the access privileges of the new user.

Step 8

Select **OK**. The new user will be added to the system.

**Note**

If the user has been added to a new group, you should now assign the group's privileges. To do this, refer to How to change the privileges of a group for a Cary Eclipse application.

How to modify user names and passwords—GLP Administration

Step 1
From the GLP Administration Application, select the **Users** tab. The Users page is displayed.

**Step 2**

Highlight the required user information in the table. A ✅ in the first column of the table indicates the currently nominated user.

**Step 3**

Select **Modify**. The **Modify Group** dialog is displayed.

**Step 4**

You can do one or more of the following:

- To change the user's group, select a new group in the **Group Name** list box.
- To change the user's password, enter a new password (and confirmation) in the **Password** and **(Confirmation)** fields.
- To modify a user's access privileges, select or clear the **Suspend** check box.

**Step 5**

Select **OK** to implement the changes you have made to the user's access.

See also:
- How to create a workgroup
- How to assign User Names and Passwords
- How to remove or restore a workgroup's access to the Cary Eclipse software
- How to change the privileges of a group for a Cary Eclipse application

**How to change the privileges of a group for a Cary Eclipse application—GLP Administration**

**Step 1**

From the GLP Administration Application, select the **Privileges** tab. The **Privileges** page is displayed.

**Step 2**

Select **Add**. The **Add Group** dialog is displayed.

**Step 3**

Highlight a nominated group and application, and select **OK** to exit the Add Privilege dialog.

Note that the applications are automatically loaded at installation. Select the **Applications page** to add or delete applications from GLP.

**Q.** Why do I get a message when I try to add an application to a group?  
**A.** You cannot add an application to a group that already has that application listed as a privilege.
Step 4

On the Privileges page, select the required check boxes to nominate the privilege level for the selected group and application. Privilege levels are assigned for running each application.

Step 5

Click Apply Changes Now to confirm the group’s privilege status or Cancel Changes to cancel privilege changes. Alternatively you can select the ESC key on your keyboard to cancel the changes.

See also:
- How to create a workgroup
- How to remove or restore a workgroup's access to the Cary Eclipse software
- How to assign User Names and Passwords
- How to modify User Names and Passwords
Kinetics Home Page

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**Kinetics Application**

**Introduction**

Use the Kinetics application to measure the increase or decrease in emission intensity as a function of time. From this you can obtain an intensity versus time plot. The features of the application include:
• Calculation of Zero order, First order and Second order reaction rates from intensity versus time data.
• Selectable time window for calculation.
• Overlay of the best-fit line on raw data.
• Automatic or manual estimates for the first order and second order Marquardt fitting.

Hot Tip:
The Kinetics application enables you to determine whether a single wavelength is changing over time. If you wish to collect scans as a function of time in order to determine whether the spectrum is changing, use the Cycle mode option in the Scan application. The Cycle mode option is available from the Options page of the Setup dialog.

To access the Kinetics Application, click the Windows Start button, point to Programs, Cary Eclipse and then select Kinetics.

Software controls

The application comprises the following:

**Instrument Status Readings**
The Instrument Status Readings, which display the Y and X values of the graph axes are displayed in the top left and right corners of the Kinetics Application Window.

- **Y value**
  Displays the current Y value, usually intensity. This value is continually updated during a data collection.

- **X value**
  Displays the current X value, which is the current reading for the Time. On the Cary page of the Setup dialog, choose whether to display the time value in minutes or seconds.

**Start and Stop buttons**
The Start and Stop buttons at the top of the Kinetics Application window are used to start and stop instrument analyses. These buttons can also be accessed from the Commands menu.

Tell me more about the Start and Stop buttons

**Graphics area**
The Graphics area refers to the plotting area of the application. It can contain traces of collected data, labels and pictures. In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area defined by the axes is referred to as a graph box.

To display or hide the Graphics area, select the Graphics command on the View menu. Double-clicking on the Graphics area enables you to toggle between half or full size display sizes. To manually size the Graphics area, place the mouse cursor over the border between the Graph and Report areas until the cursor changes to a double arrow (mouseover icon). Then hold down the mouse and drag it up or down to resize the areas.

Tell me more about the Graphics area

**Report area**
Use the Kinetics Report area to view the current report. The report may include the results of your data collection, the method parameters used to collect the results and any graphics you have included. You can select the information to include in your report in the Reports tab of the Setup dialog.
To display or hide the Report area, select the Report command from the View menu. Double-clicking on the Report area enables you to toggle between half or full size display sizes. To manually size the Report area, place the mouse cursor over the border between the Graph and Report areas until the cursor changes to a double arrow (↕). Then hold down the mouse and drag it up or down to resize the areas.

You can copy the generated report onto the Windows clipboard. To do this, highlight the report (or a relevant section of it) and press CTRL+C. You can then paste the report into a word processor such as Microsoft® Word.

**Hot Tip**
You can edit the Kinetics Report by pressing CTRL+R or selecting the **Edit Report** command from the **Edit** menu.

**Commands**
The Kinetics Application window contains the following commands, all of which can also be accessed from the various menus in the application.

Click on the buttons to view information about them:

- **Setup**
- **Zero**
- **Pre-scan**
- **Set temp**
- **Stop time**
- **Clear report**
- **Recalculate**
- **Print**

**Hot Tip**
By becoming familiar with these commands (Setup, Zero, Pre-scan, etc.), you can remove these buttons from the Kinetics window. This will enlarge the Report area and make it easier to view results. To do this, clear the Buttons item in the View menu.

**Menus**
The Kinetics Application contains the following menus:

See also:  
**File, Edit, View, Commands, Setup, Graph, Help**

**Status Display**
**Status Line**
**Keyboard function keys**
**Kinetics home page**

**Validation of your Cary Eclipse for the Kinetics application**

The following files are shipped with the Kinetics application to enable you to validate the software. The files are stored on your CD in a Validation subdirectory.

To validate your software, you should Open each file in turn and select the Recalculate button. The results calculated by the Cary Eclipse should match those given in the validation files listed below. These results were determined by exporting the data to a spreadsheet program and performing the same calculation as that used in the Kinetics application.

- **Kinetics validate_first order fit.FBKN**
- **Kinetics validate_second order fit.FBKN**
- **Kinetics validate_zero order fit.FBKN**

**Getting ready for a run—Kinetics**
Setup dialog
Setup menu
Cary page
User collect example
Bio-/Chemi-luminescence and Phosphorescence Options dialog
Gate Time for Bio-/Chemi-luminescence
Gate Time for Phosphorescence
How many flashes?
Options page
About smoothing
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Multicell/Temperature accessory
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Display Options dialog
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Go To Well dialog
Analyze page
Rate calculations
Reports page
Report toolbar
Clear Report
How to use the advanced reporting options
Auto Store page
How to use the advanced reporting options
Auto Store page
How to email the results

Setup—Kinetics

Use the Setup dialog to set up your instrument and accessories for a Kinetics run. You can also use the dialog to set up the Analysis, Report and Storage parameters associated with the run.

To access the Setup dialog select the Setup button from the Kinetics Application window or the Setup command from the Setup menu.

The Setup dialog comprises the following tabbed pages:

- Cary
- Options
- Accessories
- Analyze
- Reports
- Auto Store

See also:
Kinetics Application

Setup menu—Kinetics

Use the Setup menu to access the parameters for setting up a Kinetics read.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Choose **Setup** to display the **Setup** dialog where you can set up your instrument for a Kinetics measurement. The Setup dialog can also be accessed from the Setup command button.

Choose **Cary** to display the **Cary** page. You can use this page to set up the instrument parameters associated with the data collection.

Choose **Options** to display the **Options** page where you can set up your filters and set the detector voltage. You can also use this page to select the way the collected data is displayed in the Graphics area, and the type of smoothing applied (if any).

Choose **Accessories** to display the **Accessories** page. You can use this page to set up the accessory parameters associated with the data collection.

Choose **Analyze** to display the **Analyze** page where you can select from various mathematical equations and set the parameters used to generate the calculation of reaction rates.

Choose **Reports** to display the **Reports** page. You can use this page to set up the report parameters associated with data collection.

Choose **Auto Store** to display the **Auto Store** page where you can specify if and when data will be stored, and in what format.

---

**Cary—Kinetics**

Use the Cary page to set up the instrument parameters associated with the data collection.

From the **Setup** menu, select **Cary** to access the **Cary** page. You can also access the page from the **Setup** dialog by selecting the **Cary** tab. To access the **Setup** dialog, select the **Setup** button from the Kinetics Application window.

**Instrument setup**

**Data mode**

**Fluorescence**
Fluorescence refers to the emission of light from an excited single state. The fluorescence signal is measured when the lamp flashes.

**Bio-/Chemiluminescence**
Bio-/Chemiluminescence refers to bio- or chemi-luminescence, where the emission is generated via a chemical or biochemical reaction.

**Phosphorescence**
Phosphorescence refers to the emission of light from an excited triplet state. Phosphorescence is measured after the lamp has stopped flashing.
Wavelength setup

Multiwavelength

Use Multiwavelength to monitor more than one wavelength in your reaction. If you select Multiwavelength, the following options will be available:

**Number of wavelengths**

Use the Up/Down arrows to enter the number of emission and excitation wavelengths you wish to monitor. You can monitor up to six at one time. The number of rows in the Multiwavelength table will be expanded to match the number of wavelengths selected.

**Multiwavelength table**

Use the Multiwavelength table to enter the excitation and emission wavelengths to be monitored for each wavelengths. The number of rows in the Multiwavelength table will correspond the number of wavelengths selected. If you have selected more than two wavelengths, use the arrows to the right of the table to access all of the wavelength rows.

If you have selected Bio-/Chemi-luminescence as the Data mode, the Multiwavelength table will display the Em.wavelength only, as Ex. wavelengths are not applicable in this mode.

**Note**

The Multiwavelength option is not available if you have selected the Polarizer on the Accessories page. You can select the Multiwavelength option and then select the Polarizer on the Accessories page, however you will only be able to select Other as the Polarization Measurement Type.

**Ex. Wavelength**

Set the wavelength of the excitation monochromator, which is the wavelength value at which you want to excite your sample. Click the arrow to the right of the field to quickly select the most recent wavelength entries. The Ex. wavelength option is not available if you have selected Multiwavelength setup.

Alternatively, to set the excitation monochromator to zero order (white light), from the Commands menu, point to Zero Order and select Excitation. A tick next to this option indicates that it is selected. Clear this option to return to monochromator light and use the value set in the Ex. Wavelength field.

The Ex. Wavelength option is not available if you have selected Bio-/Chemi-luminescence as the Data mode.

**Em. Wavelength**

Set the wavelength of the emission monochromator, which is the wavelength value at which your sample emits. Click the arrow to the right of the field to quickly select the most recent wavelength entries. The Em. wavelength option is not available if you have selected Multiwavelength setup.

Alternatively, to set the emission monochromator to zero order (white light), from the Commands menu, point to Zero Order and select Emission. A tick next to this option
Collect timing indicates that it is selected. Clear this option to return to monochromator light and use the value set in the **Em. Wavelength** field.

**User Collect**
Select User Collect to display the User Collect field. The **Ex. Wavelength** and **Em. Wavelength** fields will not be available. Note that this option is not available if you select **Multiwavelength** mode.

Use the **User Collect** entry field to enter an ADL command which applies a mathematical expression to the intensity value collected. The most recent user collect entries will be saved in the drop down list to a maximum of 10.

The Syntax for ADL commands is:

```
Read (Ex. wavelength, Em. Wavelength)
```

The **User Collect** option is not available under the following conditions:

- if you have selected **Multiwavelength** setup, or
- if you have selected the **Polarizer** on the Accessories page

**Note**
You can select the **User Collect** option and then select the **Polarizer** on the **Accessories** page, however you will only be able to select **Other** as the Polarization **Measurement Type**.

**Ex. Slit (nm)**
Set the slit width of the excitation monochromator. The **Ex. Slit** option is not available if you have selected **Bio-/Chemi-luminescence** as the Data mode.

*As a general rule:*
At a fixed emission slit width, increasing the excitation slit width will result in an increase in the fluorescence excitation intensity.

**Em. Slit (nm)**
Set the slit width of the emission monochromator.

*As a general rule:*
At a fixed excitation slit width, a two-fold increase in the emission slit width will result in a four-fold increase in the emission intensity. Accordingly, a two-fold decrease in the emission slit width will result in a four-fold decrease in the emission intensity.

**Collect timing**

**Simple collect**
Select this option to set the **Number of Stages** to '1'. Use one stage if you require only one collection rate for the entire kinetics run.

**Advanced collect**
Check this option to enable the **Number of Stages** entry field and specify the collect time profile of a run, enabling you to vary the rate at which data is collected over the period of the run. Enter the times required in the fields in the **Cycles** table.

**Q.** When do I choose an Advanced Collect?

**A.** The **Advanced Collection** option allows you to divide the length of the run into different sections or ‘stages’. You can nominate a different data collection rate for each stage. This means that you can collect lots of data points during the stage when the reaction is happening quickly and then reduce the number of data points collected...
when the intensity change is slower.

**No. of stages**
Use this to set the number of time segments into which you wish to divide the total data collection time, each having a different data collection rate. The Cycles table will expand to fit the number you enter.

**Q. Why isn't the Number of Stages entry field active?**

**A.** You have selected Simple Collect. On selecting Advanced Collect, the field will be activated.

**X Mode**
This enables you to set the units of time for the abscissa (or X axis).

Choose your desired abscissa mode by selecting one of the following buttons:

**Min**
Select this to time the run in minutes.

**Sec**
Select this to time the run in seconds.

**Ave time (s)**
Set the amount of time, in seconds, for which data is averaged. To do this, enter the amount in the Ave. time field, or use the up/down arrows to the right of the field to increase or decrease the time. The averaged result is displayed in the report.

When setting the Ave Time, consider the following:

- The longer the Ave Time, the longer the signal will be averaged
- A longer Ave Time will result in more precise results as the averaging of the signal reduces any contribution of background ‘noise’.

Averaging is calculated as follows:

**For fluorescence:**
the number of readings averaged are taken from consecutive flashes at 80 Hz where:
number of consecutive flashes = averaging time/0.0125.

**For phosphorescence:**
the intensity readings are taken from different decays, where:
number of decays = averaging time/gate time.

**For bio-/chemi-luminescence:**
a signal is collected every Gate Time, and (averaging time/gate time) signals are averaged at each data interval

**Cycles Table**
This table enables you to vary the data collection rates within the one kinetics run. This feature is particularly useful when analyzing slow reactions and multi-step reactions.

**Stage**
This column displays the stage number/s.

**Cycle**
Enter a number in this numeric field to set the rate at which the system will cycle the
specified cell setup.

**Stop**
Type a number in this numeric field to set the **Stop Time** to establish how long you require a kinetics analysis to continue.

**Note**
**Stop Time** overrides **Cycle Time** so make sure that the **Stop Time** is long enough to allow the measurement of all Cycles.

**Status Display**
Select this check box to display the **Status Display** window, where you can view the status of various parameters during the data collection.

*See also:*
- **Options**
- **Accessories**
- **Example of a User Collect**
- **Example using the Stop time**
- **Example of a multi-stage Kinetics run, without Dwell time**
- **Example of a multi-stage Kinetics run, using Dwell time**
- **Status Display**

**Example of a User Collect—Kinetics**

You can extend the functionality of your Cary Eclipse instrument by using the built-in Read command. This command enables you to change the instrument to a different wavelength and take a reading. You can combine this command with the mathematical operators: ‘+’, ‘-’, ‘*’ and ‘/’. This is illustrated in the example below.

**Read(340, 400)-Read(340, 450)**
This reads the intensity at Excitation monochromator = 340, Emission monochromator = 400 and subtracts the intensity read at Excitation monochromator = 340 and Emission monochromator = 450. This reading is performed at each data point.

The **User collect** command can be selected on the **Cary** page of the **Setup** dialog. Enter the Read command in the **User collect result** field.

*See also:*
- **Cary**

**Bio-/Chemi-luminescence/Phosphorescence Options—Kinetics**

Use the **Bio-/Chemi-luminescence/Phosphorescence Options** dialog to set up the parameters associated with these two measurement modes.

To access the **Bio-/Chemi-luminescence/Phosphorescence** dialog, select the **Options** button from the **Cary** page.
Dialog controls

The Bio-/Chemi-luminescence/Phosphorescence Options dialog comprises the following options:

**Total decay time**
Enter, in seconds, the total decay time for your sample. The total decay time is the time that it takes for the phosphorescence signal to decay to dark levels (zero). The lamp will not flash again until the total decay time has elapsed.
The range is 0.001–1800.000 s.

*Note*
This option is not available in Bio-/Chemi-luminescence mode.

**No. of flashes**
Enter the number of lamp flashes required, which is how many times the lamp flashes consecutively before the delay time is counted.

If you are measuring phosphorescence samples that have a relatively long decay time (that is, a number of seconds), you can setup the Cary Eclipse to pulse the lamp multiple times, as this will increase the intensity of the phosphorescent signal. For short lived phosphorescent samples, multiple flashes will not impact on the intensity of the signal, and therefore only one flash is required.

*How many flashes should I perform?*

*Note*
This option is not available in Bio-/Chemi-luminescence mode.

**Delay time**
Enter, in milliseconds, the delay time for your sample. The delay time is the time that elapses between the last flash and the beginning of data collection. The default value is 0.100 ms, which is the time it takes for the lamp intensity to decay to zero. Setting a delay time ensures that there is no lamp effects in the phosphorescence decay curve.
The range is 0.000–5000.000 s.

*Note*
This option is not available in Bio-/Chemi-luminescence mode.

**Gate time**
Enter, in milliseconds, the gate time for your sample. The gate time is the overall reading time of the emission signal.
The range is from 10µs–10s

Understanding Average Time and Gate Time for Phosphorescence scans
Understanding Gate Time for Bio-/Chemi-luminescence scans

Example of the Bio-/Chemi-luminescence/Phosphorescence Options
Understanding Gate Time for Bio-/Chemi-luminescence scans—Kinetics

**Gate Time** is set on **Options** page, which is accessible from the **Cary** page.

The following diagram highlights the **Gate Time** setting.

The **Gate Time** is set to 10 and the **Intensity** is equal to $I_T$. 

---

**See also:**
[Cary page](#)

---

![Diagram showing Phosphorescence options and intensity over time](image)
For Phosphorescence scans, there is a relationship between Average Time (the time for which data is averaged) and Gate Time (the overall reading time of the emission signal).

Averaging is calculated as follows:

The data point at each wavelength will be the average of the signals from a number of gate time measurements that occur during this averaging time, that is:

\[
\text{Averaging Time/Gate Time}
\]

These signals are accumulated from successive decay curves so that the actual time taken to generate a point at a single wavelength will be approximately:

\[(\text{Averaging Time/Gate Time} \times \text{Total Decay Time})\]

In phosphorescence mode, the Averaging Time ranges from the Gate Time to a maximum of 1000 s.

**Note:**

*Gate Time* is not used if you select *Fluorescence* as the *Data Mode*. 

See also:

- Understanding Average Time and Gate Time for Phosphorescence scans
- Kinetics Application
- Cary page
- Options dialog

**Understanding Average Time and Gate Time for Phosphorescence Scans—Kinetics**
**Average Time** is set on the **Cary** page of the **Setup** dialog. **Gate Time** is set on **Options** page, which is accessible from the **Cary** page.

The following diagrams highlight the relationship between **Average Time** and **Gate Time**.

**Example 1**

When the Average Time and Gate Time are set to the same value:

<table>
<thead>
<tr>
<th>Average Time/Gate Time</th>
<th>1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intensity</td>
<td>1 (Area ( I_A, I_B ))</td>
</tr>
</tbody>
</table>

where:

- Total Decay Time 20 ms
- Delay Time 10 ms
- Cycle Time 1 sec
- Stop Time 3 sec
- Average Time 5 ms
- Gate Time 5 ms

**Example 2**

When the Average Time is set to five times the Gate Time:

| Average Time/Gate Time | 5          |
where:

Total Decay Time \hspace{1cm} 20 \text{ ms}

Delay Time \hspace{1cm} 10 \text{ ms}
How many flashes should I perform?—Kinetics

The Number of Flashes option is set in the Bio-/Chemi-luminescence/Phosphorescence Options dialog. To access the dialog, select the Options button from the Cary page.

Note
The Options button is only visible on the Cary page if you have selected Bio-/Chemi-luminescence or Phosphorescence as the Data mode.

If you are measuring phosphorescence samples that have a relatively long decay time (that is, a number of seconds), you can setup the Cary Eclipse to pulse the lamp multiple times, as this will increase the intensity of the phosphorescent signal (refer to Example 1, below). For short lived phosphorescent samples, multiple flashes will not impact on the intensity of the signal, and therefore only one flash is required (refer to Example 2, below).

Example 1
The above example shows that the sample does not decay before the instrument lamp flashes again. Therefore setting multiple flashes will result in an increase in the intensity of the signal.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cycle Time</td>
<td>1 sec</td>
</tr>
<tr>
<td>Stop Time</td>
<td>3 sec</td>
</tr>
<tr>
<td>Average Time</td>
<td>25 ms</td>
</tr>
<tr>
<td>Gate Time</td>
<td>5 ms</td>
</tr>
</tbody>
</table>

See also:
Understanding Gate Time for Bio-/Chemi-luminescence scans
Kinetics Application
Cary page
Options dialog
Example 2
The above example shows that the sample decays in less time than the time it takes the instrument to flash the lamp again. Therefore, setting more than 1 flash, in the No. of Flashes field will not increase the intensity of the signal.

See also: Bio-/Chemi-luminescence/Phosphorescence Options

Options—Kinetics

Use the Options page to set up your filters and set the detector voltage. You can also use this page to select the way the collected data is displayed in the Graphics area, and the type of smoothing applied (if any).

From the Setup menu, select Options to access the Options page. You can also access the page from the Setup dialog by selecting the Options tab. To access the Setup dialog, select the Setup button from the Kinetics Application window.

Display Options

Overlay traces
Select this option to superimpose the collected data of each sample in the run in one graph box. If you select this option, a graph box is created at the start of the collect in the Graphics area and each sample’s data is overlaid in that box as the data is collected. Traces will always be overlaid in the same graph, regardless of what multicell/well accessory is used. Clear this option to display all of the traces in individual graphs.

Y minimum/Y maximum
Enter the minimum and maximum Y values to be displayed on the graph axis. The software will update the Y axes to reflect these settings during the kinetics run.

Smoothing
Select Smoothing to reduce the noise of your collected data. When you select this option, smoothing will be applied to the ratioed intensity signal using the algorithms defined below. You can select one of the three types of smoothing that can be performed during a run. Only the smoothed trace is displayed during data collection, however the ratioed unsmoothed data is also saved with the experiment.

Type
Savitzky-Golay
This algorithm will take the number of Filter points, apply the Savitzky and Golay smoothing algorithm and display the smoothed value at the middle X-point of the filter. The naming of the smoothed trace will be ‘Sample name_SG’. 
Tell me more about Savitzky-Golay smoothing

**Boxcar average**
This algorithm will take the number of points as defined by the Filter size, and average these, displaying the average at the middle X-point of the filter. The naming of the smoothed trace will be 'Sample name_Box'.

Tell me more about Boxcar averaging

**Moving average**
This algorithm will take the number of points as defined by the Filter size, and average these, displaying the average at the middle X-point of the filter. The naming of the smoothed trace will be 'Sample name_Mov'.

Tell me more about Moving average

**Filter size**
Enter the Filter size, which determines the number of points that will be used in the smoothing procedures. The higher the filter size (and thus the number of points used), the greater the amount of smoothing that will be performed.

Filter sizes must be odd values. For Boxcar and Moving averages, the range is 3–101. For Savitzky-Golay smoothing, the range is 5–101.

**Excitation filter**
Set the excitation filter for the Kinetics measurement. To do this, select the down arrow to the right of the field and select an option from the list.

A range of filter settings are available:

*Auto*
If you select the Auto option, the filter wheel will be automatically moved to the appropriate position for the selected excitation wavelength. It is recommended that you set the filter to Auto (which is the default setting).

*Open*
If you select Open no filter is used.

*Closed*
If you select Closed, a blank filter is used to prevent light from getting through.

250–395, 335–620, 550–1110, 695–1110
If you are looking at a wavelength range that is within one of these filter ranges, you can select your filter manually. Otherwise it is recommended that you set the filter to Auto.

**Emission filter**
Set the emission filter for the Kinetics measurement. To do this, select the down arrow to the right of the field and select an option from the list.

A range of filter settings are available:

*Auto*
If you select the Auto option, the filter wheel will be automatically moved to the appropriate position for the selected emission wavelength. It is recommended that you set the filter to Auto (which is the default setting).
**Open**
If you select **Open** no filter is used.

**Closed**
If you select **Closed**, a blank filter is used to prevent light from getting through.

**250–395, 295–1110, 360–1110, 430–1110, 550–1110**
If you are looking at a wavelength range that is within one of these filter ranges, you can select your filter manually. Otherwise it is recommended that you set the filter to **Auto**.

**3%T Attenuator**
Select **3%T Attenuator** to decrease the fluorescence intensity and thus the amount of light going to the emission PMT. This setting is useful for highly fluorescent samples when you are using wide slits.

**PMT Detector Voltage**
Set the EHT or sample detector PMT. To do this, select one of the following:

**Low**
Sets the PMT Detector voltage to a pre-defined value of 400 v. Typically you would select this setting if you are reading highly emitting samples.

**Medium**
Sets the PMT Detector voltage to a pre-defined value of 600 v. Typically you would select this setting if you are reading moderately emitting samples.

**High**
Sets the PMT Detector voltage to a pre-defined value of 800 v. Typically you would select this setting if you are reading low emitting samples.

**Manual**
Enables you to enter any PMT voltage in the adjacent Volts field.

**Volts**
Enter the PMT voltage in the adjacent field. Note that this option is only available if you have selected Manual as the PMT Detector voltage.

**Tip**
Increasing or decreasing the **PMT Detector Voltage** by 10% will double or halve the fluorescence intensity observed.

**Status Display**
Select this check box to display the **Status Display** window. This window allows you to view the status of various parameters during the data collection.

---

**About Smoothing—Kinetics**

**Savitzky-Golay**
Savitzky-Golay smoothing is used to obtain good noise reduction with sharp peaks. The equation constants are...
specific to Filter Size. The constants for each filter size are published in the Savitzky-Golay paper (A. Savitzky, M.J.E. Golay, Anal. Chem., 36 (1964) 1627). The equation also requires that Filter Sizes entered must be odd values, between 5 and 101.

**Example**

Data \((x_1, y_1), (x_2, y_2), (x_3, y_3) \ldots (x_n, y_n)\)

Filter size 5

**Result**

The first smoothed data point will be:
\[(x_3, (Y_1*a_1+Y_2*a_2+Y_3*a_3+Y_4*a_4+y_5*a_5)/b),\]

the next:
\[(x_4, (y_2*a_1+y_3*a_2+y_4*a_3+y_5*a_4+y_6*a_5)/b) \ldots \]
\[(x_{n-2}, \text{sum}(y_{n-4}*a_1+y_{n-3}*a_2+y_{n-2}*a_3+y_{n-1}*a_4+y_n*a_5)/b),\]

where \(a_1 \ldots a_5, b\) are constants defined by Savitzky and Golay.

Each smoothed point is calculated using the previous and next data points. For this reason, calculations cannot be performed on the very first and very last data points in a data set. Hence, smoothed traces will lack the first and last X values of the original trace.

**Boxcar average**

Boxcar averaging can be used when the change in intensity is minimal.
Example

Data \((x_1, y_1), (x_2, y_2), (x_3, y_3) \ldots (x_n, y_n)\)

Filter size 5

Result

The first smoothed data point will be:
\((x_3, \text{sum}(Y_1:y_5)/5),\)

the next:
\((x_8, \text{sum}(y_6:y_10)/5) \ldots (x_{n-2}, \text{sum}(y_{n-4}:y_n)/5).\)

If there are not enough points at the end of the data set to complete the filter size, then these points will be ignored, hence, the gap at the end may be larger than the gap in the beginning.

Moving average

![Diagram of moving average]

Example

Data \((x_1, y_1), (x_2, y_2), (x_3, y_3) \ldots (x_n, y_n)\)

Filter size 5

Result

The first smoothed data point will be:
\((x_3, \text{sum}(Y_1:y_5)/5),\)
the next:
(x4, sum(y2:y6)/5) … (xn-2, sum(yn-4:yn)/5).

**Accessories—Kinetics**

Use the **Accessories** page to set up the accessory parameters associated with the data collection.

From the **Setup** menu, select **Accessories** to access the **Accessories** page. You can also access the page from the **Setup** dialog by selecting the **Accessories** tab. To access the **Setup** dialog, select the **Setup** button from the Kinetics Application window.

The **Accessories** page will vary, depending upon what accessory button you have selected from the following:

- Multicell/Temperature
- Microplate reader

See also:
Cary page

**Multicell and Temperature accessories—Kinetics**

The Multicell and Temperature accessory page is displayed when you select the **Multicell/Temperature** tab on the **Accessories** page.

The page comprises the following:

**Multicell Holder**

Select this option to use the Multicell Holder and cells with the Cary Eclipse.

**Note**
By selecting this accessory you will prohibit the use of some combinations of other accessories that are also controlled by the Accessory Controller Board.

**Multi-zero**
If you select **Multi-zero**, a Multi-zero will be performed when the **Zero** button is selected in the Kinetics Application window. Because cuvettes can have different intensities, and samples vary, this field is used when you require all blank readings to consistently start at zero intensity.

A reading will be taken at the set **Averaging Time** at the set Ex. and Em. wavelengths and at every selected cell position. These readings will be offset to zero and subsequent readings will have the corresponding offset value subtracted from each cell.

**Reset**
Select **Reset** to drive the Multicell Holder to cell position 1, which is its fully retracted or zero position.

**Go to cell…**
Select **Go to cell** to open the **Multicell Changer** dialog. You can use this dialog to manually move the Multicell Holder accessory to a new cell. To do this, simply select the corresponding button of the cell you require in the Multi Cell Changer dialog. The Multicell Holder will move the selected cell to the measurement position in the sample compartment.

The **Multicell Changer** dialog is ‘modeless’ which means that it can remain open even once you have cleared the Setup dialog. This enables you to use the **Rapid Read** command to perform a reading at the cell specified in the Cell Changer.
If you move to a new cell after you have performed a Multi-zero scan, the Cary Eclipse will display the intensity reading corrected by the determined zero-offset amount for that cell.

**Note**
You can use this option to check the intensity of a cell or start a kinetics run at a particular cell.

**Temperature Control**

Select this option to use the Temperature Controller with the Cary Eclipse.

**Note**
The **Temperature Control** option does not control temperature accessories that are not listed in the **Device** field. For example, if you have selected the **Temperature Control** option and set the **Device** to **None** because you have a different temperature control accessory, the Cary Eclipse software will not control that accessory. However, it will monitor the temperature and prompt the software to start when the accessory reaches the temperature listed in the **Temperature** field.

**Device**
Select the device that will be used to control the water temperature.

The options available will depend upon whether or not you have selected the **Multicell holder** option.

If you **have** selected the Multicell holder, the following **Device** options will be available for selection:

- Temperature controller
- PCB 150 water bath
- None

If you **have not** selected the Multicell holder, the following **Device** options will be available for selection:

- Single cell peltier
- PCB 150 Water bath
- None

**None**
Select this option if you are using a Temperature Control device not listed in this field (for example, a third party water bath). Note that the Cary Eclipse software will not control the accessory. The software will simply monitor the temperature and prompt the software to start when the accessory reaches the temperature listed in the **Temperature** field.

**PCB 150 Water Bath**
Select this option if you are using the PCB 150 water bath.

**Single cell peltier**
Select this option if you are using the Single cell peltier.

**Temperature controller**
Select this option if you are using the Temperature controller.

**Temperature**
Set the temperature at which the reaction will be performed. The default value is 20.00°C.

When you select **OK** to close the **Setup** dialog, all buttons in the Kinetics Application window will be disabled until the temperature recorded by the **Monitor** device is within ± 0.5°C of the temperature range.

If **Zero** or **Start** is selected, and the temperature is not within ± 0.5°C of the temperature
Temperature Display

Select this option to view Temperature Display information in the Status Display window. The Temperature Display option is only available if you have selected the Temperature Control option.

Port
Select from COM1, COM2, COM3 or COM4. This option is not available if you have selected None as the Device.

Monitor
Select which device will be used to monitor the reaction temperature. Select from Block, Probe 1, Probe 2, Probe 3 or Probe 4. Note that the the options in this field may change depending on the Device selected.

Temperature Display

Select this option to view Temperature Display information in the Status Display window. The Temperature Display option is only available if you have selected the Temperature Control option.

Block/PCB 150
Select this option to view the temperature of the Block (if you have selected the Single Cell Peltier device) or the PCB 150 (if you have selected the PCB 150 Water Bath) in the Status Display window.

Probes
Select one or more of these options to view the temperature of the Probes in the Status Display window.

Status Display

Select this check box to display the Status Display window. Use this window to view the status of various parameters during the data collection.

See also:
Accessibility
Multi Cell Changer
Status Display

Multi Cell Changer—Kinetics

Use the Multi Cell Changer dialog to manually move the Multicell Holder accessory to a new cell. To access the Multi Cell Changer dialog, select the Go to cell button from the Multicell/Temperature Accessories page.

The dialog comprises the following:

Multicell changer
To manually move the Multicell Holder accessory to a new cell, simply select the corresponding button of the cell you require in the Multi Cell Changer dialog. The Multicell Holder will move the selected cell to the measurement position in the sample compartment.

The Multi Cell Changer dialog is ‘modeless’ which means that it can remain open even once you have cleared the Setup dialog. This enables you to use the Rapid Read command to perform a reading at the cell specified in the Cell Changer.

If you move to a new cell after you have performed a Multi-zero scan, the Cary Eclipse will display the intensity reading corrected by the determined zero-offset amount for that cell.

Note
The Wellplate accessory page is displayed when you select the Wellplate tab on the Accessories page. The page comprises the following:

**Wellplate reader**

Select this option to use the wellplate reader with the Cary Eclipse.

- **Q. Why is the wellplate reader option unavailable?**
  - **A.** The multicell holder, temperature control and/or polarization accessory has been selected. As you cannot place any of these accessories and the wellplate reader in the Cary Eclipse sample compartment at the same time, you must clear other accessory selections before selecting the wellplate reader.

**Plate format**
Select the type of wellplate to be used. The default options are 6 wells, 12 wells, 24 wells, 48 wells, 96 wells, 192 wells or 384 wells. The graphical representation of the wellplate will change according to the number of wells chosen.

**Auto-zero**
Select this option to perform an auto-zero when the Zero button is selected from the Kinetics Application window. A zero reading will be taken at the selected well position using the current method setup. To select the well position to be zeroed, use the Select Well option. These readings will be offset to zero and subsequent readings will have this single value subtracted from each well in the sequence. The reading is taken only once, stored, and subtracted from the ratioed intensity reading.

**Select well**
Choose Select well to open the Go To well dialog where you can select the well position to be zeroed. This option is only available if you have selected Auto-zero.

**Well**
Displays the well position of the well selected for auto-zeroing. This field is not editable.

**Display options**
Select this option to open the Display Options dialog where you can set up the display parameters associated with the wellplate reader.

**Sample names**
Select Sample Names to open the wellplate Sample List dialog where you can enter or modify the sample names. Highlight one or more wells on the wellplate reader diagram to view and change their sample information.

**Go to well**
Select this option to open the Go To Well dialog.

**Well position**
Displays the well position that the mouse cursor is pointing to. This field is not editable.

**Reset**
Select Reset to move the wellplate reader to the first well position defined by the read sequence.

**Park**
Select Park to move the wellplate reader to the Park position. The wellplate reader accessory...
is moved clear of the sample compartment to enable you to easily access all wells.

The wellplate reader diagram represents the number of wells present for the chosen wellplate configuration.

The labels for the rows will be 1..12 and for the columns A..H (for a 96 wellplate). Similar labels will occur for different wellplates according. The sequence for numbering the read order will be from A1 to A12, B12 - B1, C1 - C12… etc for a 96 wellplate. A similar sequence will be for other plates.

To select or clear a single well, simply click or right-click on it.

To select or clear a block of wells, hold down the left mouse and drag around the block required. All wells within the area will be selected or cleared.

**Status Display**

Select this check box to display the **Status Display** window. This window allows you to view the status of various parameters during the data collection.

**Display Options**—**Kinetics**

Use the **Display Options** page to set up the display parameters associated with the Wellplate Reader.

The **Display Options** page is accessible from the **Wellplate Accessories** page by selecting the **Display Options** button.

The page comprises the following:

**Display**

**Individual graphs**

Select this option to display the collected data of each sample in individual graph boxes, rather than overlaid on each other in one box.

At the start of a scan, a separate box is created for each sample. Similarly, the collected data for each sample is displayed in a separate graph box.

**Overlay all traces**

Select this option to superimpose the collected data of each sample in the run in one graph box.

One box is created at the start of the collect in the Graphics area and each sample’s data is overlaid in that box as the data is collected.

**User defined**

Select this option if you wish to specify the exact number of traces to be included on one graph.
**Number of traces per graph**
Enter the number of traces to be displayed per graph.

**Overlay rows**
Select Overlay rows to view overlaid graphs of the wells you have selected. Wells in the same row of the wellplate reader will be overlaid. If you had selected wells as shown in the adjacent diagram, a total of seven graphs would be displayed (as seven rows contain selected wells). The first graph would contain overlaid scans of well A1 and A6, the second graph would contain a scan of well B8, etc.

**Overlay columns**
Select Overlay columns to view overlaid graphs of the wells you have selected. Wells in the same column of the wellplate reader will be overlaid. If you had selected wells as shown in the adjacent diagram, a total of 11 graphs would be displayed (as 11 rows contain selected wells). The first graph would contain overlaid scans of well A1 and D1, the second graph would contain a scan of well H2, etc.

See also:
[Wellplate Accessories](#)

## Wellplate Sample List—Kinetics

Use the Wellplate Sample List dialog to enter or modify the sample names of each well in the wellplate reader.

The Wellplate Sample List is accessible from the Wellplate Accessories page by selecting the Sample Names button.

The page comprises the following:

**No. of samples**
Displays the total number of samples that have been selected from the Wellplate Reader diagram on the Wellplate Accessories page. This field is not editable.

**Sample table**
The sample table lists the Sample Names and Well numbers of the selected samples.

*Sample Name*
Samples are named numerically, depending upon where they are located on the Wellplate Reader diagram. The sequence for numbering the Sample Name order is to read from A1 to A12, B1 to B12, C1 to C12…etc for a 96 wellplate. Therefore, if the B2 cell is selected, its corresponding Sample Name will be Sample 14.

To modify the name of a sample, highlight the sample name and type in a new name. You can enter up to 20 characters for each name.

*Well*
Displays the position of the well corresponding to the Sample Name. This field is not editable.

**Import names**
Select Import Names to display the Open dialog which you can use to select a text file to
import into the Sample Names column.

**Increment**  
Select **Increment** to copy and increment the sample names in the Sample Name list. This will save you from entering each sample name individually when the names consist of a descriptor followed by a number.

To use the increment option, highlight the name that you want to increment and select **Increment**. The Cary Eclipse will increment the names of all the files proceeding the first highlighted name.

**Example:**  
In this example, Sample B1 is highlighted when the Increment button is selected.

<table>
<thead>
<tr>
<th>Sample Name</th>
<th>Becomes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Samp A</td>
<td>Samp A</td>
</tr>
<tr>
<td>Sample B</td>
<td>Sample B1</td>
</tr>
<tr>
<td>Sample C</td>
<td>Sample B2</td>
</tr>
<tr>
<td>Sample D</td>
<td>Sample B3</td>
</tr>
<tr>
<td>Sample E</td>
<td>Sample B4</td>
</tr>
</tbody>
</table>

See also:  
[Wellplate Accessories page](#)  
[About the Wellplate Reader](#)

---

**Go To Well—Kinetics**

You can use the **Go To Well** dialog to manually move the Wellplate Reader accessory to a new well. This enables you to check the intensity of a well or perform a read at a particular well.

The **Go To Well** page is accessible from the **Wellplate Accessories** page by selecting the **Select well** or **Go To Well** button.

The **Go To Well** dialog is ‘modeless’ which means that it can remain open even once you have cleared the **Setup** dialog. This enables you to use the **Rapid Read** command to perform a reading at the selected well.

If you move to a new well after you have performed a Multi-zero, the Cary Eclipse will display the intensity reading corrected by the determined zero-offset amount for that well.

The page comprises the following:

**Wellplate reader diagram**  
The Wellplate Reader diagram represents the number of wells present for the chosen Wellplate configuration.

The labels for the rows will be 1 .. 12 and for the columns A .. H (for a 96 Wellplate). Similar labels will occur for different Wellplates according. The sequence for numbering the read order will be from A1– A12, B12– B1, C1– C12, etc for a 96 Wellplate. A similar sequence will be for other plates.
To move the Wellplate Reader accessory to a new well, simply click on the corresponding well on this diagram. The Wellplate Reader will move the selected well to the measurement position in the sample compartment.

To select or clear a single well, simply click or right-click on it.

To select or clear a block of wells, hold down the cursor and drag around the block required. All wells within the area will be selected or cleared.

X, Y, Well  Displays the Well Position that the mouse cursor is pointing to. This field is not editable.
Reset  Select Reset to move the Wellplate Reader to the first well position, which is well A1.
Park  Select Park to move the Wellplate Reader to the Park position. When the accessory is in the park position you have full access to the Wellplate Reader platform. This is useful for inserting or removing wellplates from the accessory.

See also:
Wellplate Accessories page
About the Wellplate Reader

Analyze—Kinetics

Use the Analyze page to select from various mathematical equations and set the parameters used to generate the calculation of reaction rates.

From the Setup menu, select Analyze to access the Analyze page. You can also access the page from the Setup dialog by selecting the Analyze tab. To access the Setup dialog, select the Setup button from the Kinetics Application window.

The page comprises the following:

**Analysis**

**Auto Calculate**  Select the Auto Calculate option to automatically perform a rate calculation on collected data at the end of each run. If you do not select this option, you can still set up the calculation options, however, you will need to perform the calculation manually at the end of the run.

**Simple Calculate**  Select this option if you require only one rate calculation for the entire kinetics run. The Number of Stages option will be unavailable as only one stage will be calculated.

**Advanced Calculate**  Check this option to specify the number of abscissa ranges. You can specify this in the Number of Stages field. Up to five stages can be chosen and individual calculations and reports will be generated for each.

**Number of Stages**  Enter the number of sections in the data collection on which the Cary Eclipse will perform rate calculations. Up to five stages can be entered. The Time Ranges table will expand to fit the number entered.

**Note**  This option is only available if you select the Advanced Calculate option.

**Factor**  Use the Factor field to enter a value that will be used to calculate enzyme activity. This factor will be multiplied by the rate calculation result.

**Conc**  Enter the initial concentration of substrate before the reaction. This concentration is only required for second order reactions. Therefore, this option is only available when you have set one of the Stages to Second in the Order column of the Time Ranges table.
Q. I have selected Second Order but the Conc (mol/L) field is still grayed out—why?
A. You must select ‘Second’ in the Order column of the Time Ranges table and then select the cursor elsewhere to activate this field.

**Time Ranges Table**

Use the Time Ranges table to enter the calculation Start and Stop times (in minutes or seconds) for each stage. These will be used to set the abscissa range over which the rate calculation is performed. Up to five stages can be chosen.

**Note**
This option is only available if you select the Advanced Calculate option.

The table comprises the following:

**Stage**
This column lists the stage number/s.

**Start**
Use these fields to enter the starting point, in minutes, for a rate calculation analysis.

**Stop**
Use these fields to enter the end point, in minutes, for a rate calculation analysis.

**Order**
Click in this field to access a drop down list where you can select the order equation to be used for calculating reaction rates using intensity versus time data. The following options are available:

* Zero
  Select this to determine the reaction rate using a zero order equation.

* First
  Select this to determine the reaction rate using a first order equation. The rate is determined using the Marquardt non-linear regression analysis.

* Second
  Select this to determine the reaction rate using a second order equation. The rate is determined using the Marquardt non-linear regression analysis.

**Plot**
Select the way in which the plot will be displayed from the following options:

* Fit
  Select Fit to automatically overlay the calculated lines of best fit onto the plotted data.

Example of Fit overlay
Manual Guess

If you have selected a 'first order' or 'second order' Simple Calculate rate calculation you can use this group to manually enter the parameters: Intensity(0), Intensity(∞) and Rate (k). You will need to have a reasonable estimate of the values for these fit parameters, as they will be used as a first guess for the Marquardt non-linear regression analysis.

For some non-linear reactions you will get a result faster if you put in an initial estimate in the manual guess field.

Note

If you do not choose Manual Guess then the Cary Eclipse system will automatically calculate the values for Intensity(0), Intensity(∞) and Rate when you select OK to exit the Setup dialog.

Manual Guess
Select this option to manually select the parameters with which to calculate a first order or second order reaction rate.

The value of these parameters will be used as a first guess for the Marquardt non-linear regression analysis.

Q. Why is the Use Manual Guess check box unavailable?
A. You can only perform a manual guess when Simple Calculate is selected in the Analyze group, and First or Second is selected in the Order column of the Time Ranges Table. Once you have made these selections and clicked the cursor elsewhere, this check box will become activated. When you select Manual Guess, the other options in the group will also be activated.

Intensity (0) Enter the estimated intensity at zero time for first and second order fits.
Intensity (∞) Enter the estimated intensity at infinite time for first or second order kinetics reaction. When Zero order calculations are performed, the value of this field is automatically set to zero by the Cary Eclipse system.
k Enter the rate estimate in this field.

Status Display

Select this check box to display the Status Display window. This window allows you to view the status of various parameters during the data collection.

See also:

Setup
Status Display
Example of multi-stage kinetics calculations
Rate calculations
How to use the kinetics ruler

Rate Calculations—Kinetics
Reaction rates may be determined in the Kinetics application using zero order, first order or second order equations using intensity versus time data over defined time ranges.

Alternatively you can manually determine the range over which to calculate the reaction rate by using the kinetics ruler.

**Zero Order**

When the Rate calculation of the Kinetics application is set to 'zero order', a linear equation is fitted to the data using a least-squares technique. The slope of the line of best fit is reported as the kinetics rate in the equation:

\[ \text{ABS} = kt + \text{ABS}_0 \]

where:

- \( \text{ABS} \) = intensity at time \( t \)
- \( \text{ABS}_0 \) = intensity at time \( t = 0 \)
- \( k \) = rate constant
- \( t \) = time

The units typically used are intensity per minute.

**First order and Second order**

First order and Second order fits are performed using a Marquardt non-linear fit technique. The values of \( \text{ABS}_0 \) and Rate are estimated using a technique outlined in 'Alternative Method of Analyzing First-Order Kinetic Data' by Lowell M. Schwartz and Robert I. Gelb (Analytical Chemistry 1978, Volume 50, Number 11).

The estimates are then used as the first guess for the Marquardt non-linear regression analysis. The Marquardt analysis calculates new values that are subsequently used as the next guess in a recursive manner, until the change in the values is very small, or until a certain number of cycles (default of 100) have been completed.

The first-order equation is:

\[ \text{ABS} = \text{ABS}_\infty + (\text{ABS}_0 - \text{ABS}_\infty) e^{-kt} \]

where:

- \( \text{ABS} \) = intensity at time \( t \)
- \( \text{ABS}_0 \) = intensity at time \( t = 0 \)
- \( \text{ABS}_\infty \) = intensity at time \( t = \text{infinity} \)
- \( k \) = rate constant
- \( t \) = time

The second-order equation is:

\[ \text{ABS} = \frac{(\text{ABS}_0 - \text{ABS}_\infty) + \text{ABS}_\infty}{X_0 .kt + 1} \]

where:
ABS = intensity at time t
ABS₀ = intensity at time t = 0
ABSₙ = intensity at time t = infinity
k = rate constant
t = time
X₀ = initial concentration of one component

See also:
How to use the kinetics ruler

Reports—Kinetics

Use the Reports page to set up the report parameters associated with data collection.

From the Setup menu, select Reports to access the Reports page. You can also access the page from the Setup dialog by selecting the Reports tab. To access the Setup dialog, select the Setup button from the Kinetics Application window.

Hot Tip
You can view your generated report before you print it out using the Print Preview command in the File menu.

If you decide to make changes to the report layout, select (Edit Report) from the Report toolbar. This enables you to directly edit the report.

The page comprises the following:

Operator

<table>
<thead>
<tr>
<th>Name</th>
<th>Enter the instrument operator name in this text entry field. Use the arrow to the right of this field to view and select one of the last ten entries in this field.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Note: In GLP mode the system will automatically enter the operator’s name and this field will not be editable.</td>
</tr>
</tbody>
</table>

| Comment | Enter operator details and comments in this text entry field. |

Options

This group enables you to select the information to include in your report.

Auto Print

Select Auto Print to print a report automatically during the data collection. If Auto Print is selected, then the system will send the report information to the specified printer(s) as well as the Report area. However if Auto Print is not selected, the report will only be sent to the Report area and can be viewed by selecting Report in the View menu.

To automatically obtain a copy of the graphics, you will need to select both the Auto Print and Graph options (see below). In this case, all graphical information displayed on screen will be printed with the report. Therefore it is advisable to arrange the graphical information as you require on screen, prior to printing.
For example, if you require a single graph print out, then the **Single/Multi Graph** option in the **Graph** menu should be changed to the correct setting.

### Parameters
Select this option to include the method setup parameters within your report.

### User data form
Select this option to include in the report any information entered in the **User Data Form**.

### Graph
Select **Graph** to include any graphs or graphic information associated with your report. If you require a printout of the displayed graphics only, you must ensure that *only* this option is selected. Select the **Clear Report** button to clear any information in the Report area before you print otherwise this will be included in your printout.

#### % Page Height
Enter the size at which you want the graph to be printed, as a percentage of total page height. For example, if you want graphical information to take up half of the page, enter 50% in this field.

### Company Logo
Check this option to include your company logo in the report. A company logo will only be included if you have added one on the Company page of the System Information Application.

#### Hot Tip
If you have not included a Company logo, select the **Header/Footer** button to open the System Information Application and use the Add Logo option to add a logo.

### Header/Footer
Select the **Header/Footer** button to open the System Information application where you can enter the Header and Footer information to be included in your report. You can also enter any additional comments, and add a logo to your report. Once you have made all your changes, select OK to exit the System Information application and return to the Reports page of the Kinetics application.

### Focused trace
Select this option to apply the **Report** selections to the focused trace only.

### All traces
Select this option to apply the **Report** selections to all visible traces.

### X-Y Pairs Table
Select this if you want to print a table showing the X-Y pairs of the collected data in your report.

#### Data Interval
- **Actual**
  Select **Actual** to report the X-Y pairs table for the trace at the actual **Data Interval** at which the data was collected.

- **Interpolated**
  Select **Interpolated** to report the X-Y pairs table for the trace at an interpolated data interval. Enter this data interval in the adjacent field. If no data has been collected at the interval you enter, the Cary Eclipse software will interpolate the data to produce an X-Y result.

### Status Display
Select this check box to display the **Status Display** window, where you can view the status of your kinetics run during the data collection.

#### Hot Tip
You can copy a picture, spreadsheet file or document from another application (for example, Microsoft
Word or Microsoft Excel) into the report area of the Lifetimes application. To edit these objects, simply double click on them to open the associated application in which they were created.

See also:
- User Data Form
- View menu
- Status Display window

Report Toolbar—Kinetics

Use the Report toolbar buttons to gain quick access to many of the Edit menu options. You can also use the toolbar to cut, copy and paste information and to change the look of your report.

To display or hide the toolbar, point to the Toolbars option in the View menu and select Reports. To display or hide the toolbar's text labels, point to the Toolbar option in the View menu and select Text Labels.

Hot Tip
To see the name of a button, point to it with the mouse.

The following commands can be accessed from the Reports Toolbar:

<table>
<thead>
<tr>
<th>Button</th>
<th>Menu command</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>![Edit Report]</td>
<td>Click this button to edit the report. This command is also accessible by pressing CTRL+R.</td>
<td></td>
</tr>
<tr>
<td>![Undo]</td>
<td>If available, this undoes the last action. This command is also accessible by pressing CTRL+Z.</td>
<td></td>
</tr>
<tr>
<td>![Cut]</td>
<td>Click this button to remove the highlighted text and place it on the Windows Clipboard. This command is also accessible by pressing CTRL+X.</td>
<td></td>
</tr>
<tr>
<td>![Copy]</td>
<td>Click this button to copy the highlighted text onto the Windows Clipboard. This command is also accessible by pressing CTRL+C.</td>
<td></td>
</tr>
<tr>
<td>![Paste]</td>
<td>Click this button to insert the contents of the Windows Clipboard (text only) at the cursor. Any highlighted text will be replaced. This command is also accessible by pressing CTRL+V.</td>
<td></td>
</tr>
</tbody>
</table>
| ![Delete] | Click this button to remove selected text in the Report area. This command is also accessible by pressing DEL.  
  **Note** This option is only available once you select Edit Report and there is text highlighted in the Report area. |
| ![Font] | Click this button to change the font of new or selected text in the report. For existing text, simply highlight the text in the report, select this button and make your changes in the Font dialog. When you select OK, the appearance of the highlighted text in the report will alter to reflect your font changes. This command is also accessible by pressing CTRL+F. |
| ![Preview] | Click this button to display the Print Preview dialog where you can view the current report as it will be printed. |

Clear Report—Kinetics
Select the **Clear Report** command to remove the contents of the Report Area in the Kinetics Application window. If you have saved the report, selecting this option does not delete the contents of the report, the information is simply removed from the Report Area.

[Clear report: To access the **Clear Report** option, select the **Clear Report** command from the **Commands** menu, or select the **Clear Report** button from the Kinetics Application window.]

See also:
- [Kinetics Application](#)
- [Commands menu](#)

### How to use advanced reporting options—Kinetics

This outlines ways to fully customize your reports. Options available include:

- Including header and footer information
- Including a logo
- Editing the text in the report
- Viewing the report on screen before printing
- Cutting and pasting to a word processor

#### Including header and footer information

You can have text appearing at the top and bottom of every page in your report. To specify what the text should be, do the following:

**Step 1**

On the Windows Taskbar, click the Windows **Start** button, point to **Programs, Cary Eclipse** and then select **System Information**. The System Information application appears with the Company page displayed.

**Step 2**

Enter the header and footer information in the **Header and Footer** fields.

**Step 3**

Click **OK** to implement your changes.

#### Including a logo

You can include a logo on the first page of your report. To specify what the logo should be, do the following:

**Step 1**

On the Windows Taskbar, click the Windows **Start** button, point to **Programs, Cary Eclipse** and then select **System Information**. The System Information application appears with the Company page displayed.

**Step 2**

Enter a logo using the **Add Logo** button. This logo will always be included on the front page of reports.
Step 3

Click **OK** to implement your changes.

Q. Why isn't the logo I selected included in my report?
A. If you are running the Cary Eclipse software under Windows NT you need to be logged on as an Administrator in order to specify the logo in the System Information application.

**Editing the text in the report**

You can edit any of the text in the Report area. Just select **Edit Report** from the **Edit** menu. You can then add, delete and change text in the Report area.

Q. The Edit Report option is not available, why not?
A. You do not have reporting editing privileges set in the GLP Administration application. The Administrator of the system will have to change your privileges before you can edit reports.

**Viewing the report on the screen before printing**

You can view a report before printing it. To do this, from the **File** menu select **Print Preview**. You can then ensure that the report format is correct before you print it.

**Hot Tip**
Use the arrow keys at the bottom of the Print Preview screen to change pages.

**Cutting and pasting to a word processor**

To have complete control over how your report looks you may wish to copy it to a word processor program, such as Microsoft Word. To do this:

Step 1
Open a report.

Step 2
From the **Edit** menu, choose **Edit Report**.

Step 3
Click in the Report area and highlight the text you want to export to your word processor.

Step 4
Right-click on the Report area and select **Copy** from the menu.

Step 5
Start up your word processor and use the Paste function (usually under the **Edit** menu) to place the text into a document.
Auto Store—Kinetics

Use the Auto Store page to specify if and when data will be stored, and in what format. You can also use this page to set up the system to automatically send an email at the end of a read.

From the Setup menu, select Auto store to access the Auto store page. You can also access the page from the Setup dialog by selecting the Auto store tab. To access the Setup dialog, select the Setup button from the Kinetics Application window.

The page comprises the following:

Storage

- **Off**
  - Select this if you do not wish to be automatically prompted to save collected data.
  - **Note** If you wish to store your unsaved data manually, choose Save Data As… from the File Menu to save your data after the run is complete. You can save your data as a Batch or Data file.

- **On; Prompt At Start**
  - Select this to display the Windows Save As dialog at the start of the collect where you can enter the Batch filename for your data.

- **On; Prompt At End**
  - Select this to display the Windows Save As dialog at the end of a collect where you can enter the Batch filename for your data.

Auto convert

Use the Auto convert options to convert your data into an ASCII file format that can be opened in spreadsheet programs such as Microsoft Excel.

- **None**
  - Select None if you do not need to generate an ASCII file.

- **ASCII (csv)**
  - Select this option to save the data as a comma delimited ASCII Spreadsheet file (*.CSV) file. Only the currently collected data is converted.

  ASCII files are stored in the same folder as the data file from which they were generated and designated the same filename as the method used to collect the data plus the extension ‘CSV’.

- **ASCII (csv) with log**
  - Select this option to save the data as a comma delimited ASCII Spreadsheet file (*.CSV) file. Only the currently collected data is converted. The key method parameters used to collect the data are also saved in this file.

  ASCII files are stored in the same folder as the data file from which they were generated and designated the same filename as the method used to collect the data plus the extension ‘CSV’.

  **Note**
  - If you choose to auto-convert the data to an ASCII file but you have set the Storage option to Off, then the ASCII file will be saved with the following format: DATE TIME, CSV
The date and time will appear in the same form that is set in the Windows Regional Settings Properties dialog that is accessed from the Windows Control Panel.

For example, 22_4_97 1;32;08.CSV

**Email**

Select the **Email** check box and enter the Email details to send an email to a person once the read is completed. This useful facility can be used to alert you to the completion of the read, particularly if you are working away from the laboratory where the Cary Eclipse instrument is. You can also attach the collected data to the email.

![Note](image)

You must set up the email **Host** and **Port** options on the **Email options** page of the System Information application in order for the email facility to work correctly.

- **To** Enter the recipient's email address. You can enter more than one recipient in this field.
- **From** Enter your email address (or the email address associated with the computer on which the Cary Eclipse software is installed).
- **Subject** Enter the subject of your email (or leave this field blank if you do not wish to include a subject).
- **Cc** Enter the email address of any recipient you want to copy the email to.
- **Message** Enter your message.
- **Attached…** Use the Attached button include a file attachment in your email. To do this, select Attached and browse your computer to select a file, using the Windows® Open dialog that appears.
- **Attach current data** Select this option to attach the data that has been collected during the run to the email.

[How to email your results](#)

**Status display**

Select this check box to display the **Status Display** window, where you can view the status of your kinetics run during the data collection.

**See also:**

- [Kinetics Application window](#)
- [Save As dialog](#)
- [Status Display window](#)

**How to email your results—Kinetics**

You may wish to send an email to a person once the read is completed. This useful facility can be used to alert you to the completion of the read, particularly if you are working away from the laboratory where the Cary Eclipse instrument is.

To do this:

**Step 1**

Ensure that you have set up the **Host** and **Port** options on the **Email options** page of the System Information
application in order for the email facility to work correctly.

Step 2

From the Setup menu, select Auto Store. The Auto Store page is displayed.

Step 3

Select the Email check box to enable the email options.

Step 4

In the To field, enter the recipient's email address (for example, rchandle@varianinc.com). To enter multiple recipients, simply type a comma ',' between each email address.

Step 5

In the From field, enter the email address of the computer on which the Cary Eclipse software is installed. This may be your email address, or a communal email address, if the Cary Eclipse software is installed on a computer that has multiple users.

Step 6

Enter information in the Subject and Message fields as required.

Step 7

Select Attach current data. The data that has been collected during the run will be attached to the email.

**Hot Tip**

You can attach other documents to your email. To do this, select the Attached button and browse your computer to select a file, using the Windows® Open dialog that is displayed.

Step 8

Select OK to clear the Auto Store dialog. Your email will be sent once the run has been completed.

See also:

Auto Store

Performing scans—Kinetics

- Starting a run
- Sample Name dialog
- Cell Loading Guide
- Sync Start dialog
- Start/Stop buttons
- Pre-scan dialog
- Stop Time dialog
- Go To dialog
Information about starting a collect—Kinetics

The sequence that occurs when you select **Start** to commence the kinetics run depends upon your storage and accessory selections.

**Storage selections**

<table>
<thead>
<tr>
<th>If you have selected … as the Storage option on the Auto Store page</th>
<th>The following occurs</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>On, prompt at start</strong></td>
<td>The Windows <strong>Save As</strong> dialog will appear as soon as you select <strong>Start</strong>. You can save the current method, collected data, report and graph template as a Batch file.</td>
</tr>
<tr>
<td><strong>On, prompt at end</strong></td>
<td>The Windows <strong>Save As</strong> dialog will appear at the end of the data collection. You can save the current method, collected data, report and graph template as a Batch file.</td>
</tr>
<tr>
<td><strong>Off</strong></td>
<td>The Windows <strong>Save As</strong> dialog will not appear.</td>
</tr>
</tbody>
</table>

**Accessory selections**

<table>
<thead>
<tr>
<th>If you have selected … on the Accessories page</th>
<th>The following dialog is displayed</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>No accessories</strong></td>
<td>The <strong>Sample Name</strong> dialog will appear when you select <strong>Start</strong>, prompting you to load the sample and enter your sample name. Select <strong>OK</strong> to display the <strong>Sync Start</strong> dialog and commence the kinetics run.</td>
</tr>
<tr>
<td><strong>Multicell Holder</strong></td>
<td>The <strong>Cell Loading Guide</strong> dialog will appear. Enter the sample name of each activated cell then select <strong>OK</strong> to display the <strong>Sync Start</strong> dialog. When you select <strong>OK</strong> to clear the <strong>Cell Loading Guide</strong> dialog, the cell changer will move to cell position 4, enabling you to easily access all of the cell positions and load your samples. <strong>If you need to start data collection immediately:</strong> Load the cells into the cell changer and select <strong>Reset Slide</strong> from the <strong>Sync Start</strong> dialog. The cell changer will move to cell position 1. You should then add the reactants to the cells as quickly as possible, close the lid and select <strong>OK</strong> on the Sync Start dialog. The collect will start immediately without performing another reset command. <strong>If you do not need to start the data collection immediately:</strong> Add the reactants to all the cuvettes and select <strong>OK</strong> on the <strong>Sync Start</strong> dialog. The system will automatically reset the cell changer to cell position 1 and start measurements on the selected cuvettes.</td>
</tr>
<tr>
<td><strong>Wellplate Reader</strong></td>
<td>The <strong>Sync Start</strong> dialog will appear after you press <strong>Start</strong>. The sample</td>
</tr>
</tbody>
</table>
Sample Name—Kinetics

If you are using a single cell holder, the Sample Name dialog appears once you select the **Start** button to commence an analysis.

The page comprises the following:

- **Sample Name**
  Enter the name of the sample to be used in the kinetics run. Click on the arrow to the right of the field to view and select the most recently entered sample names.
  Once you have entered the **Sample Name**, select **OK** to display the **Sync Start** dialog and commence the kinetics run.

See also:
- Sample Name
- Cell Loading Guide
- Sync Start
- Start/Stop buttons
- Setup
- Accessories
- Auto Store

Cell Loading Guide—Kinetics

If you are using a Multicell Holder, the **Cell Loading Guide** dialog appears once you select the **Start** button to commence an analysis.

The **Cell Loading Guide** dialog prompts you to insert blank samples in the Multicell Holder to collect the Kinetics scans. You can also use the dialog to name the samples in each selected cell in the Multicell Holder.

The dialog comprises the following:

- **Loading Guide/Sample Name entry**
  Enter the names of the sample to be used in the kinetics run in each available field. This sample name will be used as the Name of Trace in the **Trace Preferences** dialog.

**Note**

The **Cell Loading Guide** dialog appears only if you have selected the Multicell Holder option on the **Accessories** page of the **Setup** dialog. Only cells selected in the Multicell Holder diagram on the **Accessories** page will be available in the Loading Guide.

In the adjacent example, cells 1—3 will be available in the Loading Guide dialog, as they have been selected in the Multicell Holder diagram.

Once you have entered the **Sample Name**, select **OK** to display the **Sync Start** dialog and commence the Kinetics run.
Sync Start—Kinetics

Use the Sync Start dialog to synchronize the start of a reaction and the start of the kinetics data collection. Before displaying the Sync Start dialog, the Cary Eclipse will set up all the instrument and graphing parameters so that data collection can start as soon as the countdown finishes or you select OK.

The sequence for a Sync Start is as follows:

- Set up your data collection parameters
- Click the Start button (and load the samples if necessary)
- Once the Sync Start dialog is displayed, add your active reagent
- Commence the data collection by selecting the OK button

The dialog comprises the following:

**Count Down** This field displays a two minute countdown in seconds. If you want to start your data collection before the two minute countdown ends select the OK button. Alternatively, you can wait for the countdown to end and the data collection will start automatically.

**Note** If you are using a Multicell Holder and you select the OK button, the system will automatically Reset this accessory and immediately start the kinetics run.

**Reset Slide** Click this button to drive the Multicell Holder to its zero point (Cell position 1).

Start/Stop Instrument buttons—Kinetics

The following buttons may be displayed at the top of the Kinetics Application window. The **Start**, **Pause**, **Continue** and **Connect** buttons will change depending upon the status of the run. For example, if a run has not been started, the **Start** button will be visible to indicate that the system is ready to commence a run.

Select **Start** to commence a kinetics run using the current Setup parameters. If any unsaved data exists in the Graphics area you will be prompted to save the data. If you choose not to save, any unsaved data, pictures and labels will be deleted when you start the kinetics run.

When a kinetics run is started, the **Start** button will change to **Pause**.

The **Pause** button is displayed in place of the **Start** button once you have selected **Start** to commence a run. Click **Pause** to temporarily suspend the data collection, so that you can modify your solutions during the kinetics run. While the system is in the paused state, it will continue to keep track of the elapsed time, however data collection will stop until the **Continue** button is selected.

The benefit of using **Pause** instead of **Stop** is that you are able to add a secondary substrate to your kinetics solution(s) and then continue to monitor a secondary reaction, without having to Stop and Start the collect again. It also means that you will not get a spike in your data when you open the sample compartment to add the reagent.
When a kinetics run is paused, the Pause button will change to Continue.

The Continue button is displayed in place of the Pause button when you have paused the run. Click Continue to proceed with the kinetics run after it has been paused.

Once you select Continue, the button changes back to Pause.

If another application is already online with the Cary Eclipse, the Start button will change to Connect, and you will need to select Connect to bring this application online before the Start button will re-appear. (You will not be able to Connect if the other application is collecting data.)

Select this to end a kinetics run.

If you have selected Storage On in the Auto Store page of the Setup dialog, all data collected is automatically stored in the file specified in the Batch name field of the Windows Save As dialog before or after the run commenced.

See also:
Graphics area
Commands menu

Zero—Kinetics

Use the Zero button to zero the current intensity value. The intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before commencing a data collection if you want your data to start at 0 intensity. The Cary Eclipse will prompt you to load a blank before performing the zero.

Before you select Zero, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.

When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the Kinetics Application window.

Select the Clear Zero command from the Commands menu to remove zero correction and thus remove the subtraction effect.

To access the Zero option, select the Zero command from the Commands menu, or select the Zero button from the Kinetics Application window.

See also:
Kinetics Application
Commands menu
Cell Loading Guide

Pre-scan—Kinetics

Select Pre-scan to determine the optimum instrument parameters to be used for your sample. At the end of this collection sequence the software will report the optimum Ex and Em wavelengths and PMT voltage. Once you are satisfied with the parameters, the software will automatically update your method setup to reflect these new parameters.

To access the Pre-scan option, select the Pre-scan command from the Graph menu, or select
The Pre-scan dialog comprises the following:

**Type of scatter**

Select the type of scatter you wish to search for from the following:

- **Rayleigh scatter**: Rayleigh scatter refers to the scatter of the excitation light. It is displayed at the peak of the excitation wavelength.
- **Raman scatter**: Raman scatter refers to scatter due to the solvent. Its peak is at a fixed energy from the excitation wavelength, and is solvent dependent. Thus, the type of solvent used must be selected from the Solvent list.
- **2nd order**: Second order scatter refers to the scatter of higher order excitation light. Its peak is a multiple of the excitation wavelength.

See also:
- Graph menu
- Kinetics Application

**Stop Time—Kinetics**

Use the **Stop Time** dialog to extend the length of a kinetics run while the run is in progress. Extending a run may be necessary if you decide whilst a run is in progress that the data collection period is too short. The **Stop Time** command is not available until a run is in progress (that is, the **Start** button is selected).

To access the **Stop Time** dialog, select the **Stop Time** button from the Kinetics Application window or the **Stop Time** command from the **Commands** menu.

**To extend the length of a kinetics run in progress:**

1. Select the **Stop time** option from the Kinetics Application window or the **Commands** menu. The **Stop Time** dialog is displayed.

2. In the **Stop Time** field, the minimum allowable Stop Time will be displayed. This is the Stop Time that has been specified on the **Cary** page of the **Setup** dialog. In the **Stop Time** field, enter the new Stop Time and select **OK**. The X axis (Time) of the graph in the Graphics Area will be updated to reflect the new setting.

See also:
- Kinetics Application
- Cary page

Use the **Go to Wavelength** dialog to temporarily change the instrument to a new excitation or emission wavelength. You would mainly use this dialog if you want to quickly go to a wavelength and check the intensity reading of the sample. The lamp will flash continuously in this mode. This option is only available if the instrument is not currently collecting data.

To access the **Go to Wavelength** dialog, select the **Go To Wavelength** command from the **Commands** menu. You can also select **F4** to open the dialog.
Note

This is not the wavelength used in the application method. If your method uses a wavelength that is different from the 'Go to wavelength' setting you select, then when you select Start the system will change to the wavelength selected. (In most applications, this is set in the Cary page of the Setup dialog.)

The Go to Wavelength page comprises the following:

**Excitation**

*Wavelength*

Enter the new wavelength in this numeric entry field. When you select Go to the instrument will change to the new value.

*Zero Order*

Select Zero Order to drive the Cary Eclipse fluorescence spectrophotometer to 0 nm (white light). You can then use this light for aligning accessories.

**Emission**

*Wavelength*

Enter the new wavelength in this numeric entry field. When you select Go to the instrument will change to the new value.

*Zero Order*

Select Zero Order to drive the Cary Eclipse fluorescence spectrophotometer to 0 nm (white light). You can then use this light for aligning accessories.

**Go To**

Click Go to once you have entered a new wavelength, to change the instrument to the entered value.

See also:

- Commands menu
- Function Keys

---

**Go to Temperature—Kinetics**

Use the Go to Temperature dialog to change the current temperature to a new value.

To access the Go to Temperature dialog, select the Set temp button from the Kinetics Application window or the Set temp command from the Commands menu.

The Set Temp button is only accessible if you are using a thermostatted accessory. (On the Accessories page, the Temperature control option must be selected.)

You can access the Go to Temperature dialog prior to starting a run or once a run is in progress.

**To change the current temperature of a run in progress:**

1. Access the Set Temp button by selecting the Pause button to pause the run. You can then select the Set Temp button.

2. In the Set temp field, enter the temperature you want the instrument to change to. Select OK to return to the Kinetics Application window.

3. Select Continue to proceed with the run.

See also:

- Kinetics Application
Rapid Result—Kinetics

Use the Rapid Result dialog to take a reading of a blank and sample/s, without having to change the current application method. You can use the Multi Cell Changer dialog to specify the cell you want to read. This means that between runs, while new samples are being prepared and loaded, other samples can quickly be measured. The result is printed in the Report area.

To access the Rapid Result dialog, select the Rapid Result command from the Commands menu or press F6.

Excitation  

Wavelength
Enter the wavelength at which you want to take a reading.

Zero order
Select this option to set the excitation monochromator to zero order (white light).

Emission  

Wavelength
Enter the wavelength at which you want to take a reading.

Zero order
Select this option to set the excitation monochromator to zero order (white light).

Zero
Click Zero to change the wavelength to the entered value and measure a blank.

Result
Click Result to change the wavelength to the entered value and measure a sample. The X,Y results are corrected for the blank and the results are displayed in the Report window.

See also:
Multi Cell Changer
Commands menu

Commands menu—Kinetics

Use the Commands menu to access major operational commands in the Kinetics application. The menu is accessible from the menu line at the top of the Kinetics Application window.

Alternatively you can use function key shortcuts for some of the commands. These appear next to the item in the Commands menu. Note that not all commands appear as both button and menu items.

Hot Tip
If you access these commands using the Commands menu, rather than from the application buttons, you can remove these buttons from your application window. To do this, clear the Buttons command in the View menu. This will enlarge the Graphics area, making viewing and selection of scans easier.

Tell me how to remove or display the command buttons

The Commands menu comprises the following commands:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Start</td>
<td>Select this to start a data collection using the currently set up parameters. If the Connect command is displayed in place of the Start command, this indicates that another Cary Eclipse application is in use and has control of the Cary Eclipse instrument. To make the Kinetics application active so that you can perform a read, you will need to select Connect. However, you will not be able to select Connect if another application is</td>
</tr>
<tr>
<td><strong>Stop</strong></td>
<td>Select this to stop the data collection. If you have selected Storage On in the <strong>Auto Store</strong> page of the <strong>Setup</strong> dialog, all data collected is automatically stored in the file specified in the <strong>Batch name</strong> field of the Windows <strong>Save As</strong> dialog before or after the run commenced.</td>
</tr>
<tr>
<td><strong>Clear Zero</strong></td>
<td>Select this option to clear the <strong>Zero</strong> command. When you select the <strong>Zero</strong> command (from the <strong>Commands</strong> menu or by pressing F5), the intensity at that time will be reset to zero and all subsequent results will have the intensity value that was reset to zero subtracted from them. Selecting <strong>Clear zero</strong> cancels this zeroing (and thus subtraction) effect.</td>
</tr>
<tr>
<td><strong>Pre-scan</strong></td>
<td>Select <strong>Pre-scan</strong> to quickly determine the optimum instrument parameters to be used for your sample. At the end of this collection sequence the software will report the optimum Ex and Em wavelengths and PMT voltage. Once you are satisfied with the parameters, the software will automatically update your method setup to reflect these new parameters.</td>
</tr>
<tr>
<td><strong>Zero order…</strong></td>
<td>Select the <strong>Zero order</strong> command to set one of the monochromators to zero order. If you use this command to zero either the excitation or emission monochromators, this will override the <strong>Ex. Wavelength</strong> or <strong>Em. Wavelength</strong> set on the <strong>Cary</strong> page. The Cary Eclipse software will use the zero setting when collecting data. The <strong>Zero order</strong> command is generally used during alignment procedures.</td>
</tr>
<tr>
<td><strong>Excitation</strong></td>
<td>Select this command to set the excitation monochromator to zero order (white light).</td>
</tr>
<tr>
<td><strong>Emission</strong></td>
<td>Select this command to set the emission monochromator to zero order (white light).</td>
</tr>
<tr>
<td><strong>Reset</strong></td>
<td>Select this command to re-initialize the instrument. This will perform the same initializing procedure that is performed when the instrument is powered on.</td>
</tr>
<tr>
<td><strong>Rapid Result</strong></td>
<td>Select this to display the <strong>Rapid Result</strong> dialog. Use this dialog to take a reading of a blank and multiple sample readings without changing the application method. This means that between runs, while new samples are being prepared and loaded, other samples can quickly be measured. This command can also be accessed by pressing F6.</td>
</tr>
<tr>
<td><strong>Zero</strong></td>
<td>Select this to zero the current intensity value. The intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before selecting <strong>Start</strong> if you want your data to start at 0 intensity. This command is also accessible by pressing F5.</td>
</tr>
<tr>
<td><strong>Note</strong></td>
<td>Before you select <strong>Zero</strong>, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.</td>
</tr>
<tr>
<td><strong>Go to…</strong></td>
<td>Select <strong>Go To</strong>… to display the <strong>Go To Wavelength</strong> dialog where you can change the instrument to a new wavelength. Use this command if you want to quickly go to a wavelength to check the intensity reading. This command is only available if the instrument is not currently collecting data. It can also be accessed by pressing F4.</td>
</tr>
<tr>
<td><strong>Note</strong></td>
<td>This is not the wavelength used in the application method. If your method uses a wavelength that is different from the 'Go To wavelength', then when you select <strong>Start</strong> the system will change to the wavelength selected. In most applications, this is set in the <strong>Cary</strong> page of the <strong>Setup</strong> dialog.</td>
</tr>
<tr>
<td><strong>Set temp</strong></td>
<td>Select <strong>Set temp</strong> to open the <strong>Set Temperature</strong> dialog where you can change the temperature</td>
</tr>
</tbody>
</table>
How to perform a single-wavelength kinetics run—Kinetics

This demonstrates how to perform a fluorescence single-wavelength kinetics run at ambient temperature.

Follow the steps through, adjusting them to meet the requirements of your particular data collection.

- Start the Kinetics application
- Set up data collection parameters
- Set up instrument parameters
- Set up rate parameters
- Set up the options
- Set up accessories
- Set up rate calculation, rate order and other rate parameters
- Set up reporting and printing requirements
- Set up storage of collected data
- Set up email options
- Set up visual system monitoring
- Finish Setup
- Zero the instrument
- Start the kinetics run
- Save your data

Start the Kinetics application

Step 1
Click the Windows Start button, point to Programs, Cary Eclipse and then select Kinetics. The Kinetics Application is displayed.

**Note**
If you are running a GLP system, you may be prompted to enter a password before accessing the Kinetics application.

**Set up data collection parameters**

Step 2 [Setup dialog]

Select Setup to display the Setup dialog and specify the method parameters for a new method.

To do this:

**Set up instrument parameters**

Step 3 [Setup dialog | Cary page]

a. In the **Data mode** field set the mode to **Fluorescence**.

b. In the **Ex. Wavelength** field, set the wavelength of the excitation monochromator to a wavelength at which the chromophore absorbs light. For example, if you were measuring quinine, this absorbs at 350 nm.

c. In the **Em. Wavelength** field, set the wavelength of the emission monochromator. For quinine this would be approximately 450 nm.

d. Set the slit width of the excitation and emission monochromators to 5. Note that as a general rule, at a fixed emission slit width, increasing the excitation slit width will result in an increase in the fluorescence excitation intensity. You can therefore choose to increase the slit width if the resultant intensity is too low, however it is recommended that you initially try to increase intensity by using the PMT Detector Voltage setting (on the Options page).

**Set up rate parameters such as the number of different rates in the reaction, the speed, and the duration of the data collection**

Step 4 [Setup dialog | Cary page]

a. In the **Collect Timing** group, select the **Simple Collect** option. This sets up a single rate for your reaction.

b. Enter a **Cycle time** of 0. The **Ave time** will therefore be used to determine the distance between points.

**Hot Tip**
If you have a long, slow reaction, use the Cycle time to limit the number of data points that are collected.

c. Specify the duration of the kinetics run by setting the **Stop** time for the rate stage. You will need to have knowledge of your experiment to determine this value.

d. In the **Ave Time** field enter the required value. If the sample you are measuring is highly fluorescent,
then 0.1 sec is a good starting point. If the sample fluoresces quickly, you may need to decrease the Ave time to 0.0125 sec in order to ensure that you produce a data point each time the lamp is flashed. Note however that decreasing the Ave time will increase the signal-to-noise ratio.

**Set up the options**

Step 5 [Setup dialog | Options page]

a. Select the Options tab. The Options page is displayed.

b. In the Display Options group, select the way in which you want the data displayed as it is collected. For example, select the Overlay Data button to superimpose the collected data of each sample in the kinetics run in one graph box.

c. Enter the minimum and maximum Y values to be displayed on the graph axis during the data collection.

d. Set the Excitation filter and the Emission filter for the concentration measurement to Auto. It is not necessary to set the Emission filter to the default setting of Open, as you are only looking at a single wavelength.

e. Set the PMT Detector voltage to Medium. To determine whether this setting is most suitable for obtaining the highest possible intensity, use the Rapid Result option, which is accessible from the Commands menu.

**Set up accessories**

Step 6 [Setup dialog | Accessories page]

Select the Accessories tab and ensure that no accessories are selected.

**Set up rate calculation, rate order and other rate parameters**

Step 7 [Setup dialog | Analyze tab]

a. Select the Analyze tab. The Analyze page is displayed.

b. Select the Auto Calculate check box to automatically perform a rate calculation on collected data at the end of each run.

c. Select Simple Calculate to set up one rate calculation for the entire kinetics run.

d. Enter a Start time of 0 min and Stop time that is the same as the Stop time entered on the Cary page.

e. The Order you set will depend upon the reaction you are performing. Choose from Zero, First or Second.

**Set up reporting and printing requirements**

Step 8 [Setup dialog | Reports page]

a. Select the Reports tab to display the Reports page where you can specify your reporting requirements for this method.
b. Enter your name in the **Name** field.

c. Enter a comment relating to your experiment in the **Comment** field (*optional*).

d. Set up your report style by selecting the appropriate check boxes in the **Options** group.

   For example, select the **Auto Print** check box to obtain a printout of your report automatically. Select the **Parameters** check box to include your experimental parameters in the report. Select the **Graph** check box to include a calibration graph in the generated report.

   **Note**

   If **Auto Print** is selected, then the system will send the report information to the specified printer as the Report area. However if **Auto Print** is not selected, the report will only be sent to the Report area and can be viewed by selecting the **Report** command from the **View** menu.

e. Select the **X-Y Pairs Table** check box to view a list of abscissa values and their corresponding intensity values.

**Set up storage of collected data**

Step 9 [Setup dialog | Auto Store page]

a. Select the **Auto Store** tab to display the **Auto Store** page.

b. Set the **Storage** option to **Off**. The method (with calibration), collected data and report will not be automatically saved. However, you can manually save it all at the end of the collection.

c. Select the **Auto Convert** option you require. If you select **ASCII (csv)** or **ASCII (csv) with log**, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format. The data file will be saved in the current folder.

**Set up email options**

Step 10 [Setup dialog | Auto Store page]

Refer to [How to email your results](#) for instructions on setting up the email options.

**Set up visual system monitoring**

Step 11 [Setup dialog]

Select the **Status Display** check box on any of the Setup pages to display various information fields on your current reaction.

**Finish Setup**

Step 12

Once you are satisfied with your method setup select **OK** to confirm any changes you have made and clear the **Setup** dialog.
Zero the instrument

Step 13

a. Place the blank solution in the sample compartment of the Cary Eclipse instrument.

b. Select the Zero button to zero the system. Alternatively, select Zero from the Commands menu to perform a zero. When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the Scan Application window.

Start the kinetics run

Step 14

a. Select the Start button to commence a data collection. Alternatively, select Start from the Commands menu. Do not add your active reagent at this time. The Sample Name dialog is displayed.

b. Place the sample in the sample compartment and select OK. The Sync Start dialog will be displayed.

c. Add your active reagent just before the Count Down reaches 0:00 or commence the data collection by selecting the OK button.

Save your data

Step 15

a. Once the run is finished, select the Save As command from the File menu.

b. In the File name field, enter a file name for this kinetics run.

c. Select Save. The data will be stored as a Batch file.

See also:
How to perform post run calculations on previously stored data
How to export collected data

How to perform a multi-wavelength, temperature controlled kinetics run—Kinetics

This demonstrates how to perform a multi-wavelength, temperature controlled kinetics run using the PCB 150 Water bath.

Follow the steps through, adjusting them to meet the requirements of your particular data collection.

- Start the Kinetics application
- Set up data collection parameters
- Set up instrument parameters
- Set up rate parameters
- Set up collection display options
- Ensure Multicell Holder accessory is not selected
- Set up accessories for reaction temperature control and temperature display
- Set up rate calculation, rate order an other rate parameters
- Set up reporting and printing requirements
- Set up storage of collected data
- Set up the email options
- Set up visual system monitoring
- Finish Setup
- Start the kinetics run

Start the Kinetics application

Step 1

Click the Windows Start button, point to Programs, Cary Eclipse and then select Kinetics. The Kinetics Application is displayed.

**Note**

If you are running a GLP system, you may be prompted to enter a password before accessing the Kinetics application.

Set up data collection parameters

Step 2 [Setup dialog]

Select Setup to display the Setup dialog and specify the method parameters for a new method.

To do this:

Set up instrument parameters

Step 3 [Setup dialog | Cary page]

a. In the **Data mode** field set the mode to **Fluorescence**.

b. In the **Wavelength setup** group, select **Multiwavelength**. The Multiwavelength table will be displayed. Enter the following parameters in the Multiwavelength table. For the purpose of this example, FURA-2 will be used.

<table>
<thead>
<tr>
<th>Name</th>
<th>Ex. wavelength (nm)</th>
<th>Em. wavelength (nm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wavelength 1</td>
<td>340</td>
<td>510</td>
</tr>
<tr>
<td>Wavelength 2</td>
<td>380</td>
<td>510</td>
</tr>
</tbody>
</table>

Note that as a general rule, at a fixed emission slit width, increasing the excitation slit width will result in an increase in the fluorescence excitation intensity. You can therefore choose to increase the slit width if the resultant intensity is too low, however it is recommended that you initially try to increase intensity by using the PMT Detector Voltage setting (on the **Options** page).

Set up rate parameters

Step 4 [Setup dialog | Cary page]

a. In the **Collect Timing** group, select the **Advanced Collect** option. This enables you to set up different data collection procedures for the multiple rates in your reaction.
b. Set the value in the Number of Stages field to 3. The table below will expand to three rows.

c. Set the Ave time to 0.0125 sec.

d. Enter the following parameters in the Stages table:

<table>
<thead>
<tr>
<th>Stage</th>
<th>Cycle (min)</th>
<th>Stop (min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.0000</td>
<td>20.0000</td>
</tr>
<tr>
<td>2</td>
<td>1.0000</td>
<td>60.0000</td>
</tr>
<tr>
<td>3</td>
<td>5.0000</td>
<td>100.0000</td>
</tr>
</tbody>
</table>

Therefore, from 1–20 min, a cycle will be performed every 0.0125 sec. (When the Cycle time is set to 0, the Ave time is used to determine the distance between points.) From 21–60 min, a cycle will be performed every 1.0000 min and from 61–100 min, a cycle will be performed every 5.0000 min.

### Set up collection display options

**Step 5 [Setup dialog | Options page]**

a. Select the Options tab. The Options page is displayed.

b. Enter the minimum and maximum Y values to be displayed on the graph axis during the data collection.

c. Set the Excitation filter for the concentration measurement. It is recommended that you set the filter to Auto (the default setting). This will automatically move the filter wheel to the appropriate position for the selected excitation wavelength.

d. Set the Emission filter for the concentration measurement. In order to minimize steps in the spectra, it is recommended that you set the filter to Open (the default setting).

e. Set the PMT Detector Voltage. The setting you choose will depend upon your sample. For example, a Low setting is recommended for highly emitting samples, while a High setting is recommended for low emitting samples.

### Ensure Multicell Holder accessory is not selected

**Step 6 [Setup dialog | Accessories page]**

Select the Accessories tab to display the Accessories page. On the Multicell/Temperature tab, ensure that the Multicell Holder accessory is not selected.

### Set up accessories for reaction temperature control and temperature display

**Step 7 [Setup dialog | Accessories page]**

a. Select the Temperature control check box to enable the Temperature group.

b. Click on the down arrow adjacent to the Device field and select the PCB 150 Water bath accessory.

---

**Note**

The Advanced Collect facility enables you to collect data more frequently during the crucial stages of your reaction and to collect data less frequently where you know there will not be much activity.
c. Set the **Temperature** to 37°C.

d. Set the appropriate Communications port from the **Port** drop down list.

e. Set the **Monitor** to PCB 150.

f. In the **Temperature Display** group, select **Probe 1** to view the temperature of one temperature probe in the **Status Display** window.

**Set up rate calculation, rate order and other rate parameters**

**Step 8** [Setup dialog | Analyze page]

a. Select the **Analyze** tab. The **Analyze** page is displayed.

b. Select the **Auto Calculate** check box so that a rate calculation will be automatically performed on collected data at the end of each run.

c. Select **Advanced Calculate** to set up multiple rate calculations for the kinetics run. If you select **Advanced Calculate**, the Cary Eclipse system will also automatically calculate the values: A0, AInf and Rate when the Start button is selected.

d. In the **Number of stages** field, enter a value of 3. The Stages table will be expanded to three rows.

e. Enter the following parameters in the Stages table:

<table>
<thead>
<tr>
<th>Stage</th>
<th>Start (min)</th>
<th>Stop (min)</th>
<th>Order</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.00</td>
<td>20.00</td>
<td>Depends on reaction</td>
</tr>
<tr>
<td>2</td>
<td>21.00</td>
<td>60.00</td>
<td>Depends on reaction</td>
</tr>
<tr>
<td>3</td>
<td>61.00</td>
<td>100.00</td>
<td>Depends on reaction</td>
</tr>
</tbody>
</table>

**Set up reporting and printing requirements**

**Step 9** [Setup dialog | Reports page]

a. Select the **Reports** tab to display the **Reports** page where you can specify your reporting requirements for this method.

b. Enter your name in the **Name** field.

c. Enter a comment relating to your experiment in the **Comment** field *(optional)*.

d. Set up your report style by selecting the appropriate check boxes in the **Options** group.

For example, select the **Auto Print** check box to obtain a printout of your report automatically. Select the **Parameters** check box to include your experimental parameters in the report. Select the **Graph** check box to include a calibration graph in the generated report.

**Note**

If **Auto Print** is selected, then the system will send the report information to the specified printer as well as the Report area. However if **Auto Print** is not selected, the report will only be sent to the Report area and can be viewed by selecting the **Report** command from the **View** menu.
e. Select the **X-Y Pairs Table** check box to view a list of abscissa values and their corresponding intensity values.

**Set up storage of collected data**

Step 10 [Setup dialog | Auto Store page]

a. Select the **Auto Store** tab to display the **Auto Store** page.

b. Set the **Storage** option to **On; prompt at start**. You will be prompted to save your method (with calibration), collected data and report when you commence the run.

c. Select the **Auto Convert** option you require. If you select **ASCII (csv)** or **ASCII (csv) with log**, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format. The data file will be saved in the current folder.

**Set up email options**

Step 11 [Setup dialog | Auto Store page]

Refer to [How to email your results](#) for instructions on setting up the email options.

**Set up visual system monitoring**

Step 12 [Setup dialog]

Select the **Status Display** check box on any of the Setup pages to display various information fields on your current reaction.

**Finish Setup**

Step 13

Once you are satisfied with your method setup select **OK** to confirm any changes you have made and clear the **Setup** dialog.

**Zero the instrument**

Step 14

a. Place the blank solution in the sample compartment of the Cary Eclipse instrument.

b. Select the **Zero** button to zero the system. Alternatively, select **Zero** from the **Commands** menu to perform a zero. When the result is zeroed, the word ‘Zeroed’ will appear in the Y display box in the top left corner of the Scan Application window.

**Start the kinetics run**

Step 15

a. Select the **Start** button to commence a data collection. Alternatively, select **Start** from the **Commands**
menu. **Do not add your active reagent at this time.** The system will display the **Save As** dialog.

b. Enter the file name for this kinetics run in the **File name** field and select **Save**. The system will display the **Sample Name** dialog.

c. Place the sample solution in the correct cell position and select **OK**. The **Sync Start** dialog will be displayed.

d. Add your active reagent just before the **Count Down** reaches 0:00 or commence the data collection by selecting the **OK** button.

---

See also:

- How to perform post run calculations on previously stored data
- How to export collected data

### Viewing run progress and results—Kinetics

- Reports page
- Reports toolbar
- View menu
- Edit menu
- Status display
- Print Preview dialog
- Print

### View menu—Kinetics

The **View** menu is accessible from the menu line at the top of the Kinetics Application window.

**Hot Tip**

Highlight a method or data file in Explorer and simply drag and drop it onto the application to open it. Any data will be displayed in the Graphics area.

The following commands can be accessed from the **View** menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Toolbar</td>
<td>Select this option to toggle the <strong>Graphics</strong> and <strong>Reports</strong> toolbar on and off. You can also select to display or hide the toolbar <strong>Text Labels</strong>.</td>
</tr>
<tr>
<td>Buttons</td>
<td>Select this option to toggle the left-hand button display (button area) on and off.</td>
</tr>
<tr>
<td>Graphics</td>
<td>Select this option to toggle the Graphics area on and off.</td>
</tr>
<tr>
<td>Report</td>
<td>Select this option to toggle the Report area on and off.</td>
</tr>
<tr>
<td>Report Vertical</td>
<td>If the Graphics option is selected, select <strong>Report Vertical</strong> to display the Graphics area, Report area and/or ADL Editor side by side vertically.</td>
</tr>
<tr>
<td>ADL</td>
<td>Select this option to toggle the ADL Editor on and off.</td>
</tr>
<tr>
<td>ADL Vertical</td>
<td>If the <strong>ADL</strong> and <strong>Report</strong> options are selected, select <strong>ADL Vertical</strong> to display the ADL Editor vertically on the right hand side of the Report area.</td>
</tr>
<tr>
<td>ADL Quick Reference</td>
<td>This displays the <strong>Quick Reference</strong> window where you can easily add a command in the correct syntax to your ADL program. A detailed description of all the commands in the</td>
</tr>
</tbody>
</table>
Quick Reference is available by selecting the Help button.

| ADL Hooks | Select this to activate a link (or hook) to an ADL program at a specific point in the application, and then run that ADL program. For example, ADL hooks have been supplied to run specific programs:

- on start-up of the Cary Eclipse software;
- when the system goes online or offline;
- when the Start button is selected;
- when a run finishes;
- when a zero is performed etc.

For the hook to work, **ADL Hooks** must be selected, and the appropriate ADL file for the application must be present in the ADL folder in the directory.

⚠️ **Note**
Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.

| Hints | Select this option to toggle the Hints bubble text on and off. The bubble text appears when you place the arrow cursor over a field, button or text entry field for a short period. It offers a quick explanation of the item’s function.

| Status Display | Select this option to display the **Status Display** window, where you can view the status of various parameters during the data collection.

See also:
- Kinetics Application
- Graphics area
- Status Display
- ADL Hooks
- How to insert ADL commands using the Quick Reference

**Edit menu—Kinetics**

Use the **Edit** menu to copy, cut, paste and delete information. You can also use the menu to access the Report editing commands. The menu is accessible from the menu line at the top of the Kinetics Application window.

The following commands can be accessed from the **Edit** menu:

⚠️ **Note**
Some options are only available once you select **Edit Report**.

<table>
<thead>
<tr>
<th>Command</th>
<th>Button</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Undo</td>
<td><img src="undo_icon.png" alt="Undo" /></td>
<td>If available, this undoes the last action. This command is also accessible by pressing CTRL+Z.</td>
</tr>
<tr>
<td>Cut</td>
<td><img src="cut_icon.png" alt="Cut" /></td>
<td>Select this command to remove the highlighted text and place it on the Windows Clipboard. This command is also accessible by pressing CTRL+X.</td>
</tr>
<tr>
<td>Copy</td>
<td><img src="copy_icon.png" alt="Copy" /></td>
<td>Select this command to copy the highlighted text onto the Windows Clipboard. This command is also accessible by pressing CTRL+C.</td>
</tr>
</tbody>
</table>
Use the Status Display window to view the status of your kinetics run during the data collection. Depending upon settings you have for the Kinetics run, the following parameters may be displayed:

- Current Excitation Wavelength (nm)
- Current Emission Wavelength (nm)
- Excitation Slit Width (nm)
- Emission Slit Width
- PMT Voltage (V)

To access the Status Display window, select Status Display from the View menu, or from any page of the Setup dialog. You can choose to view the Status Display at any time during a kinetics run.

See also:
- View menu
- Setup
- Status Line

Print Preview—Kinetics
Use the Print Preview dialog to view the current report before you print it. From the File menu, select Print Preview to access the dialog.

The contents of the Graphics area appear first in the preview area followed by the contents of the Report area. You can reduce or enlarge the print preview on screen. To do this, click the left mouse button in the preview area to toggle through the five available views. Click the right mouse button in the preview area to return to the full page view.

The page comprises the following commands:

- **First arrow** [↩] Use this arrow to view the first page of the report.
- **Previous arrow** [↩] Use this arrow to view the previous page of the report.
- **Next arrow** [↩] Use this arrow to view the next page of the report.
- **Last arrow** [↩] Use this arrow to view the last page of the report.
- **Print** [↩] Use this to display the Windows Print dialog where you can choose the printer and pages to print as well as the number of copies you wish to print.

**Q. Why are the arrow buttons grayed out?**

**A.** The buttons will be grayed out if the report has only one page of data.

See also:
- **File menu**

**Print**—Kinetics

Use the Print dialog to select the printer and pages to print of the currently displayed report as well as the number of copies you wish to print.

To access the Print option, select the Print command from the File menu, or select the Print button from the Kinetics Application window.

All graphical information displayed on screen will be printed with the report, therefore it is advisable to edit the report on the screen prior to printing. You can also preview the report prior to printing by using the Print Preview command.

If you require a printout of the graphics in the Graphics area only, select the Clear Report button to erase the contents of the Report area before you print.

**Note**

The options on the Report page are used to generate the contents of the Report area during a run or on a recalculation. If Auto Print has been selected the Cary Eclipse will automatically print a report at the end of a data collection.

See also:
- **Print Preview**
- **Report page**
- **Edit Report**
- **Kinetics Application**
Performing Calculations—Kinetics—Kinetics

- Recalculate
- How to perform post run calculations on previously stored data
- How to generate a report from previously stored data
- How to use advanced reporting options
- Analyze
- Rate Calculations
- Maths Calculator
- Maths Operations
- Maths How To
- User Data Form

Recalculate—Kinetics

Use the pages accessible from the Recalculate option to perform re-calculations on data that has been analyzed. These re-calculations can be made using new Setup parameters. For example you may wish to change the order of a reaction. You can also use the Recalculate button to access Report options.

To access the Recalculate option, select the Recalculate button from the Kinetics Application window.

When you select this option, the Setup dialog will appear, containing the following tabbed pages:

- Analyze
- Reports

See also: Kinetics Application

How to perform post run calculations on previously stored data—Kinetics

- Retrieve the data
- Set up the recalculation
- Print the report

Retrieve the data

Step 1

From the File menu, select Open to display the Windows Open dialog.

Step 2

A list of previously stored Batch files will appear. Select the Batch file you require then select Open.

The system will automatically retrieve the collected data into the Graphics area. The method that was used to collect the data will also be retrieved.

Note
You can also retrieve data from Data (*.FDKN), Method (*.FMKN) and Spreadsheet ASCII (*.CSV) file types.

**Set up the recalculation**

**Step 3**

Select the **Recalculate** button to display the **Setup** dialog. The **Analyze** and **Reports** pages will be displayed to enable you to set up the recalculation parameters.

**Step 4**

Select the **Analyze** tab to access the **Analyze** page where you can set the analysis parameters you require.

For example, select the **Auto Calculate** option to automatically perform a rate calculation on collected data at the end of each run.

**Step 5**

a. Select the **Reports** tab to display the **Reports** page where you can specify your reporting requirements for this method.

b. Enter your name in the **Name** field.

c. Enter a comment relating to your experiment in the **Comment** field *(optional)*.

d. Set up your report style by selecting the appropriate check boxes in the **Options** group.

   - For example, select the **Auto Print** check box to obtain a printout of your report automatically.
   - Select the **Parameters** check box to include your experimental parameters in the report.
   - Select the **Graph** check box to include a calibration graph in the generated report.

**Note**

If **Auto Print** is selected, then the system will send the report information to the specified printer as well as the Report area. However if **Auto Print** is not selected, the report will only be sent to the Report area and can be viewed by selecting the **Report** command from the **View** menu.

**Step 6**

Select **OK** to clear the **Setup** dialog and start the recalculations. The results will be displayed in the Report area.

**Print the report**

**Step 7**

a. From the **File** menu, select **Print**. The Windows **Print** dialog is displayed.

b. Set up your printing parameters and select **OK** to print your report.

**See also:**

*How to generate a report from previously collected data*
How to generate a report from previously stored data—Kinetics

Step 1
Select **Open...** from the **File** menu to display the Windows **Open** dialog.

Step 2
A list of previously stored files will appear. If this is not a list of batch files, select the down arrow at the right of the **Files of type** list box and select 'Batch' to list all the batch files.

Step 3
Select the Batch file you require then select **Open**.

The system will automatically retrieve the collected data into the Graphics area. The method that was used to collect the data will also be retrieved.

**Note**
You can also retrieve data from Methods (*.FMKN), Data (*.FDKN), Report (*.FRKN), Spreadsheet ASCII (*.CSV), GRAMS (*.SPC) and Rich Text Format (*.RTF) file types.

Step 4
Select the **Recalculate** button to display the **Setup** dialog.

Step 5
Select the **Reports** tab and on the **Reports** page specify the information you would like to include in the report. Select **Auto Print** if you require the new report to print to the Reports area and to a printer.

Step 6
Click **OK** to display the results in the Report area (and send to the printer if **Auto Print** is selected).

**Note**
If you require a printout of only the current graphics in the Graphics area of a previously stored data file:

a. Select the **Clear Report** option in the Commands menu to clear any text that may be displayed in the Report area.

b. Press the **Print** button to obtain a printout of the graphics.

Alternatively tick only the **Graph** check box in the **Reports** page to obtain a printout of graphics only.

See also:
[How to use advanced reporting options](#)

**Maths window**

Use the **Maths** window to perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator.
To access the Maths window, select the Calculator command from the Graph menu, or select the Calculator button on the Graphics toolbar. The Maths window is only available in the Kinetics, Lifetimes, Scan and Validate applications.

The Maths window comprises the following:

**Calculation display field**
View the current calculation, including the trace/s being used, the trace operation being carried out and any functions that have been applied to the trace.

**Hot Tip**
Click on the down arrow adjacent to this field to view the last ten unique calculations that have taken place. Use this list to keep track of previous manipulations and repeat calculations quickly by simply selecting and then applying the desired calculation.

**Selected Trace/Focused Trace**
Choosing either of these options selects the highlighted (red) trace in the current Graph box for manipulation. When choosing between these options, use this simple rule:

- Use the **Selected Trace** option when the selected calculation is only to be applied to one trace.
- Use the **Focused Trace** option when the same calculation is to be applied to a number of traces.

The following example illustrates the difference between these options.

To perform the same manipulation on several traces:

a. Highlight a trace by clicking on it with the mouse. The trace will become red.
b. Select **Selected Trace**, **Apply** and then = to plot the result of the calculation
c. Highlight the next trace and repeat step b.

OR

a. Highlight the first trace.
b. Select **Focused Trace**, **Apply** and then = to plot the result of the calculation;
c. Highlight the next trace and select the = button to perform the same calculation on the newly selected trace.

**Trace**
Use the Trace group options to select the operation to be performed on the traces, the filter size and the interval (where applicable).

Click **Apply** to insert the selected operation into the equation
Click **=** (equals) to display the result of the calculations.

**Operation**
Click on an operation to find out more about it

- Anisotropy
- Polarization
- Sum Function
- Mean
- Boxcar Averaging
- Moving Average
- Savitzky-Golay
- Spline
- Deriv 1
- Deriv 2
Display Options
Select whether the results of the operation performed on the trace will be viewed in a New Graph box or in the Selected Graph box. If you choose the Selected Graph option, you will be able to directly compare the results with existing traces.

Functions
This group contains number and function buttons that can be used as a standard calculator in addition to being applied to traces.

To use the functions:

1. select a trace or value
2. select either a second trace or number
3. select the equals (=) button to display the result

All the basic mathematical functions are available (addition, subtraction, multiplication and division), as well as logarithm (Log) and square root (Sqrt) functions. When used as a calculator, the results of equations appear in the Report area.

Additional buttons
- Click C to clear the entire operation in the calculation display field.
- Click CE to clear the last entry entered in the calculation display field.
- Click = (equals) to start the operation shown in the calculation display field.

See also:
- Maths Home Page
- Maths How To

Maths Operations

The following operations can be selected on the Maths window. To access the Maths window, select the Calculator command from the Graph menu, or select the Calculator button on the Graphics toolbar.

Anisotropy, Polarization and Sum Function
The term Anisotropy refers to an unequal angular distribution about a point in space. Anisotropy also refers specifically to a simple expression for measuring the degree of polarization of the fluorescence. It is almost identical to the expression for polarization apart from having a factor of two in the denominator. Anisotropy is usually represented by the lowercase letter "r", while polarization by an uppercase "P".

The two expressions are listed below:

Anisotropy
\[ r = \frac{(I_{vv} - G_{Ivh})}{(I_{vv} + 2G_{Ivh})} \]

Polarisation
\[ P = \frac{(I_{vv} - G_{Ivh})}{(I_{vv} + G_{Ivh})} \]

where

I_{vv} equals the intensity of fluorescence with excitation polarizer = vertical, emission polarizer = vertical
**Ivh** equals the intensity of fluorescence with excitation polarizer = vertical, emission polarizer = horizontal

G is a factor that accounts for the polarization bias of the instrument. In particular it accounts for the wavelength-dependent polarization bias of the emission side of the instrument (dominated by the polarization bias of the emission grating).

The G factor is given by:

\[ G = \frac{I_{hv}}{I_{hh}} \]

where

**Ihv** equals the intensity of fluorescence with excitation polarizer = horizontal, emission polarizer = vertical

**Ihh** equals the intensity of fluorescence with excitation polarizer = horizontal, emission polarizer = horizontal

The expression for the anisotropy is a quotient. The denominator of the expression, as given by \( I_{vv} + 2GI_{vh} \) is the ‘sum’ function, and actually represents the total emission unbiased by any sample polarization effects.

**Mean**

The Mean operation allows the average of two or more traces to be taken and the resulting trace to be plotted. The mean of the traces can also be used in conjunction with other functions.

The equation used in mean calculations is:

\[ \frac{(scan \ 1 + scan \ 2...+ \ scan \ x)}{x} \]

The calculation is performed over the common X range of the traces and the trace with the most dense data is used to determine the data points. The other traces are interpolated at the relevant data points.

**Boxcar Averaging**

This algorithm will take the number of points as defined by the Filter size, and average these, displaying the average at the middle X-point of the filter.

For example, for the following:

Data: \((x1, y1), (x2, y2), (x3, y3) \ldots (xn,yn)\)

Filer size: \(5\)

The first smoothed data point will be \((x3, \text{sum}(Y1:y5)/5), \) the next \((x8, \text{sum}(y6:y10)/5) \ldots (xn-2, \text{sum}(yn-4:yn)/5).\)

If there are not enough points at the end of the data set to complete the filter size, then these points will be ignored, hence, the gap at the end may be larger than the gap in the beginning. There will be a gap before the first point and after the last point of the smoothed continuum, with respect to the original unsmoothed data. This algorithm is only valid for evenly x-spaced data points.

Within a time scan, (e.g., Advanced collect in kinetics) the data points may be unevenly spaced between different stages. Each stage will be treated as a new collect, hence, there may be large gaps between stages. If there are not enough points at the end of a stage to
complete the filter size, then these points are ignored, hence, there may be large gaps at the end of a stage. The filter size will be limited to odd numbers, with the minimum of 3 to a maximum of 101.

**Moving Average**

This algorithm will take the number of points as defined by the Filter size, and average these, displaying the average at the middle X-point of the filter.

For example, for the following:

Data: \((x_1, y_1), (x_2, y_2), (x_3, y_3) \ldots (x_n, y_n)\)

Filter size: 5

The first smoothed data point will be \((x_3, \text{sum}(Y_1:y_5)/5)\), the next \((x_4, \text{sum}(y_2:y_6)/5) \ldots (x_{n-2}, \text{sum}(y_{n-4}:y_n)/5)\). There will be a gap before the first point and after the last point of the smoothed continuum, with respect to the original unsmoothed data. This algorithm is only valid for evenly x-spaced data points. Within a time scan, (e.g., Advanced collect in kinetics), the data points may be unevenly spaced between different stages. This will be treated by interpolation of data from the previous stage so as to give evenly spaced data points into the next stage. If there is not enough data for interpolation, then the next stage will be treated as a new collect, hence, there may be gaps and further delays in displaying the point. The filter size will be limited to odd numbers, with the minimum of 3 to a maximum of 101.

**Savitzky-Golay**

The Savitzky-Golay technique operation enables you to reduce the noise in the currently selected Maths scan by using smoothing.

When you select this operation you will be prompted to enter a Filter Size from 5 to 101. The larger the Filter Size, the more data points that will be smoothed.

Q. I cannot enter a particular value; it keeps changing to another number — why?
A. The Savitzky-Golay technique requires an odd number to be entered, not an even number.

You will also need to enter an Interval. The Interval refers to how many of the averaged points will be plotted on the smoothed trace. The smaller the interval, the more detail there will be in the trace.

Consider selecting an Interval of 1. This would mean that when the smoothed continuum is plotted, every smoothed point will be displayed.

Q. Why can't I see the Filter Size and Interval fields?
A. These will be displayed when you select Smooth in the Operation field.

**Spline**

The Deriv operations allow you to obtain derivatives of a selected scan. The derivative can be of the orders 1 to 4. The resulting trace is then plotted. The Savitzky-Golay technique is used for derivative calculations.

On selection of the Deriv operation you will be prompted to enter a Filter Size from 5 to 101. The Filter Size refers to the number of points around a particular point that will be averaged to produce a new derived Y value. The lower the Filter Size, the more data points that will be used in the derivative trace.

Q. I cannot enter a particular value; it keeps changing to another number — why?
A. The Savitzky-Golay technique requires an odd number to be entered, not an even number.

You will also need to enter an Interval. The Interval refers to how many of the derived points will be plotted on the resulting trace. The smaller the Interval, the more detail there will be in the trace.

Q. Why can't I see the Filter Size and Interval fields?
A. These will be displayed when you select one of the ‘Derivs’ in the Operation field.

See also:
About smoothing

Maths How To…

- How to perform a spectral subtraction
- How to perform a mean calculation
- How to smooth a trace
- How to derive a trace

See also:
Maths Home Page

User Data Form—Kinetics

Use the User Data Form to enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data.

To access the User Data Form, select the User Data Form command from the Graph menu, or select the User Data Form button on the Graphics toolbar. The User Data Form command is only available if you have selected a graph box.

The dialog comprises the following:

Data Name  This column lists the names of each visible trace in the highlighted graph box. Use the extra columns for recording additional information about the experimental conditions you used to collect the data, eg. pH.

Other Columns  Use this to record the name of the parameter in which you are interested as a column heading, (for example, pH, ionic concentration, substrate concentration etc.).

Note  You must enter a column heading, or else the data in that column will not be saved.

See also:
Graph menu
Graphics toolbar

Displaying data graphically—Kinetics

- Graphics area
- Graphics How To
- Graph menu
- Graphics toolbar
The Graphics area of the Kinetics application appears in the top half of the Kinetics Application window and is used to display traces of collected data. In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area surrounding the graph is referred to as the graph box. A graph will appear in the Graphics area when you select Start to commence a Kinetics run.

To display or hide the Graphics area, select the Graphics command on the View menu. Double-clicking on the Graphics area enables you to toggle between half or full size display sizes.

You can access the functions associated with the Graphics area by doing the following:

- selecting one of the Graphics toolbar buttons;
- selecting a command from the Graph menu; or
- clicking the right mouse button in the Graphics area and select a command from the menu

Note
Not all functions are available on every menu.

You can customize the graphics area to suit your requirements in the following ways:

**Note**
All of the commands and dialogs listed are accessible from the **Graph menu**.

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<th>Use the…</th>
<th>Tell me how…</th>
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How to track the focused trace

How to select the focused trace

How to display a trace that is not appearing in a graph box

How to remove a trace from a graph

How to remove a trace from the trace List

How to resize graph boxes

How to scale your graph manually

How to scale your graph automatically

How to alter the look of your graphs

How to zoom a graph

How to add a label to a graph

How to edit a label

How to delete a label

How to move a label

How to add a picture to a graph

How to move a picture

How to delete a picture

How to replace a picture

Move pictures in the graph box

Resizing pictures in the graph box

Replace pictures in the graph box

Delete pictures from the graph box

Save the Graphics area you have set up as a graphics template

Cursor

Edit Picture dialog

Delete annotation command

Save As dialog (from the File menu)

Note

The red trace in any graph box is the one on which all information appears and the one used for the cursor tracking. It is called the focused trace.

See also:

Toolbar

Graphics How To

Graphics How to—Kinetics

Traces

How to track the focused trace

How to select the focused trace

How to display a trace that is not appearing in a graph box

How to remove a trace from a graph

How to remove a trace from the trace List

Sizing and scale

How to resize graph boxes

How to scale your graph manually

How to scale your graph automatically

How to alter the look of your graphs

How to zoom a graph

Labels

How to add a label to a graph

How to edit a label

How to delete a label

How to move a label

Pictures

How to add a picture to a graph

How to move a picture

How to delete a picture

How to replace a picture

Graph menu—Kinetics
Introduction

Use the Graph menu to view data in various graphical formats or in multiple graphs in the Graphics area. You can double-click on the Graphics area to toggle the display through graph and report to graph only.

In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area surrounding the graph is referred to as the graph box. A graph will appear in the Graphics area when you select Start to commence a Kinetics run.

The Graph menu can be accessed in the following ways:

- from the main menu at the top of the Kinetics Application window
- by clicking the right mouse button in a graph box or the Graphics area

**Note**
The red trace in any graph box is the one on which all information appears and the one used for the cursor tracking. It is called the focused trace.

Menu commands

The following commands can be accessed from the Graph menu:

<table>
<thead>
<tr>
<th>Menu command</th>
<th>Button</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Print XY point</td>
<td></td>
<td>Select this to print the cursor's current X and Y coordinates in the report area.</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Note:</strong> This option is only available from the Graph menu that appears when you right-click in a graph box.</td>
</tr>
<tr>
<td>Copy Graph</td>
<td>Select Copy Graph to copy the entire Graphics area to the Windows Clipboard. All of the graphs in the Graphics area will be copied to the Clipboard.</td>
<td></td>
</tr>
<tr>
<td>Paste to Graph</td>
<td>Select Paste to Graph to paste the contents of the Windows Clipboard into the Graphics area. Text is pasted as a label, graphics are pasted as pictures.</td>
<td></td>
</tr>
<tr>
<td>Add Graph</td>
<td>Select Add Graph to add a new graph box to the Graphics area. You may want to do this if you have, for example, a graph containing existing data and you want to collect some new data in a new graph.</td>
<td></td>
</tr>
<tr>
<td>Remove Graph</td>
<td>Select Remove Graph to delete the selected graph box from the Graphics area. The graph box is removed, but the data is not deleted and is still available from the Trace Preferences window. You may want to use remove graph if you have a large number of graph boxes open but only want to focus on some of these. <strong>Note:</strong> You can remove multiple graph boxes by first holding down the CTRL key and selecting the graphs to be removed and then selecting the Remove Graph button or menu item. Use the SHIFT key to highlight a range of graphs for removal.</td>
<td></td>
</tr>
<tr>
<td>Clear All Traces</td>
<td>Select Clear All Traces to delete all traces from the graph/s. Note that selecting this option simply turns off the Visible property for the traces in the Trace Preferences window. You can redisplay the traces by making the traces Visible again in the Trace Preferences window.</td>
<td></td>
</tr>
<tr>
<td>Add Label</td>
<td>Select <strong>Add Label</strong> to display the <strong>Add Label</strong> dialog where you can enter a text label to be placed on a graph or in the background of the Graphics area.</td>
<td></td>
</tr>
<tr>
<td>-----------</td>
<td>--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------</td>
<td></td>
</tr>
<tr>
<td>Add Picture</td>
<td>Select <strong>Add Picture</strong> to display the <strong>Add Picture</strong> dialog where you can select a picture, bitmap or chemical structure to be placed on a graph or in the background of the Graphics area.</td>
<td></td>
</tr>
</tbody>
</table>
| Edit Annotation | Select **Edit Annotation** to alter the content or look of the selected annotation, (either a label or a picture) in the Graphics area.  
**Note:**  
This option can only be activated if an annotation is selected.  
If the selected annotation is a label, selecting **Edit Annotation** will open the **Edit Label** dialog where you can change the text of the selected label.  
If the selected annotation is a picture, selecting **Edit Annotation** will open the **Edit Picture** dialog where you can replace the selected picture with a new picture. |
| Delete Annotation | Select **Delete Annotation** to remove an annotation, (such as a label or a picture) from the Graphics area.  
You can use this menu option to remove highlighted annotations in the Graphics area such as labels, pictures and ruler/fit information.  
**Note:**  
This option can only be activated if an annotation is selected. |
| Trace Preferences | Select this command to display the **Trace Preferences** dialog where you can select the trace(s) to display in the selected graph. |
| Graph Preferences | Select this command to display the **Graph Preferences** dialog where you can change the appearance of your graph. Any changes you make to the appearance of the graph will be applied to all graphs. |
| Cursor Mode… | Select this command to display the **Cursor Modes** dialog where you can choose the type of cursor to be used on the screen. For example, you can select to use a free cursor that can move anywhere in the graph or exactly trace the focused scan (red scan). Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the **Least Squares** method or the **Point to Point** method. |
| Axes Scales… | Select this command to display the **Axes Scales** dialog where you can set the scale range for the X and Y axes. |
| Auto scale graph | To automatically scale the graph, point to one of the following options:  

**Autoscale[XY]**  
Select Autoscale[XY] to automatically scale collected data by scaling the Y height and X width to full screen.  

**Autoscale[X]**  
Select Autoscale[X] to automatically scale collected data by scaling the X width to full screen.  

**Autoscale[Y]**  
Select Autoscale[Y] to automatically scale collected data by scaling the Y height to full screen. |
| Zoom Out | Select **Zoom Out** to Zoom back to the original X and Y ranges which were used to collect the data (i.e. the method setup conditions of the focused scan).  
For example, the system will revert back to the X and Y ranges set in the method if |
you zoom in and out during a data collection.

Single/Multi Graph

Select **Single/Multi Graph** to toggle between displaying one or all graphs.

**Note:**
The graph highlighted in the **Multi Graph** view will be displayed when you toggle the **Single Graph** option.

Auto Arrange Graphs

Select **Auto Arrange Graphs** to view and arrange all graphs simultaneously.

**Note:**
You can move graph boxes manually by clicking the left mouse button outside the graph axes and dragging the graph box to a new position. You can also resize graphs.

User Data Form

Select **User Data Form** to display the **User Data Form** where you can enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data, such as the pH at which data was collected.

Calculator

Select **Calculator** to display the **Maths** window where you can perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator.

History List

This displays a list of the current graphs.

See also:
- Add Label
- Add Picture
- Edit Label
- Edit Picture
- Trace Preferences
- Graph Preferences
- Cursor Modes
- Axes Scales
- Single/Multi Graph
- User Data Form
- Maths
- Graphics toolbar
- Graphics area

**Graphics Toolbar—Kinetics**

Use the Graphics toolbar buttons to gain quick access to many of the **Graph** menu options. You can also use the toolbar to select the graph you want to view and to change the look of your graphs.

To display or hide the toolbar, point to the **Toolbars** option in the **View** menu and select **Graphics**. To display or hide the toolbar's text labels, point to the **Toolbars** option in the **View** menu and select **Text Labels**.

**Hot Tip**
To see the name of a button, point to it with the mouse.

The following commands can be accessed from the Graphics toolbar:
<table>
<thead>
<tr>
<th>Tool</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Trace Preferences</strong></td>
<td>Click this button to display the Trace Preferences window where you can select the trace(s) to display in the selected graph.</td>
</tr>
<tr>
<td><strong>Graph Preferences</strong></td>
<td>Click this button to display the Graph Preferences dialog where you can change the appearance of your graph. Any changes you make to the appearance of the graph will be applied to all graphs.</td>
</tr>
<tr>
<td><strong>Cursor Mode</strong></td>
<td>Click this button to display the Cursor Modes dialog where you can choose the type of cursor to be used on the screen. For example, you can select to use a free cursor that can move anywhere in the graph or exactly trace the focused scan (red scan). Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the Least Squares method or the Point to Point method.</td>
</tr>
<tr>
<td><strong>Axes Scales…</strong></td>
<td>Click this button to display the Axes Scales dialog where you can set the scale range for the X and Y axes.</td>
</tr>
<tr>
<td><strong>Autoscale [XY]</strong></td>
<td>Click the Autoscale[XY] button to automatically scale collected data by scaling the Y height and X width to full screen.</td>
</tr>
<tr>
<td><strong>Autoscale [X]</strong></td>
<td>Click the Autoscale[X] button to automatically scale collected data by scaling the X width to full screen.</td>
</tr>
<tr>
<td><strong>Autoscale [Y]</strong></td>
<td>Click the Autoscale[Y] button to automatically scale collected data by scaling the Y height to full screen.</td>
</tr>
<tr>
<td><strong>Zoom Out</strong></td>
<td>Click the Zoom Out button to Zoom back to the original X and Y ranges which were used to collect the data (i.e. the method setup conditions of the focused scan). For example, the system will revert back to the X and Y ranges set in the method if you zoom in and out during a data collection.</td>
</tr>
<tr>
<td><strong>Add Label</strong></td>
<td>Click this button to display the Add Label dialog where you can enter a text label to be placed on a graph or in the background of the Graphics area.</td>
</tr>
<tr>
<td><strong>Add Picture</strong></td>
<td>Click this button to display the Add Picture dialog where you can select a picture, bitmap or chemical structure to be placed on a graph or in the background of the Graphics area.</td>
</tr>
</tbody>
</table>
| **Single/Multi Graph** | Select Single/Multi Graph to toggle between displaying one or all graphs.  
  **Note:** The graph highlighted in the Multi Graph view will be displayed when you toggle the Single Graph option. |
| **Auto Arrange Graphs** | Click the Auto Arrange Graphs button to view and arrange all graphs simultaneously.  
  **Note:** You can move graph boxes manually by clicking the left mouse button outside the graph axes and dragging the graph box to a new position. You can also resize graphs. |
| **Add Graph** | Click this button to add a new graph box to the Graphics area. You may want to do this if you have, for example, a graph containing existing data and you want to collect some new data in a new graph. |
| **Remove Graph** | Click this button to delete the selected graph box from the Graphics area. The graph box is removed, but the data is not deleted and is still available from the Trace Preferences window. You may want to use remove graph if you have a large number of graph boxes open. |
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but only want to focus on some of these.
Note:
You can remove multiple graph boxes by first holding down the CTRL key
and selecting the graphs to be removed and then selecting the Remove Graph
button or menu item. Use the SHIFT key to highlight a range of graphs for
removal.
Clear All
Graphs

Click this button to delete all traces from the graph/s. Note that selecting this option
simply turns off the Visible property for the traces in the Trace Preferences
window. You can redisplay the traces by making the traces Visible again in the
Trace Preferences window.

User Data
Form

Click this button to display the User Data Form where you can enter extra
information about the traces in the highlighted graph box. For example, you might
want to record information about the experimental conditions you used to collect the
data, such as the pH at which data was collected.

Calculator

Click this button to display the Maths window where you can perform mathematical
manipulations on collected traces. In addition, it can act as a simple numeric
calculator.

See also:
Add Label
Add Picture
Edit Label
Edit Picture
Trace Preferences
Graph Preferences
Cursor Modes
Axes Scales
Single/Multi Graph
User Data Form
Maths
Graphics area
Graph menu
Graphics How to

Graph Labels—Kinetics
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Add Label dialog
Edit Label dialog
How to add a label to a graph
How to edit a label
How to move a label
How to resize a label
How to delete a label

Add Label—Kinetics
Use the Add Label dialog to place a label on your graph or into the background of the Graphics area.
To add a label to a graph box, you must have at least one trace Visible in the graph box. (You can select which
traces you want to make visible in the Trace Preferences dialog.) If you are viewing multiple graphs, the label


will be placed in the graph box that is currently selected. However, you can add a label outside of a graph box in the background of the Graphics area at any time.

**Q.** Can I add a label that will print with all my graphs.

**A.** Yes, you can add your label to the background of the Graphics area. To do this, right-click on a section of the background (gray area) and select **Add Label** from the menu. (You will notice that this menu has less options than the menu that appears when you right-click on a graph.) In the **Add Label** dialog, enter the label to be added to all graphs and select OK. The label will appear in the background of the Graphics area.

To access the **Add Label** dialog, select the **Add Label** command from the **Graph** menu, or select the **Add Label** button on the Graphics toolbar.

The **Add Label** dialog comprises the following:

| **To selected** | **Select this option to associate a label with the selected trace. When you associate a label with a trace and make the trace visible in the Graph box, the label will also be displayed with the trace.** |
| **label** | **To selected** | **Select this option to associate the label with the selected graph. When you associate a label with a graph, the label will always be displayed in the Graph box, regardless of what traces are visible.** |
| **trace** | **Selected trace** | **This will identify the trace that is currently selected and display the name of the trace.** |
| **graph** | **Font** | **Click Font to open the Windows Font dialog. In this dialog you can specify the font type, style and size to be used for the label.** |
| | **Label** | **Use the Label field to enter the text for the label. You can enter as many lines of text as you require.** |

**Hot Tip**

If you wish to include certain XY co-ordinates on the graph as a label, position your cursor at the appropriate place, right-click and select **Add Label** from the menu. When the **Add Label** dialog appears, the co-ordinate positions will be added to the **Label** field.

**Note**

When you save the data (as a Batch or Data file) the label is saved with its associated trace or graph. Labels outside graph boxes can be saved in a graphics template. To do this, choose **Graph Template** as the **Save As** type in the **Save As** dialog. See **Example of a Graphics Template**

See also:

- **Trace Preferences dialog**
- **Graph menu**
- **Graphics toolbar**
- **How to add a label to a graph**
- **How to move a label**
- **How to edit a label**
- **How to resize a label**
- **How to delete a label**

**Edit Label—Kinetics**

Use the **Edit Label** dialog to alter the content or look of your label.

To access the **Edit Label** dialog, right-click on the label you wish to change and select **Edit Annotation** from
the menu.

The dialog comprises the following:

**Font**
Select **Font** to open the Font dialog, where you can specify the font type, style and size to be used for the label.

**Label**
Use the **Label** field to alter the text for the label.

See also:
- [How to edit a label](#)
- [How to add a label to a graph](#)
- [How to resize a label](#)
- [How to move a label](#)

## How to add a label to a graph—Kinetics

You can add a label to your graph or Graphics area background. To do this:

**Step 1**

Click on the graph box you require or click outside the graph box(es) to choose the Graphics area background.

**Note**
There must be at least one trace Visible in the graph box before you can add a label to it.

**Step 2**

Click the **Add Label** button on the toolbar or right-click on the graph and select **Add Label** from the menu. The **Add Label** dialog will appear. The **Add Label** command is also accessible from the **Graph** menu.

**Step 3**

Click the **Font** button. The Windows **Font** dialog appears. Select the font type, style and size you require for your label. A sample of the font style appears in the **Sample** field. When you are satisfied with your selections, select **OK** to return to the **Add Label** dialog.

**Step 4**

Enter the label in the **Label** field and select **OK**. The label will be displayed on the selected graph. If necessary, you can move a label so that it is more appropriately placed on your graph.

See also:
- [Graphics Area](#)
- [How to move a label](#)
- [How to edit a label](#)
- [How to resize a label](#)

## How to edit a label—Kinetics

**Step 1**

Place the cursor on the label and click the left mouse button to select the label.
Step 2

Click the right mouse button and select **Edit Annotation** from the drop-down menu. Alternatively you can select the **Edit Annotation** command from the **Graph** menu. The **Edit Label** dialog is displayed.

Step 3

Use the **Edit Label** dialog to change the label.

For example, if you want to change the font size of the label, select the **Font** button. In the **Size** field, scroll through the available sizes for the font and select the size you require. Alternatively, highlight the currently selected size in the **Size** entry field and enter the size you require. Click the **OK** button in this dialog to return to the **Edit Label** dialog.

Step 4

Click **OK** to clear the **Edit Label** dialog. Your label will appear with the new font size.

See also:
- [How to add a label to a graph](#)
- [How to move a label](#)
- [How to resize a label](#)
- [How to delete a label](#)

### How to move a label—Kinetics

To move a label so that it is more appropriately placed on your graph or Graphics area background, do the following:

Step 1

Place the cursor on the label.

Step 2

Click and hold down the left mouse button.

**Hot Tip**

If you are having trouble selecting the label, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 3

Drag the label to the correct position and release the mouse button. The label will be displayed at the new position.

**Note**

Labels are transparent so as not to interfere with any traces on graphs.
How to resize a label—Kinetics

To resize a label so that it is more appropriately sized on your graph or Graphics area background, do the following:

Step 1

Select the label by clicking on it with the mouse. A border will be displayed around the label to indicate it is selected.

Hot Tip
If you are having trouble selecting the label, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Position the cursor over one of the corner edges of the label until the cursor becomes a diagonal double-headed arrow. Click and hold down the left mouse button.

Example of label showing the border that indicates it is selected and the arrow to be used for re-sizing

Step 3

Increase or decrease the size of the label by dragging the mouse. The label text will also be resized. Release the mouse button when the label and text are the correct size.

Note
Labels are transparent so as not to interfere with any traces on graphs.

How to delete a label—Kinetics

To delete a label from a graph or Graphics area background, do the following:

Step 1

Position the cursor on the label and click the left mouse button to select it.

Hot Tip
If you are having trouble selecting the label it may be that a trace is interfering with the label selection. Try zooming in and clicking in a different part of the label.

Step 2

Click with the right mouse button and select **Delete Annotation** from the drop-down menu displayed. Alternatively, you can select the **Delete Annotation** command from the **Graph** menu. The label will be deleted from the graph or Graphics area.

See also:
- How to add a label to a graph
- How to move a label
- How to edit a label
- How to resize a label

**Graph Pictures—Kinetics**

- **Add Picture dialog**
- **Edit Picture dialog**
- **How to add a picture**
- **How to replace a picture**
- **How to move a picture**
- **How to resize a picture**
- **How to delete a picture**

**Add Picture—Kinetics**

Use the **Add Picture** dialog to insert a picture or bitmap into the Graphics area. The picture can be added as a background picture or to a particular graph box.

To add a picture to a graph box you must have at least one trace **Visible** in the graph box. (You can select which traces you want to make visible in the **Trace Preferences** dialog.) If you are viewing multiple graphs, the picture will be placed in the graph box that is currently selected. However, you can add a picture outside of a graph box in the Graphics area at any time.

**Q.** Can I add a picture that will print with all my graphs.
**A.** Yes, you can add your picture to the background of the Graphics area. To do this, right-click on a section of the background and select **Add Picture** from the menu. (You will notice that this menu has less options than the menu that appears when you right-click on a graph.) In the **Add Picture** dialog, load the picture to be added to all graphs and select **OK**. The picture will appear in the background of the Graphics area.

To access the **Add Picture** dialog, select the **Add Picture** command from the **Graph** menu, or select the **Add Picture** button on the Graphics toolbar.

The **Add Picture** dialog comprises the following commands:

- **To selected trace** Select this option to associate a picture with the selected trace. When you associate a picture with a trace and make the trace visible in the Graph box, the label will also be displayed with the trace.
To selected graph

Select this option to associate the picture with the selected graph. When you associate a picture with a graph, the label will always be displayed in the Graph box, regardless of what traces are visible.

Load

Click **Load** to open the Windows **Open** dialog. Use this dialog to browse your computer for the picture you want to insert. The picture must be one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.

If you are viewing multiple graphs the picture will be placed on the graph that is currently selected. If no graph is selected, the picture will be placed on the Graphics area background.

**Note**

When you save the data (as a Batch or Data file) the label is saved with its associated trace or graph. Labels outside graph boxes can be saved in a graphics template. To do this, choose **Graph Template** as the **Save As** type in the **Save As** dialog.

See 'Example of a Graphics Template'.

See also:

- **Trace Preferences**
- **Graph menu**
- **Graphics toolbar**
- **How to add a picture to a graph**
- **How to move a picture**
- **How to delete a picture**
- **How to resize a picture**
- **How to replace a picture**

**Edit Picture—Kinetics**

Use the **Edit Picture** dialog to change a picture you have inserted into the Graphics area.

To access the **Edit Picture** dialog, right-click on the picture you wish to change and select **Edit Annotation** from the menu.

The **Edit Picture** dialog comprises the following commands:

**To selected trace/graph**

Displays whether you originally selected to add the picture to a trace or a graph. This option is not editable.

**Load**

Click **Load** to open the Windows **Open** dialog. Use this dialog to browse your computer for the picture you want to insert. The selected picture will replace the current picture. The picture must be one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.

See also:

- **How to replace a picture**
- **How to add a picture**
- **How to move a picture**
- **How to resize a picture**
- **How to delete a picture**

**How to add a picture to a graph—Kinetics**

You can add a picture to your graph or the Graphics area background. To do this:

**Step 1**
Click on the graph box you require or click outside the graph box(es) to choose the Graphics area background.

**Note**
There must be at least one trace Visible in the graph box before you can add a picture to it.

**Step 2**

Click the **Add Picture** button on the toolbar or right-click on the graph and select **Add Picture** from the menu. The **Add Picture** dialog will appear. The **Add Picture** command is also accessible from the **Graph** menu.

**Step 3**

Click the **Load** button. The Windows **Open** dialog appears.

**Step 4**

Browse through your directories to find the picture you want to insert. All pictures must be in the bitmap format and will have the extension .BMP.

**Step 5**

Highlight the .BMP that you want to insert and select **Open**.

**Step 6**

The **Add Picture** dialog will appear showing a preview of the bitmap. If the bitmap is the correct one select the **OK** button. The bitmap will be placed onto the graph.

**Note**
You should take care not to add a bitmap that is too big as it will obscure part or all of your graph.

You can move a picture so that it is more appropriately placed on your graph.

**See also:**
[How to move a picture to a graph](#)
[How to delete a picture](#)
[How to replace a picture](#)
[How to resize a picture](#)

### How to replace a picture—Kinetics

To change a picture that you have added to a graph or Graphics area background do the following:

**Step 1**

Move the cursor to the picture and select the left mouse button to select it.

**Hot Tip**
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.
Step 2

Click the right mouse button and select **Edit Annotation** from the drop-down menu or select **Edit Annotation** from the **Graph** menu. The **Edit Picture** dialog is displayed.

Step 3

From the **Edit Picture** dialog, select the **Load** button. The Windows **Open** dialog is displayed.

Step 4

Browse through your directories to find the picture to replace the current bitmap. Picture files must be in one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.

Step 5

Highlight the file that you want to insert and select **Open** to return to the **Edit Picture** dialog.

Step 6

The **Edit Picture** dialog will display a preview of the bitmap. If the bitmap is the correct one select the **OK** button. The bitmap will be placed onto the graph. If you need to, move the picture so that it is more appropriately placed on your graph.

**Note**

Do not to add a bitmap that is too big as it will obscure part or all of your graph.

**See also:**

- How to add a picture to a graph
- How to move a picture
- How to delete a picture
- How to resize a picture

**How to move a picture—Kinetics**

To move a picture so that it is more appropriately placed on your graph or Graphics area background, do the following:

Step 1

Place the cursor on the picture.

Step 2

Click and hold down the left mouse button.

**Hot Tip**

If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 3

Drag the picture to the correct position and release the mouse button. The picture will be displayed at the new
How to resize a picture—Kinetics

To resize a picture so that it is more appropriately sized on your graph or Graphics area background, do the following:

Step 1

Select the picture by clicking on it with the mouse. A border will be displayed around the picture to indicate it is selected.

Hot Tip
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Position the cursor over one of the corner edges of the picture until the cursor becomes a diagonal double-headed arrow. Click and hold down the left mouse button.

Example of picture showing the border that indicates it is selected and the arrow to be used for re-sizing

Step 3

Increase or decrease the size of the picture by dragging the mouse. The picture will also be resized. Release the mouse button when the picture is the correct size.

How to delete a picture—Kinetics

To delete a picture from a graph or Graphics area background, do the following:

Step 1
Position the cursor on the picture and click the left mouse button to select it.

**Hot Tip**
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

**Step 2**

Click with the right mouse button and select **Delete Annotation** from the drop-down menu displayed. Alternatively, you can select the **Delete Annotation** command from the **Graph** menu. The picture will be deleted from the graph or Graphics area.

See also:
- How to add a picture to a graph
- How to move a picture
- How to zoom a graph
- How to resize a picture

**Trace Preferences—Kinetics**

A single graph box can display many traces. This window enables you to select the trace(s) to display (make **Visible**) in the selected graph box.

To access the **Trace Preferences** dialog, select the **Trace Preferences** command from the **Graph** menu, or select the **Trace Preferences** button on the Graphics toolbar.

The dialog comprises the following:

**Hot Tip**
The **Trace Preferences** menu is also available on clicking the right mouse button anywhere in the trace listing.

**Trace Preferences table**

<table>
<thead>
<tr>
<th><strong>Visible</strong></th>
<th>Use the <strong>Visible</strong> field to select which traces will appear on the selected graph. To select or clear the <strong>Visible</strong> field, simply click the left mouse button in this column. A tick will appear next to traces that are to be displayed in the selected graph.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>If you are in single graph view, all selected traces will appear on the one graph. If you are in <strong>Multi Graph</strong> view the traces will appear on the graph that is currently selected.</td>
</tr>
<tr>
<td><strong>Hot Tip</strong></td>
<td>To make all traces Visible, right-click on the table and select the <strong>Select All</strong> command from the menu. Then right-click on the table again and select the <strong>Show Traces</strong> command from the menu.</td>
</tr>
</tbody>
</table>

| **Color** | The **Color** field enables you to view the color of each trace that appears on the selected graph. To change the color of a trace, highlight it, click the right mouse button in the **Trace Preferences** table and select **Change Trace Color**. You can then select a color from the **Color** dialog that is displayed. |
The red trace in any graph box is the one on which all information appears. This trace, which is used for the cursor tracking, is called the focused trace and it is always red. When you select a trace in the **Color** column it will become the focused trace in the selected graph box.

**Note**
When you change the trace line width in the **Graph Preferences** dialog it also will change the width of the color trace line in this dialog.

**Name of trace**  The **Name of Trace** field contains a description of the traces currently available for display in the selected graph box.

**Filename**  This field displays the filename and path for the trace. If the name is too long to appear in the field you can use the keyboard arrow keys to scroll to the end of the filename or you can resize the **Trace Preferences** window. If the file path for a trace shows 'not saved' then the trace is unsaved data and is only stored in memory.

**Trace Audit Log**

The **Trace Audit Log** displays information about the highlighted trace. This includes instrument parameters and data form information relating to the trace that has been selected. It also logs all information about data manipulation. When more than one trace is highlighted, the **Trace Audit Log** will display information about the trace that was first selected.

**Trace Preferences menu—Kinetics**

The **Trace Preferences** menu is displayed when you click the right mouse anywhere on the **Trace Preferences** table. The menu contains the following commands:

- **Select All**  This selects or highlights all the traces in the listed on the **Trace Preferences** page.
- **Select None**  This de-selects all the traces in the listing.
- **Show Traces**  Select **Show Traces** to make all of the traces currently highlighted Visible, meaning that they will be displayed in the current graph box.
- **Hide Traces**  Select **Hide Traces** to remove the Visible property from all the traces currently highlighted, which will remove them from the current graph box.
- **Delete Traces**  Select **Delete Traces** to delete all the highlighted traces in all graphs. Note that if you save the data file before you select **Delete Traces**, the traces are not deleted from the saved file, and they can be retrieved at a later date. Selecting **Delete Traces** will delete the traces from the application memory, and it is recommended that you select this option if you need to free up memory on your computer. This is especially relevant for the Kinetics application where the data files can be quite large.

**Note**
If the application is controlled by the GLP Administration application, you will be prompted to save an unsaved data file before it is removed from the application.
How to select the focused trace—Kinetics

To select the scan that you want to be the focused (red) trace, do one the following:

- From the Graph, click the left mouse button on the trace that you want to make the focused trace.
- From the Graph Legend (if displayed), click the left mouse button on the trace that you want to make the focused trace.
- From the Trace Preferences dialog, click in the Color column of the trace that you want to make the focused trace.

Note
Select the Show Legend check box in the Graph Preferences dialog to display the Graph Legend.

See also:
Trace Preferences page
Graph Preferences page
How to remove a trace from the trace list
How to remove a trace from a graph

How to display a trace that is not appearing in the graph box—Kinetics

Step 1
Click on the graph in the Graphics area.

Step 2
Select the Trace Preferences button from the Graphics toolbar, or select Trace Preferences from the Graph menu. The Trace Preferences dialog is displayed.

Step 3
Find that trace that you wish to see and select the Visible option. A tick will appear in the Visible column to indicate that it will now be displayed.

If you wish to make multiple traces visible, do the following:

a. Hold down the CTRL key select all the traces that you want to display.

b. Click the right mouse button and select the Show Traces command from the drop-down menu. Ticks will appear next to the selected traces to indicate that they will now be displayed.
Step 4

Press **OK** to clear the **Trace Preferences** dialog. The selected trace(s) will be displayed.

See also:
- How to remove a trace from a graph
- How to remove a trace from the trace list

**How to remove a trace from a graph—Kinetics**

Step 1

Select a graph in the Graphics area.

Step 2

Click the **Trace Preferences** button on the Graphics toolbar or select **Trace Preferences** from the **Graph** menu. The **Trace Preferences** dialog is displayed.

Step 3

Find that trace that you no longer wish to see and select the **Visible** option. The tick will be removed from the **Visible** column to indicate that it will not be displayed.

If you wish to make multiple traces visible, do the following:

a. Hold down the CTRL key select all the traces that you no longer want to display.

b. Click the right mouse button and select the **Hide Traces** command from the drop-down menu. The ticks will be removed from the selected traces to indicate that they will not be displayed.

Step 4

Select **OK** to clear the **Trace Preferences** dialog. The trace will be removed from the graph.

See also:
- How to display a trace that is not appearing in the graph box
- How to remove a trace from the trace list

**How to remove a trace from the trace list—Kinetics**

If your graphs contain unwanted traces, you may wish to delete them (rather than simply making them invisible) in order to free up application memory and aid system performance.

Step 1

Select a graph in the Graphics area.

Step 2

Click the **Trace Preferences** button on the Graphics toolbar or select **Trace Preferences** from the **Graph** menu. The **Trace Preferences** dialog is displayed.
Step 3

Select the trace to be removed by clicking on it.

Step 4

Click the right mouse button to display the *Trace Preferences* menu.

Step 5

Select **Delete Traces** to delete the sample from the trace list. You will be prompted to confirm this action.

Step 6

Select **OK** to clear the *Trace Preferences* dialog. The trace will be removed from all graphs.

See also:
- How to remove a trace from a graph
- How to display a trace that is not appearing in the graph box

**Graph Preferences—Kinetics**

Introduction

Use the *Graph Preferences* dialog to change the appearance of your graph.

All graphs will appear labeled with the X axis label being horizontal and the Y axis vertical. You cannot change the orientation of these labels. However you can alter the font and size of the axes and their labels.

Any changes you make to the appearance of the graph will apply to all graphs until the settings are changed again.

To access the *Graph Preferences* dialog, select the *Graph Preferences* command from the *Graph* menu, or select the *Graph Preferences* button on the Graphics toolbar.

Dialog controls

The *Graph Preferences* dialog comprises the following:

**Axes Preferences**

<table>
<thead>
<tr>
<th><strong>Axes Color</strong></th>
<th>Select the color that you want to assign to the axes of the graph/s from the drop down list.</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Axes Width</strong></td>
<td>Select the width of the axes from the drop down list. The width, which is in pixels, ranges between one and five pixels.</td>
</tr>
<tr>
<td><strong>Grid Style</strong></td>
<td>Select the style of grid for the graph from the drop down list. If you do not wish to include a grid on the graph, select None.</td>
</tr>
<tr>
<td><strong>Plot Area Color</strong></td>
<td>Select the color to be used as the background of the graph from the drop down list. If you do not want a background color on the graph, select White.</td>
</tr>
</tbody>
</table>
**Trace Width**  
Select the width of the trace from the drop down list. The width is given in pixels and ranges between one and five pixels.

**Font**  
Click the **Font** button to display the Windows **Font** dialog where you can define the font style for the axes of the graph/s. The style and size of the font will be applied to all graphs. The style, name and point size of the font appears on the font button.

**Preview Box**  
Preview any changes you make to the axes preferences in the preview box. The changes shown in this box will not be made until you select **OK**.

**Advanced Options**

**Auto tick marks**  
The **Auto tick marks** option enables you to approximately set how many tick marks (or points) will be included on the graph axes. The tick marks generated will be whole, rounded, even numbers. The **Auto tick marks** option is not available if you have selected **Major ticks**.

**Y-axis**  
Use the Y-axis slider bar to increase or decrease the number of tick marks (or points) to be included on the Y axis.

**X-axis**  
Use the X-axis slider bar to increase or decrease the number of tick marks (or points) to be included on the X axis.

**Major ticks**  
Use the **Major Ticks** option to enable you to set exactly how many ticks (or points) will be added to the X and Y axes. The number of marks you specify will be divided by the axis range, which can result in odd numbers on the axis. Note that any changes are not reflected in the graph bitmap at the top of the screen.

**Decimals**  
Use the **Decimals** option to set the number of X and Y decimal places to be displayed on the axes. To set the number of decimal places, select the **Decimal** option and enter the number in the Y-axis and X-axis combo boxes. Note that any changes are not reflected in the graph bitmap at the top of the screen.

**Trace Style**

**Points**  
Select **Points** to display each individual data point collected by the Cary Eclipse on the graph.

**Points and solid lines**  
Select **Points and solid lines** to display each individual data point collected by the Cary Eclipse on the graph. Additionally, an interpolated line that connects the points will be drawn.

**Solid lines**  
If you select **Solid lines**, the Cary Eclipse system will interpolate the data between the actual, collected data points and show the data as a continuous line.

**Style cycles automatically**  
If you select **Style cycles automatically**, the Cary Eclipse system will display each of the above three trace styles. This makes it easier to differentiate between traces, especially on black-and-white printers.

**Show Legend**
**Show Legend**  Select **Show Legend** to display a legend in the right side of each graph box. A single left click on a sample name or example line within the legend will make it the focused or red scan. Information about the trace will be displayed below the X axis.

**Hot Tip**
Double-clicking on the legend will automatically display the **Trace Preferences** window, allowing you to view more detailed information about your trace (e.g. audit log, File name, etc.). You can also use the legend to select which trace will be the focused trace.

See also:
- [Graph menu](#)
- [Graphics toolbar](#)
- [Graphics area](#)
- [Graphics How To](#)

### How to alter the look of your graphs—Kinetics

**Step 1**

Click the **Graph Preferences** button on the Graphics toolbar, or select **Graph Preferences** from the **Graph** menu. The **Graph Preferences** dialog is displayed.

**Step 2**

Use the available options available to choose your preferred appearance for the graphs. A preview box in the top right-hand corner will demonstrate how the graphs will appear.

**Step 3**

Select OK to clear the **Graph Preferences** dialog and update all graphs to reflect your selections.

**Note**
You can also add labels and pictures to your graphs to alter their appearance.

**Step 4**

To re-use your graphics setup at a later date, save the setup as a graphics template.

See also:
- How to add a label to a graph
- How to add a picture to a graph
- How to move a label
- How to move a picture
- How to resize graph boxes
- How to make a graphics template

### Cursor Modes—Kinetics

Use the Cursor Modes dialog to choose the type of cursor to be used on screen. For example, you can select to use a free cursor that can move anywhere in the graph. Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the Least Squares method or the
Point to Point method.

To access the Cursor Modes dialog, select the Cursor Modes command from the Graph menu, or select the Cursor Modes button on the Graphics toolbar.

The dialog comprises the following:

**Cursor Modes**

- **None**
  - Select this for no cursor tracking.

- **Free**
  - The Free cursor is not confined to a particular trace on the graph. It can move anywhere in a graph box, and its ‘X,Y’ position is shown in the bottom right-hand side of the Graphics area. You can use the Free cursor to zoom in on a particular part of a graph.
  
  **Note**
  - The Free cursor moves from pixel point to pixel point in the selected graph box. Hence you need to be careful when positioning the cursor over a scan to obtain an accurate (X,Y) reading. It is recommended that you use the Track cursor mode for this.

- **Track**
  - Use the Track cursor to exactly trace the focused (red) scan in a graph box. The cursor position is displayed in the X and Y at the bottom right-hand side of the Graphics area.
  
  **Hot Tip**
  - If you wish to fix the cursor on a particular position on the curve, move the cursor to the required position. Click the left mouse button, press ‘Alt’ on the keyboard, then release the mouse button.

- **Ruler-Least Squares**
  - This uses a least-squares technique to determine the slope and thus the reaction rate between the start and end points.
  - This kinetics ruler function can be used for measuring zero order reaction rates in kinetics traces. You apply the ruler after kinetics data is collected to manually select the range over which to calculate the reaction rate. The slope is then determined and thus the rate of reaction.

- **Ruler-Point to Point**
  - This uses the slope of the line drawn between two selected points to determine the point to point slope and thus the reaction rate.
  - This kinetics ruler function can be used for measuring zero order reaction rates in kinetics traces. You apply the ruler after kinetics data is collected to manually select the range over which to calculate the reaction rate. The slope is then determined and thus the rate of reaction.

See also:
- About the kinetics ruler
- How to use the kinetics ruler
- How to zoom a graph
- How to track the focused trace

**How to track the focused trace—Kinetics**

To track the focused (red) trace in the graph:

Step 1

Click the **Cursor Modes** button from the Graphics toolbar, or select **Cursor Mode** from the **Graph** menu.
The **Cursor Modes** dialog will appear.

**Step 2**

Select **Track** and select the **OK** button.

**Step 3**

Place the blue tracking cursor in the graph box you require.

**Step 4**

Move the cursor left and right across the graph box to trace the focused (red) scan. The cursor coordinates change (in the X, Y field at the bottom left hand corner of the Graphics area) as you move along the curve.

**Hot Tip**

If you wish to fix the cursor on a particular position on the curve, move the cursor to the required position. Press 'Alt' on the keyboard, then release the mouse button.

**See also:**
- How to select the focused trace
- How to view a particular trace

**Axes Scales—Kinetics**

Use the **Axes Scales** dialog to manually change the axes range. This can assist in zooming in on a particular part of the graph.

To access the **Axes Scales** dialog, select the **Axes Scales** command from the **Graph** menu, or select the **Axes Scales** button on the Graphics toolbar.

The dialog comprises the following:

**X Axis**

- **Mode**
  - View the current units for the X axis.

- **Scale**
  - Use the **Scale** field to enter a new range for the X axis, in the format n1 - n2 (where n1 is the start of the scale and n2 is the end of the scale).

  **Hot Tip**
  - The Cary Eclipse will store the ten most recently entered ranges, which you can access by selecting the down arrow adjacent to the **Scale** field.

**Y Axis**

- **Mode**
  - View the current units for the X axis.

- **Scale**
  - Use the **Scale** field to enter a new range for the Y axis, in the format n1 - n2 (where n1 is the start of the scale and n2 is the end of the scale).

  **Hot Tip**
  - The Cary Eclipse will store the ten most recently entered ranges, which you can access by selecting the down arrow adjacent to the **Scale** field.
See also:
Graph menu
Graphics toolbar
How to scale your graph manually
How to scale your graph automatically
How to zoom a graph

How to scale your graph manually—Kinetics

Step 1

Click on the graph whose axes you want to change to make it the focused trace.

Step 2

Click the Axes Scales button on the toolbar, or select Axes Scales from the Graph menu. The Axes Scales dialog is displayed.

Step 3

Type in the new ranges for the X and Y axes in the format ‘n-n’, or choose an option for each from the drop-down list.

Step 4

Select OK. The axes on the selected graph will change to reflect your chosen settings.

Hot Tip
Select the Zoom Out command to resize the graph to its original settings.

How to scale your graph automatically—Kinetics

Step 1

Click on the graph whose axes you want to change to make it the focused trace.

Step 2

To autoscale the graph, select one of the following:

- Click the Autoscale XY button on the toolbar to autoscale both X and Y axes.
- Click the Autoscale X button on the toolbar to autoscale the X axis only.
- Click the Autoscale Y button on the toolbar to autoscale the Y axis only.

The focused trace will be scaled according to the option selected.
Hot Tip
Select the **Zoom Out** command to resize the graph to its original settings.

See also:
- [How to zoom a graph](#)
- [How to scale your graph manually](#)

**Single/Multi Graph—Kinetics**

You can use the **Single/Multi Graph** command to toggle between a display of either one or multiple graph boxes in the Graphics area.

The command is accessible from the **Graph** menu or by selecting the **Single/Multi Graph** button from the Graphics toolbar.

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Example of a single graph box display

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Example of a multi graph box display

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**Note**
Up to 100 graphs can be displayed in the Graphics area at the one time.

See also:
- [Graph menu](#)
- [Graphics toolbar](#)
- [Graphics How To](#)

**How to make a graphics template—Kinetics**

A graphics template lets you set up and store the style of a graphics report for future use. You can do this by
adding labels and pictures to the background of the Graphics area and altering any axes styles etc. One advantage of this over the application's built-in reporting features is that you have more flexibility as to where you can position graph boxes and any text that you may want to appear (e.g. operator name, batch number etc.) and also where you add pictures, e.g. company logos etc.

Step 1

Start the application and open the data file from which you want to make a graphics template. Alternatively, collect some new data.

Step 2

Make as many graph boxes as you require in your template by using the Add Graph function.

Step 3

Adjust the graph boxes to the correct size and position them as you want them to appear in the template.

Step 4

Do any of the following, if required:

- Change the appearance of the graph boxes in the Graph Preferences dialog
- Use the Add Label function to append text to the Graphics area
- Use the Add Picture function to add pictures, such as your company's logo to the Graphics area

Step 5

From the File menu, select Print Preview. Use the Print Preview dialog displayed to check the template that you have created.

Step 6

When you are satisfied with the appearance of the graphics, select the Save As… command from the File menu. The Save As dialog is displayed.

Step 7

A list of stored data files will appear. Select the down arrow at the right of the Files of type field and select 'Graph Template' to list all the template files. Ensure that the Save only focused trace check box is cleared.

Step 8

Enter a name for the template in the File name field.

Step 9

Select OK to save the graphic template. The template will be used each time you create a new file.

Note

You can also make a graphic template in a similar manner from existing data. No data will be saved in the template.
The ‘ruler’ function in the Kinetics Application enables you to measure zero order reaction rates in kinetics traces. Use the ruler after kinetics data is collected to manually select the range over which to calculate the reaction rate. From this data, the slope the rate of reaction is determined.

To access the Ruler Rate dialog, do the following:

1. From the Graph menu select Cursor Modes. In the Cursor Modes dialog, select Ruler-Least Squares or Ruler-Point to Point option as the cursor mode. See More Information on Least Squares and Point to Point calculations.
2. Select a region of the trace that is relatively straight and place the cursor on the point of the trace where you wish to start drawing the ruler. Click and hold down the left mouse button and drag the cursor to the second point on the trace. A ruler line will draw as you move the cursor.
3. Release the mouse button. The Ruler Rate dialog is displayed.

The Ruler Rate dialog comprises the following:

- **Slope**: Displays the slope of the line of best fit in the region you selected.
- **X Start/Stop**: Displays the beginning and end X co-ordinates of the region you selected.
- **Y Start/Stop**: Displays the beginning and end Y co-ordinates of the region you selected.
- **Retry**: Select Retry to re-select a region of the trace.
- **Add Label**: Select Add Label to add a label to the graph that displays the slope of the line you have selected.

**More information on Least Squares and Point to Point calculations**

**Least squares slope calculation**

You can calculate the rate of a zero order reaction by manually selecting the range over which to calculate the reaction rate of a kinetics plot using the ruler on a particular graph.

A linear equation is fitted to the data using a Ruler-Least Squares technique. The slope of the line of best fit is reported as the kinetics rate in the Ruler Rate dialog using the previous zero order equation.

**Point to point slope calculation**

You can choose to calculate the point to point slope instead of the least squares slope value.

The point to point slope is calculated from the slope of a line drawn between two ordinate values at two specified abscissa points.
If you choose OK or Add Label in the Ruler Rate dialog then a corresponding Results table will appear in the Report area that includes the manually selected abscissa, ordinate range, calculated reaction rate (Slope) and Standard Deviation (SD).

The SD results means that the system looks at the difference between the actual Y value and the fitted Y value after the linear least squares fit or point to point fit has been applied. The system takes the difference of each point over the selected region and calculates the SD using the following equation:

\[ S = \sqrt{\frac{1}{n-2} \sum_{i=1}^{n} (f_{m,i} - f_{c,i})^2} \]

where,

- \( f_{m,i} \) are measured values
- \( f_{c,i} \) are calculated values
- \( n \) is the number of points

The Factor* Slope result is also displayed. This is the product of the Factor multiplied by the calculated kinetics ruler slope. The Factor value (entered in the Analyze tab) also appears above the result table in the report.

**Note**

You can only perform a manual zero order rate calculation after the data is collected. Once the data is collected, select Cursor modes in the Graph menu and choose the kinetics ruler type you require. You can then select the range over which to calculate the reaction rate.

**See also:**

How to use the kinetics ruler

**How to use the kinetics ruler—Kinetics**

The ‘ruler’ function can be used for measuring zero order reaction rates in kinetics traces. You can use the ruler after kinetics data is collected to manually select the range over which to calculate the reaction rate. The slope is then determined and thus the rate of reaction.
To use the ruler:

Step 1

From the Graph menu, select **Cursor Modes**. Alternatively, select the button on the toolbar. The **Cursor Modes** dialog is displayed.

Step 2

Select **Ruler—Least Squares** to use a least-squares technique to determine the slope and thus the rate of the reaction over the selected range. Alternatively, select **Ruler—Point to Point** to use a line drawn between two selected points to determine the point to point slope and thus the reaction rate.

Step 3

Select **OK** to clear the **Cursor Modes** dialog and return to the Kinetics Application window.

Step 4

Select a region of the trace that is relatively straight and then place the cursor on the first point on the trace where you wish to start drawing the ruler. Press and hold down the left mouse button.

Step 5

Move the cursor to the second point on the trace (you will see the ruler line drawing as you move the cursor) and release the mouse button.

The **Ruler Rate** dialog will be displayed to enable you to view the start and end positions of the calculation range that you selected. The value of the calculated slope is also displayed.

If you are not satisfied with the rate result, select the **Retry** button to repeat Steps 3 to 5, this time selecting another range.

**Note**

If you wish to exit the kinetics ruler mode, select **Cancel**. The kinetics ruler trace will be removed from the graph.

Step 7

Select **Add Label** to add the determined slope value of the ruler trace to the graph. The **Ruler Rate** dialog will be cleared.

If you do not wish to add a label, simply select **OK** to clear the **Ruler Rate** dialog.

The results of the manual determination of the reaction rate will appear in a table in the Report area. The ordinate and abscissa start and end values are reported as well as the calculated slope. The result table will also include the calculated standard deviation as well as the Factor*slope result.

See also:

**Cursor modes**
Examples—Kinetics

- User Collect
- Multi-stage kinetics run
- Multi-stage kinetics calculations
- Single-stage, multicell kinetics run
- Example of a graphics template
- Using the stop time

Example of a multi-stage kinetics run—Kinetics

Number of Stages = 3, Single Cell kinetics

<table>
<thead>
<tr>
<th>Stage</th>
<th>Cycle (min)</th>
<th>Stop (min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2</td>
<td>10</td>
</tr>
<tr>
<td>2</td>
<td>4</td>
<td>26</td>
</tr>
<tr>
<td>3</td>
<td>5</td>
<td>45</td>
</tr>
</tbody>
</table>

The Ave Time is set to 0.500 s

Graphical representation of data points for the above example

Stage 1: 0-10 minutes. The run will commence taking data readings at the set Ave Time of 0.5 s and cycle every 2 minutes. Five cycles will be completed in the first 10 minutes (Stop Time 10 mins) of the run.

Stage 2: 10-26 minutes. The run will continue taking data readings at the set Ave Time of 0.5 s but cycle every 4 minutes. Four cycles will be completed in the next 16 minutes (Stop Time 26 (10+16) mins) of the run.

Stage 3: 26-45 minutes. The run will continue taking data readings at the set Ave Time of 0.5 s but cycle every 5 minutes. Four cycles will be completed in the next 20 minutes (Stop Time 45 (10+16+20) mins) of the run.

See also:
- Example using the Stop time
- Example of a User Collect

Example of multi-stage kinetics calculations—Kinetics
The table to follow indicates the regions that would be selected to perform an Advanced analysis, on the above example.

Number of stages (steps) = 2

<table>
<thead>
<tr>
<th>Stage</th>
<th>Start (min)</th>
<th>Stop (min)</th>
<th>Order</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1</td>
<td>2</td>
<td>Zero</td>
</tr>
<tr>
<td>2</td>
<td>4</td>
<td>5</td>
<td>Zero</td>
</tr>
</tbody>
</table>

**Examples of single-stage, multicell kinetics runs using Dwell and Cycle Time—Kinetics**

**Example 1**

Number of Stages = 1, Multicell kinetics (cells chosen = 3), Ave Time = 0.0125 seconds

<table>
<thead>
<tr>
<th>Stage</th>
<th>Dwell (s)</th>
<th>Cycle (s)</th>
<th>Stop (s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.13</td>
<td>6</td>
<td>24</td>
</tr>
</tbody>
</table>
In the example above, the run will commence by taking readings of the first cell for the Dwell Time of 0.13 s. Since the Ave Time is set to 0.0125 s, the Cary will take ten readings during that Dwell Time. The system will then move to the next cell and take further data readings for 0.0125 s for a Dwell Time of 0.13 s. The system will continue to collect data, at cells 2 and 3 in this manner.

The system will then wait until a Cycle time of 6 s has expired before recommencing the run. The run will end at the selected Stop Time of 24 s.

### Example 2

Number of Stages = 1, Multicell kinetics (cells chosen = 6), Ave Time = 0.5 seconds

<table>
<thead>
<tr>
<th>Stage</th>
<th>Dwell (s)</th>
<th>Cycle (min)</th>
<th>Stop (min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2</td>
<td>2</td>
<td>10</td>
</tr>
</tbody>
</table>

Ave Time = 0.5 s  
Dwell time = 2 s  
Cycle time = 2 min  
(Cells chosen = 6)

The run will average the readings every 0.5 s to produce a data point. It will do this for a Dwell Time of 2 s. It will then take data readings of the next five cells using these parameters.

The system will then wait until a Cycle Time of 2 min has elapsed before continuing with further readings of the selected cells. The run will end at the selected Stop Time.

See also:
Example of a Graphics Template—Kinetics

By using the Add Label command in the Cary Eclipse graphics system, you can customize a graphics template such as the one shown below:

The above is created as follows:

- The graph on the left is the collected data.
- The text on the top right is a multi-line label (added using the Add Label command).
- The logo is an inserted picture (added using the Add Picture command).

See also:
Add Label
How to make a graphics template

Example using the Stop time—Kinetics

Example

Cycle time = 5 minutes
Stop time = 30 minutes

In the above example the run will complete 6 cycles of the chosen cells, each with a Cycle time of 5 minutes. The run will end at the selected Stop time of 30 minutes.

Note
If the Stop time was set to 28 mins, the run would continue for 30 mins, as the Cycle time overrides the Stop time.
File Management—Kinetics

- File menu
- Open dialog
- Save As dialog
- Filename extensions
- ASCII spreadsheet file
- Auto Store page
- How to perform post run calculations on previously stored data
- How to combine data files into a batch file
- How to export collected data
- How to combine data files into a batch file

File menu—Kinetics

Use the File menu to open or save a method or data file, print or preview a report, or exit the Cary Eclipse application.

Hot Tip
Highlight a method or data file in Explorer and simply drag and drop it onto the application to open it. Any data will be displayed in the Graphics area.

The following commands can be accessed from the File menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>New</td>
<td>Select New to clear all method, report and graphics information and create a new file containing the default settings.</td>
</tr>
<tr>
<td>Open Method…</td>
<td>Select Open Method to display the Open dialog where you can search for and load a stored method.</td>
</tr>
<tr>
<td>Open…</td>
<td>Select Open to display the Open dialog where you can search for and load method, data, report, graph template and batch files, as well as other file types compatible with the Cary Eclipse software. When you load a batch file, the method which collected the data is loaded as well as the associated report and graphics layout.</td>
</tr>
<tr>
<td>Save Method As…</td>
<td>Select this option to display the Save As dialog and store the current method. You can use the Save As dialog to store the method, data, report, or graphics template separately or together as a Batch file. The data can also be saved as a comma delimited ASCII Spreadsheet file.</td>
</tr>
<tr>
<td>Save As…</td>
<td>Select this option to display the Save As dialog and store the collected data. You can use the Save As dialog to store the method, data, report, or graphics template separately or together as a Batch file. The data can also be saved as a comma delimited ASCII file.</td>
</tr>
<tr>
<td>Print Setup…</td>
<td>Select this option to display the Windows Print Setup dialog where you can select the printer, printer page size and orientation.</td>
</tr>
<tr>
<td>Print</td>
<td>Select this option to display the Print Preview dialog where you can view the current report</td>
</tr>
</tbody>
</table>
Use the dialog to open a file previously saved by the application. To access the Open dialog, select Open or Open Method from the File menu.

**Look in**

The Look In field lists the available folders and files. Click the down arrow on the right of this field to view the location of the current folder in your computer's hierarchy. You can also select a new location simply by selecting a different folder. The folders and files in the selected folder are displayed in the list box below the Look In field.

**Note**
To open the folder in the level that is one higher than the current folder, select on the toolbar.

**Look in list box**

The Look In List Box, which is directly below the Look In field, displays the folders and files in the selected location. To view the contents of a folder, double-click it.

- Click to create a new folder. Type the name of the new folder in the highlighted text entry field that appears and press Enter. Then double-click on the new folder to open it.

- Click to display the directory structure of the drive.

- Click to display the files as a list.

- Click to display files as a detailed list including the file size, file type and the date the file was last modified.

- Click to preview the selected trace before you load it into the application.

**File name**

Enter the name of the file you want to open in this field. The easiest way to do this is to find the file in the Look in list box and then select the file. However, you can type the full path of a file and then select Open to open the file.

**Files of type**

Click on the arrow on the right of this field to view a list of the different types of application files that can be opened in the list box.
Save As—Kinetics

Choose this file type to save the data, method, report or graphics template individually or together as a Batch file.

Overlay Data
Select this check box to overlay individual data files in one graph box. If this is not checked the data from each file will appear in individual graph boxes. This option is only available if the Open dialog has been accessed by selecting 'Open...' from the File menu.

Hot Tip
Use the SHIFT or Control key to select multiple data files.

Open
This button opens the highlighted file/s or the files listed in the File name field.

See also:
File menu
ASCII Spreadsheet File

Save As—Kinetics

Look in | File name | Files of type | Save

Use the Save As dialog to save the data, method, report or graphics template individually or together as a Batch file.

Note
This dialog appears when you select either the Save Method As or Save As menu option in the File menu. When you select Save Method As this dialog appears with 'Methods' displayed in the Files of type field. When you select Save As this dialog appears with 'Batch' displayed in the Files of type field.
Traces (or data) that are currently ‘Visible’ in the Graphics area are saved, regardless of how they are displayed (full screen graph box or tile).

For example, if six graph boxes are displayed in the Graphics area and each graph box has one Visible trace then all six traces will be saved.

If you do not wish to save particular traces, make them not Visible in the Trace Preferences dialog. To save only the focused trace, select the Save only focused trace check box.

The Save As dialog comprises the following:

**Look In**
- The Look In field lists the available folders and files. Click the down arrow on the right of this field to view the location of the current folder in your computer's hierarchy. You can also select a new location simply by selecting a different folder.
- The folders and files in the selected folder are displayed in the list box below the Look In field.

**Note**
- To open the folder in the level that is one higher than the current folder, select on the toolbar.

**Look in list box**
- The Look In list box, which is directly below the Look In field, displays the folders and files in the selected location. To view the contents of a folder, double-click it.
- Click to create a new folder. Type the name of the new folder in the highlighted text entry field that appears and press ENTER. Then double-click on the new folder to open it.
- Click to display the directory structure of the drive.
- Click to display the files as a list.
- Click to display files as a detailed list including the file size, file type and the date the file was last modified.
- Click to preview the selected trace before you load it into the application.

**File Name**
- Enter the name of the file you want to save in this field.
- You can also type the full path of a file then select the Save button to save the file.

**Files of Type**
- Click on the arrow on the right of this field to view a list of the different types of application files that can be opened in the list box.
  - **Methods (*.FMKN)** Choose this file type to only save the current method. Any collected data is not stored.
  - **Data (*.FDKN)** Choose this file type to only save the data currently Visible in the Trace Preferences dialog (for any graph box). Note that the method used to collect the data is not stored with the data.
  - **Report (*.FRKN)** Choose this file type to only save the contents currently shown in the Report area.
  - **Graph Template (*.FGKN)** Choose this file type to save the layout of the Graphics area as a template. For example, any bitmaps you have added to your Graphics area will be saved as well as the range and style of the graph axes.
  - **Batch (*.FBKN)** Choose this file type to save the data that is currently Visible in the
Filename extensions—Kinetics

The Cary Eclipse system uses the filename extension to distinguish between data types (e.g. method or data) and also to differentiate applications. (e.g. Concentration or Kinetics).

The Syntax for file extensions is:

<table>
<thead>
<tr>
<th>Letter</th>
<th>Represents</th>
<th>Available Options</th>
</tr>
</thead>
<tbody>
<tr>
<td>First</td>
<td>Type of analysis</td>
<td>F Fluorescence</td>
</tr>
<tr>
<td>Second</td>
<td>Type of file</td>
<td>B Batch</td>
</tr>
<tr>
<td></td>
<td></td>
<td>D Data</td>
</tr>
<tr>
<td></td>
<td></td>
<td>G Graph Template</td>
</tr>
<tr>
<td></td>
<td></td>
<td>M Method</td>
</tr>
<tr>
<td></td>
<td></td>
<td>R Report</td>
</tr>
<tr>
<td>Third and fourth</td>
<td>Application identifier</td>
<td>AB Advanced Reads</td>
</tr>
<tr>
<td></td>
<td></td>
<td>CN Concentration</td>
</tr>
<tr>
<td></td>
<td></td>
<td>KN Kinetics</td>
</tr>
<tr>
<td></td>
<td></td>
<td>LT Lifetimes</td>
</tr>
<tr>
<td></td>
<td></td>
<td>RT Ratio</td>
</tr>
<tr>
<td></td>
<td></td>
<td>SR Simple Reads</td>
</tr>
</tbody>
</table>

**Spreadsheet Ascii (*.CSV)**

Choose this file type to save the raw intensity data currently Visible in the Trace Preferences dialog (for any graph box) as a comma delimited ASCII Spreadsheet file (*.CSV) file. The data is saved as XY pairs delimited by a comma.

*Note*

This file type can be easily imported into a spreadsheet program. For example, Microsoft Excel. You can also double-click on the stored *.CSV file from Microsoft Explorer to load Excel automatically with the ASCII XY data displayed.

**GRAMS (*.SPC)**

Choose this file type to only save the data currently Visible in the Trace Preferences dialog (for any graph box) as a Cary Eclipse GRAMS data file.

**Tell me more about Filename Extensions**

Save only focused trace

Select this option to save only the focused trace in the graph box. If this option is not selected, all visible traces in the graph box will be saved.

Save

Click this button to save the file currently present in the File name field in the directory specified in the Look in field.

See also:

File menu

ASCII Spreadsheet file (*.CSV)

**Filename extensions—Kinetics**

The Cary Eclipse system uses the filename extension to distinguish between data types (e.g. method or data) and also to differentiate applications. (e.g. Concentration or Kinetics).

The Syntax for file extensions is:
The following files may be opened and saved in the Kinetics Application:

<table>
<thead>
<tr>
<th>Extension</th>
<th>Available action</th>
<th>File type</th>
</tr>
</thead>
<tbody>
<tr>
<td>*.FMKN</td>
<td>Open/Save</td>
<td>A <em>method</em> file (*.FMKN) consists of the software setup by which data is collected.</td>
</tr>
<tr>
<td>*.FDKN</td>
<td>Open/Save</td>
<td>A <em>data</em> file (*.FDKN) contains data collected that is currently visible in the Trace Preferences dialog (for any graph box).</td>
</tr>
<tr>
<td>*.FRKN</td>
<td>Open/Save</td>
<td>A <em>report</em> file (*.FRKN) contains the report generated by the Cary Eclipse about the data.</td>
</tr>
<tr>
<td>*.FGKN</td>
<td>Open/Save</td>
<td>A <em>graph template</em> file (*.FGKN) contains information that sets up the layout of the Graphics area including any bitmaps as well as the range and style of the graph axes.</td>
</tr>
<tr>
<td>*.FBKN</td>
<td>Open/Save</td>
<td>A <em>batch file</em> (*.FBKN) contains the collected data that was Visible in the Trace Preferences dialog (for any graph box), the method by which the data was collected and the report and graphics template.</td>
</tr>
<tr>
<td>*.CSV</td>
<td>Open/Save</td>
<td>An <em>ASCII spreadsheet</em> file (*.FCSV) has the same extension for all applications. It consists of raw intensity data (that was currently Visible in the Trace Preferences dialog (for any graph box) saved as XY comma-delimited pairs. This file can be imported into a spreadsheet program such as Microsoft Excel.</td>
</tr>
<tr>
<td>Data.*</td>
<td>Open</td>
<td>A <em>Cary DOS</em> (Data.*) file is the equivalent of a Cary Windows Data file—it does not contain any Batch information.</td>
</tr>
<tr>
<td>*.ADL</td>
<td>Open only</td>
<td>An <em>ADL</em> file (*.ADL) is a program file generated in the Applications Development Language (ADL).</td>
</tr>
<tr>
<td>*.SPC</td>
<td>Open/Save</td>
<td>A <em>Cary GRAMS</em> file (*.SPC ) is the equivalent of a Cary Eclipse Windows Data file—it does not contain any Batch information.</td>
</tr>
<tr>
<td>*.FADL</td>
<td>Open</td>
<td>An <em>Eclipse ADL</em> (*.FADL) file is a program file generated in the Applications Development Language (ADL) specifically for the Cary Eclipse. Eclipse ADL files will be associated with the relevant Eclipse application. When creating ADL files for the Cary Eclipse, it is recommended that you save them with the FADL extension.</td>
</tr>
</tbody>
</table>

**Note**
Selecting *All Files* in the Windows Open dialog will list all available file types.

See also:
- Open
- Save As
ASCII Spreadsheet file (*.CSV)—Kinetics

An ASCII Spreadsheet file consists of raw intensity data saved as XY comma-delimited pairs. You can choose to import or export Cary Eclipse files to ASCII files, which can then be imported in spreadsheet programs such as Microsoft Excel.

The format is as follows:

- Line 1 contains the sample name plus a comma
- Line 2 contains X and Y labels. The name of the X label must be the same as that used to label the graph. For example, in Kinetics, the graph label is Time (secs). The Y axis must be labeled as Intensity (a.u.).
- Lines 3 and onward contain the X,Y data
- After the last line of X,Y pairs there must be a blank line separating the data from any text audit strings or the end of the file.

For example, if a Kinetics file was collected as:

\[
\begin{align*}
  s &= 0.025, \text{ Intensity} = 471.607 \\
  s &= 5.025, \text{ Intensity} = 462.810 \\
  s &= 10.025, \text{ Intensity} = 438.042 \\
\end{align*}
\]

etc.

It would appear as follows in the ASCII file:

```
Kinetics,
Time (secs); Intensity (a.u.)
0.025, 471.607,
5.025, 462.810,
10.025, 438.042,

Kinetics,
Collection time;
```

Microsoft’s Excel generates a CSV file with the above format.

There is also an alternative format for CSV files. For example,

```
Kinetics
Time (secs); Intensity (a.u.)
0.025, 471.607,
5.025, 462.810,
10.025, 438.042,

Kinetics,
Collection time;
```

**Note**

The Time units for Kinetics can be either (mins) or (secs).
How to combine data files into a batch file—Kinetics

To combine a number of data files into a batch file, do the following:

Step 1
Select **Open**… from the **File** menu to display the Windows **Open** dialog.

Step 2
A list of stored batch files will appear. Select the down arrow to the right of the **Files of type** field and select 'Data' to list all the data files.

Step 3
Select the **Overlay Data** check box.

Step 4
Highlight the data files that you require. Hold down the CTRL key to select multiple files.

Step 5
Select **Open**. The highlighted data files will load into the application and appear in the same graph box.

Step 6
Select **Save As**… from the **File** menu to display the Windows **Save As** dialog.

Step 7
A list of stored data files will appear. Select the down arrow at the right of the **Files of type** field and select 'Batch' to list all the batch files. Ensure that the **Save only focused trace** check box is cleared.

Step 8
In the **File name** field, type in the file name for your new batch file.

Step 9
Select **Save** to create the new batch file. The current method will be stored with the batch file.

All the data files are now combined into the one batch file.
How to export collected data—Kinetics

If you want to export data from a Cary Eclipse application into a third-party application, then you simply need to save your data as an Spreadsheet ASCII (*.CSV) file or a Rich Text Format (*.RTF) file. Spreadsheet ASCII files are recommended if you are planning to use the data in a spreadsheet program such as Microsoft® Excel. Rich Text Format files are recommended if you are planning to open the data in a word processor program such as Microsoft® Word.

To export collected data:

Step 1

From the File menu, select Save As. The Save As dialog is displayed.

Step 2

Click on the down arrow next to the Files of type list box and select the Spreadsheet ASCII (*.CSV) or Rich Text Format (*.RTF) option.

Step 4

Type the name of your new file in the File name field.

Step 5

Select Save to export your data to this new file.

Software and Screen features—Kinetics

- Menu line
- Status display
- Status line
- Buttons
- Function keys
- Right mouse menu
- How to remove or display the command buttons

Menu line—Kinetics

- File menu
- Edit menu
- View menu
- Commands menu
- Setup menu
- Graph menu
- Help menu

Status Line—Kinetics

The Status Line is displayed at the very bottom of the Kinetics Application window. The line will display the following information (from left to right):
The major commands in this application can be accessed both from the menu items at the top of the main window and from the application buttons. These buttons, which appear in the left side of the Kinetics Application window, comprise the following:

**Hardware Status**
This field displays status messages indicating the current instrument activity.

**Sequence Information**
This field displays status messages indicating the current system activity.

**Current Results**
This field displays information on the last reading.

### Kinetics Application window

#### Buttons—Kinetics

The major commands in this application can be accessed both from the menu items at the top of the main window and from the application buttons. These buttons, which appear in the left side of the Kinetics Application window, comprise the following:

**Hot Tip**
If you wish to access these commands from the main menus, rather than from these buttons, you can remove the Command buttons from your application window. To do this, clear the **Buttons** command in the **View** menu. This will enlarge the Graphics area, making viewing and selection of scans easier.

<table>
<thead>
<tr>
<th>Button</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Setup</strong></td>
<td>Click this button to display the <strong>Setup</strong> dialog where you can set up your Cary Eclipse for the collection, storage and analysis of the kinetics run. You can also set up any accessories that you may have and define the reporting options.</td>
</tr>
<tr>
<td><strong>Zero</strong></td>
<td>Select this to zero the current ordinate value. This command can also be accessed by pressing F5. <strong>Note</strong> Before you select <strong>Zero</strong>, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.</td>
</tr>
<tr>
<td><strong>Pre-scan</strong></td>
<td>Select <strong>Pre-scan</strong> to quickly determine the optimum instrument parameters to be used for your sample. At the end of this collection sequence the software will report the optimum Ex and Em wavelengths and PMT voltage. Once you are satisfied with the parameters, the software will automatically update your method setup to reflect these new parameters.</td>
</tr>
<tr>
<td><strong>Set Temp</strong></td>
<td>Click <strong>Set temp</strong> to open the <strong>Set Temperature</strong> dialog where you can change the temperature during a kinetics run. <strong>Q. Why is the <strong>Set Temp</strong> button unavailable?</strong> <strong>A.</strong> The <strong>Set Temp</strong> button is only accessible if you are using a thermostatted accessory. (On the <strong>Accessories</strong> page, the <strong>Temperature control</strong> option must be selected.) Once you have started a run, you can access the <strong>Set Temp</strong> button by selecting the <strong>Pause</strong> button to pause the run. You can then select the <strong>Set Temp</strong> button.</td>
</tr>
<tr>
<td><strong>Stop Time</strong></td>
<td>Click the <strong>Stop Time</strong> button to access the <strong>Stop Time</strong> dialog. Use this dialog to extend the length of a kinetics run while the run is in progress. Extending a run may be necessary if you decide whilst a run is in progress that the data collection period too short.</td>
</tr>
<tr>
<td><strong>Clear Report</strong></td>
<td>Select <strong>Clear Report</strong> to remove all of the text that appears in the Report area. If you have saved the report, you can view the information again. To do this, simply open the report file using the <strong>Open…</strong> command in the <strong>File</strong> menu.</td>
</tr>
<tr>
<td><strong>Recalculate</strong></td>
<td>Use the pages accessible from the <strong>Recalculate</strong> option to perform re-calculation on data that has been analyzed.</td>
</tr>
<tr>
<td><strong>Print</strong></td>
<td>Select <strong>Print</strong> to display the Windows <strong>Print</strong> dialog where you can choose what printer the report will be printed to. You can also select what pages to print as well as the number of...</td>
</tr>
</tbody>
</table>
Function keys—Kinetics

The function keys at the top of your keyboard perform basic Cary Eclipse operations simply and quickly.

<table>
<thead>
<tr>
<th>Function key</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>This displays the Help for the current dialog or window.</td>
</tr>
<tr>
<td>F4</td>
<td>Press F4 to display the Go to Wavelength dialog.</td>
</tr>
<tr>
<td>F5</td>
<td>Press F5 to perform an instrument Zero. The current intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before selecting Start if you want your data to start at 0 intensity. This command is also accessible from the Commands menu or by selecting the Zero button.</td>
</tr>
<tr>
<td></td>
<td>Note: Before you select Zero, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.</td>
</tr>
<tr>
<td></td>
<td>Select the Clear Zero command from the Commands menu to remove zero correction.</td>
</tr>
<tr>
<td>F6</td>
<td>Press F6 to display the Rapid Result dialog where you can read a blank and multiple sample readings without changing the application method. This command is also accessible from the Commands menu.</td>
</tr>
<tr>
<td>F9</td>
<td>If the application is online, press F9 to start a data collection. This is equivalent to selecting the Start button. If the application is not online, you will need to press F9 to first connect to the Cary Eclipse. Then press F9 to start the data collection. This command is also accessible from the Commands menu or by selecting the Start button.</td>
</tr>
<tr>
<td></td>
<td>Note: You will not be able to perform a Connect if another application is collecting data.</td>
</tr>
<tr>
<td>F12</td>
<td>Select this to stop the current measurement. This is equivalent to selecting the Stop button. This command is also accessible from the Commands menu or by selecting the Stop button.</td>
</tr>
</tbody>
</table>

See also:
- Go to Wavelength
- Zero
- Rapid Result
- Start/Stop Instrument Buttons
Right mouse menu—Kinetics

The following commands are accessible by clicking the right mouse button on editable fields in the Cary Eclipse software:

- **Undo** If available, this reverts to the previous value before the Edit on the field.
- **Cut** If available, this cuts the highlighted text to the clipboard.
- **Copy** If available, this copies the highlighted text to the clipboard.
- **Paste** If available, this pastes the text from the clipboard to the cursor position.
- **Delete** If available, this deletes the highlighted text/number.
- **Select All** If available, this selects all entries.

How to remove or display the command buttons—Kinetics

The command buttons (Setup, Zero, Pre-scan, Set Temp, Stop Time, Clear Report, Recalculate and Print) appear on the left side of the Kinetics Application window.

To remove the command buttons

Step 1

From the Kinetics Application window, select the View menu.

Step 2

Clear the Buttons menu item.

The buttons will disappear from the application screen, giving you more room to display reports.

To show the command buttons

Step 1

From the Kinetics Application window, select the View menu.

Step 2

Select the Buttons menu item.

The buttons will re-appear in the application screen.

ADL—Kinetics

- [ADL Command Line](#)
- [ADL Hooks](#)
- [How to insert ADL commands](#)

ADL Command Line—Kinetics

Introduction to ADL
Use the ADL Command Line to run Single-line or Multi-line Applications Development Language (ADL) commands that enable you to perform functions over and above that of the existing program functionality.

ADL is not a general purpose programming language. It has been developed specifically by Varian to meet the needs of spectroscopists. It can be used to handle all the data collected from the Cary Eclipse—both photometric values and entire data collections. ADL programs that you develop should be stored with the extension 'ADL'.

To access the ADL Command Line, select the ADL Command item from the Commands menu. You can also press ALT+Y to open the dialog.

**Note**
Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.

---

**Software controls**

**Single Line**
Selecting this lets you enter ADL commands on a single line into the Command Line entry box.

Q. Can I enter more than one ADL command on a single line?  
A. Yes, provided that the commands are separated by a semicolon ‘;’.

**Multi-line**
The Multi-line capability enlarges the Command Line entry box. Use the Multi-line capability to create a more complex ADL program by adding a series of commands over multiple lines. When Multi-line mode is selected, the Quick Reference option becomes available.

**Quick Reference**
Select Quick Reference to open the Quick Reference dialog where you can easily add a command in the correct syntax to your ADL program. This option is only available for Multi-line commands.

Show me How to insert ADL commands using the Quick Reference

A detailed description of all the commands in the Quick Reference is available by pressing the Help button on the Quick Reference dialog.

**Browse**
Select Browse to access the Open dialog where you can browse for and select an existing ADL program to load and run.

**Command Line (entry box)**
This entry box enables you to use ADL commands. You can enter single ADL commands into this field, or you can enter a series of ADL commands. If you have the entry type set to Single Line, you can review previous ADL entries by selecting the down arrow to the right of the list box.

The ADL commands in the Command Line are executed when you select OK.

**Note**
On entering this window, the Cary Eclipse will recall the last command or program executed from the Command Line entry box. The ADL commands entered in the last executed program are typically stored in the Varian\Cary Eclipse WinFLR\ADL directory on the drive on which the Cary Eclipse software is installed.
**ADL Hooks—Kinetics**

The **ADL Hooks** command is accessible from the **View** menu.

Instead of writing a full ADL program you can use ADL hooks to link into an existing application. This is particularly useful if you want to make small changes to the way an application works, e.g. you want to pop up a dialog for the user to enter some specific data before the run starts. ADL hooks are links at specific points in an application at which you can run an ADL program. For example, ADL hooks have been supplied to run the following ADL programs:

<table>
<thead>
<tr>
<th>Hook</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>KN_Startup.ADL</td>
<td>This is run when the Kinetics application starts. For this to to work the application must be started from a batch or method file that has ADL Hooks on.</td>
</tr>
<tr>
<td>KN_PreSeq.ADL</td>
<td>This runs when the <strong>Start</strong> button is selected.</td>
</tr>
<tr>
<td>KN_PostSeq.ADL</td>
<td>This runs at the end of the <strong>Start</strong> button sequence.</td>
</tr>
<tr>
<td>KN_Online.ADL</td>
<td>This runs when Cary Eclipse is detected online.</td>
</tr>
<tr>
<td>KN_OffLine.ADL</td>
<td>This runs when Cary Eclipse goes offline.</td>
</tr>
<tr>
<td>KN_Zero.ADL</td>
<td>This runs when the Zero button is selected.</td>
</tr>
<tr>
<td>KN_Recalc.ADL</td>
<td>This runs when the <strong>Recalculate</strong> button is selected.</td>
</tr>
<tr>
<td>KN_Shutdown.ADL</td>
<td>This runs when the application is shutting down.</td>
</tr>
<tr>
<td>KN_PreRead.ADL</td>
<td>This runs just before a read is taken.</td>
</tr>
<tr>
<td>KN_PostRead.ADL</td>
<td>This runs just after a read is taken (It has the reading result stored in the <strong>UserResult</strong> variable.)</td>
</tr>
<tr>
<td>KN_PreSetup.ADL</td>
<td>This runs when the <strong>Setup</strong> button is selected.</td>
</tr>
<tr>
<td>KN_PostSetup.ADL</td>
<td>This runs when the <strong>OK</strong> button on the <strong>Setup</strong> dialog is selected.</td>
</tr>
<tr>
<td>KN_CellChange.ADL</td>
<td>This runs instead of the Cary Eclipse driving the Multicell Holder. It enables you to add your own instructions for the Cary Eclipse Multicell Holder or to use your own cell changing accessory.</td>
</tr>
<tr>
<td>KN_PreStage.ADL</td>
<td>This runs prior to each collection stage. It determines the stage by checking the <strong>UserResult</strong> variable as UserResult = stage number.</td>
</tr>
</tbody>
</table>

**Note**
Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.
How to insert ADL commands using the Quick Reference—Kinetics

The Quick Reference window is accessed from the ADL Command window. It enables you to easily add a command in the correct syntax to your ADL program. To do this:

Step 1

Press Alt-Y to open the ADL Command Line window.

Step 2

Select the Multi-line option. The Command Line entry box will expand to take multiple lines of ADL commands and the Quick Reference check box is displayed.

Step 3

Select the Quick Reference check box. The Quick Reference window is displayed.

Step 4

If necessary, use the Go To field to jump to the appropriate part of the Quick Reference listing.

Step 5

Highlight the command you require in the Quick Reference list.

Step 6

Click the Insert in ADL button. The selected ADL command will appear in the Command Line entry box.

Step 7

Where necessary, alter the defaults supplied by the inserted Quick Reference command in the Command Line entry box to suit your program.
Step 8

Repeat steps 4–7 to add more commands as required.

See also:
ADL Command Line

Help and Safety information—Kinetics

- Help menu
- Safety home page

Help menu—Kinetics

Use the Help menu to view the online Help and software information for this application.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
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<tbody>
<tr>
<td>Kinetics Help</td>
<td>Select this to display the Kinetics Home Page. This contains a list of online Help topics specific to the Kinetics Application.</td>
</tr>
<tr>
<td>Help Topics</td>
<td>Select this to display the Cary Eclipse Windows Online Help Home Page.</td>
</tr>
<tr>
<td>Tip of the day</td>
<td>View useful tips about using your Cary Eclipse instrument and software.</td>
</tr>
<tr>
<td>Eclipse Tour</td>
<td>Select this to display the Cary Eclipse Multimedia Online Help. This will give you an animated introduction to the Cary Eclipse software and hardware.</td>
</tr>
<tr>
<td>About…</td>
<td>Select About to access a dialog that displays information about the application as well as Varian email and Internet contact information. To clear this About dialog, select OK.</td>
</tr>
</tbody>
</table>

Safety Home Page

Click on the desired index entry to view the Help.

- About Safety
- Lamp module
- Ultraviolet radiation
- Electrical hazards — Cary Eclipse
- Panels, covers and modules
- Other precautions
- Warnings and cautions
- Warning symbols
- Color coding
- Information symbols
- FCC advisory
- CE compliant products

Kinetics How To—Kinetics

Select the procedure you require below for a step-by-step informative guide on running the application software.
Procedural

- How to perform a single-wavelength kinetics run
- How to perform a multi-wavelength, temperature controlled kinetics run

Calculations and reporting

- How to use the kinetics ruler
- How to generate a report from previously collected data
- How to perform post run calculations on previously stored data
- How to use advanced reporting options

General

- How to email your results
- How to export collected data
- How to combine data files into a batch file
- How to remove or display the command buttons
- How to insert ADL commands using the Quick Reference

See also:
Kinetics Home Page
Graphics How To

How to resize graph boxes—Kinetics

Step 1
Click on the graph that you want to resize so that the highlight box appears around it.

Step 2
Move the cursor to the edge of the highlight box and place it over one of the square marks (e.g. □) in the box until it turns into a double headed arrow (←→).

Step 3
Click and hold the left mouse button and drag the box to the desired size.

Step 4
Release the left mouse button.

See also:
How to alter the look of your graphs

How to zoom a graph—Kinetics

To zoom a graph, do the following:

Step 1
Click on the button on the toolbar, or select Cursor Mode from the Graph menu. The Cursor Modes
dialog will appear.

Step 2

Select **Free** and select the **OK** button.

Step 3

Click and hold down left mouse button in the highlighted graph box.

Step 4

Drag the mouse sideways and up or down. You will see a zoom box made up of a dotted line appear on the screen. (See picture below.)

![The dotted line is a zoom box.](image)

**Hot Tip**

To move the box on the screen, hold down the left and right mouse buttons together. You can then position the box anywhere in the graph box. Releasing the right mouse button will still allow you to resize the zoom box.

Step 5

Release the left mouse button. The area defined by the zoom box will be redrawn to fill the entire graph box, and the scans within the box will be enlarged accordingly. The abscissa (X) and ordinate (Y) coordinates displayed below the graph will also be changed to reflect the positions of the corners of the zoom box.

You can repeat this process if you need to zoom in even further.

**Note**

The system may limit the extent to which you can zoom.

See also:

- [How to scale your graph manually](#)
- [How to scale your graph automatically](#)

**How to take the derivative of a trace**

Some uses of derivatives are:

- To resolve partly overlapping peaks
- To quantitatively determine trace compounds
- To characterize pure compounds
- Purity testing

To take the derivative of a collected trace and plot the result, follow these steps:

Step 1
Click on the trace to be smoothed. It will become red (highlighted).

Step 2

Click the Selected Trace or Focused Trace button. The name of the highlighted trace will appear in the calculation display field.

Step 3

In the Trace group, select your derivative order in the Operation field. Choose from derivatives 1 to 4 (Deriv1, Deriv2, Deriv3, Deriv4). You may have to scroll to find the derivative you require.

Step 4

Enter a Filter Size and an Interval in the corresponding fields.

Step 5

In the Display Options group, select New Graph to plot the result in a new graph box, or Selected Graph to plot the result in the current graph box.

Step 6

Click the Apply button in the Trace group. Your Deriv selection will be displayed before the trace name in the calculation display field.

Step 7

Click on the equals (=) button. The result will be displayed in the Graphics area.

See also:

About derivatives

**How to smooth a trace**

To smooth a collected trace and plot the result, follow these steps:

Step 1

Click on the trace to be smoothed. It will become red (highlighted).

Step 2

Click on the Maths button in the toolbar (it's on the far right) or select the Maths menu.

Step 3

Click the Selected Trace or Focused Trace button. The name of the highlighted trace will appear in the calculation display field.

Step 4

In the Trace group, select Smooth in the Operation field. (You may have to scroll to find it.)
Step 5

Enter a Filter Size and an Interval in the corresponding fields.

Step 6

In the Display Options group select New Graph to plot the result in a new graph box, or Selected Graph to plot the result in the current graph box.

Step 7

Click the Apply button in the Trace group. The word ‘Smooth’ will appear before the trace name in the calculation display field.

Step 8

Click on the equals (=) button. The result will be displayed in the Graphics area.
**Lifetimes Home Page**

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<td>• How to use advanced reporting options</td>
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<tr>
<td></td>
<td></td>
<td>data</td>
</tr>
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</table>

**To find out about…**

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<td></td>
<td>• Report Toolbar</td>
</tr>
<tr>
<td></td>
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</tr>
<tr>
<td>ADL</td>
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<tr>
<td>Help and safety information</td>
<td>• Help menu</td>
</tr>
<tr>
<td></td>
<td>• Safety Home Page</td>
</tr>
</tbody>
</table>

**Lifetimes Application**

**Introduction**

The Lifetimes application is used to measure phosphorescence lifetimes, and therefore is only available in phosphorescence mode.

The Lifetimes application measures the average amount of time a molecule remains in the excited state following excitation. Where:

Lifetimes (time resolved)

\[ A + \text{Excitation} \rightarrow A^* \]
A* <- A + Emission

It is a time-based measurement (like Kinetics) where the change in intensity is usually due to uni-molecular processes.

The shape of the decay curve produced by a Lifetimes measurement is sensitive to the environment that the molecule is in. For this reason, it is often used as a probe technique to determine what the molecule is experiencing.

To access the Lifetimes Application, click the Windows Start button, point to Programs, Cary Eclipse and then select Lifetimes.

Software controls

The application comprises the following:

<table>
<thead>
<tr>
<th>Instrument Status Readings</th>
<th>The Instrument Status Readings, which display the Y and X values of the graph axes are displayed in the top left and right corners of the Lifetimes Application Window.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Y value</td>
<td>Displays the current Y value, usually intensity. This value is continually updated during a data collection.</td>
</tr>
<tr>
<td>X value</td>
<td>Displays the current X value, which is the current reading for the Time. On the Cary page of the Setup dialog, choose whether to display the time value in minutes or seconds.</td>
</tr>
<tr>
<td>Start and Stop buttons</td>
<td>The Start and Stop buttons at the top of the Lifetimes Application window are used to start and stop instrument analyses. These buttons can also be accessed from the Commands menu.</td>
</tr>
<tr>
<td></td>
<td><strong>Tell me more about the Start and Stop buttons</strong></td>
</tr>
<tr>
<td>Graphics area</td>
<td>The Graphics area refers to the plotting area of the application. It can contain traces of collected data, labels and pictures. In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area defined by the axes is referred to as a graph box.</td>
</tr>
<tr>
<td></td>
<td>To display or hide the Graphics area, select the Graphics command on the View menu. Double-clicking on the Graphics area enables you to toggle between half or full size display sizes.</td>
</tr>
<tr>
<td></td>
<td><strong>Tell me more about the Graphics area</strong></td>
</tr>
<tr>
<td>Report area</td>
<td>Use the Lifetimes Report area to view the current report. The report may include the results of your data collection, the method parameters used to collect the results and any graphics you have included. You can select the information to include in your report on the Reports page of the Setup dialog.</td>
</tr>
<tr>
<td></td>
<td>To display or hide the Report area, select the Report command from the View menu. Double-clicking on the Report area enables you to toggle between half or full size display sizes.</td>
</tr>
</tbody>
</table>
You can copy the generated report onto the Windows clipboard. To do this, highlight the report (or a relevant section of it) and press CTRL+C. You can then paste the report into a word processor such as Microsoft® Word.

**Hot Tip**
You can edit the Lifetimes Report by pressing CTRL+R or selecting the **Edit Report** command from the **Edit** menu.

**Commands**
The Lifetimes Application window contains the following commands, all of which can also be accessed from the various menus in the application.

*Click on the buttons to view information about them:*

- Setup...
- Zero
- Pre-scan...
- Clear report
- Recalculate
- Print...

**Hot Tip**
By becoming familiar with these commands (**Setup**, **Zero**, **Pre-scan**, etc.), you can remove these buttons from the Lifetimes window. This will enlarge the Report area and make it easier to view results. To do this, clear the **Buttons** item in the **View** menu.

**Menus**
The Lifetimes Application contains the following menus:

- **File**
- **Edit**
- **View**
- **Commands**
- **Setup**
- **Graph**
- **Help**

See also:
- Status Display
- Status Line
- Keyboard function keys
- Lifetimes Home Page

**Validation of your Cary Eclipse for the Lifetimes application**

The files listed below are shipped with the Lifetimes application to enable you to validate the software. The files are stored on your CD in a Validation subdirectory.

To validate your software, you should Open each file in turn and select the Recalculate button. The results calculated by the Cary Eclipse should match those given in the validation files listed below. These results were determined by exporting the data to a spreadsheet program and performing the same calculation as that used in the Lifetimes application.

- **Lifetimes validate_single exp.FBLT**
- **Lifetimes validate_double exp.FBLT**

**Getting ready for a run—Lifetimes**

- **Setup dialog**
- **Setup menu**
- **Cary page**
- **About Gate Time**
- **How many flashes?**
- **Options page**
Use the Setup dialog to set up your instrument and accessories for a Lifetimes run. You can also use the dialog to set up the Analysis, Report and Storage parameters associated with the run.

To access the Setup dialog select the Setup button from the Lifetimes Application window or the Setup command from the Setup menu.

The dialog comprises the following tabbed pages:

- **Cary**
- **Options**
- **Accessories**
- **Analyze**
- **Reports**
- **Auto Store**

See also: Lifetimes Application

### Setup—Lifetimes

Use the Setup menu to access the parameters for setting up a Lifetimes read.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Setup</strong></td>
<td>Choose Setup to display the Setup dialog where you can set up your instrument for a Kinetics measurement. The Setup dialog can also be accessed from the Setup command button.</td>
</tr>
<tr>
<td><strong>Cary</strong></td>
<td>Choose Cary to display the Cary page. You can use this page to set up the instrument parameters associated with the data collection.</td>
</tr>
<tr>
<td><strong>Options</strong></td>
<td>Choose Options to display the Options page where you can set up your filters and set the detector voltage. You can also use this page to select the way the collected data is displayed in the Graphics area, and the type of smoothing applied (if any).</td>
</tr>
<tr>
<td><strong>Accessories</strong></td>
<td>Choose Accessories to display the Accessories page. You can use this page to set up the accessory parameters associated with the data collection.</td>
</tr>
<tr>
<td><strong>Analyze</strong></td>
<td>Choose Analyze to display the Analyze page where you can select from various mathematical equations and set the parameters used to generate the calculation of reaction rates.</td>
</tr>
</tbody>
</table>
Use the **Cary** page to set up the instrument parameters associated with the data collection.

From the **Setup** menu, select **Cary** to access the **Cary** page. You can also access the page from the **Setup** dialog by selecting the **Cary** tab. To access the **Setup** dialog, select the **Setup** button from the Lifetimes Application window.

**Note**
The Lifetimes Application operates in **Phosphorescence** mode only.

### Instrument setup

**Delay time**
- Enter, in milliseconds, the delay time for your sample. The delay time is the time that elapses between the last flash and the beginning of data collection. The default value is 0.100 ms, which is the time it takes for the lamp intensity to decay to zero. Setting a delay time ensures that there is no lamp effects in the phosphorescence decay curve.
  - The range is 0.000–5000.000 s.

**Gate time**
- Enter, in milliseconds, the gate time for your sample. The gate time is the overall reading time of the emission signal.
  - The range is from 10µs–10s.

**No. of flashes**
- Enter the number of lamp flashes required, which is how many times the lamp flashes consecutively before the delay time is counted.

### Wavelength setup

**Ex. Wavelength (nm)**
- Set the wavelength of the excitation monochromator, which is the wavelength value at which you want to excite your sample. Click the arrow to the right of the field to quickly select the most recent wavelength entries.

Alternatively, to set the excitation monochromator to zero order (white light), from the **Commands** menu, point to **Zero Order** and select **Excitation**. A tick next to this option
indicates that it is selected. Clear this option to return to monochromator light and use the value set in the **Ex. Wavelength** field.

**Em. Wavelength (nm)**

Set the wavelength of the emission monochromator, which is the wavelength value at which your sample emits. Click the arrow to the right of the field to quickly select the most recent wavelength entries.

Alternatively, to set the emission monochromator to zero order (white light), from the **Commands** menu, point to **Zero Order** and select **Emission**. A tick next to this option indicates that it is selected. Clear this option to return to monochromator light and use the value set in the **Em. Wavelength** field.

**Ex. Slit (nm)**

Set the slit width of the excitation monochromator. Choose from 1.5, 2.5, 5, 10 or 20 nm or 10 Round.

**Em. Slit (nm)**

Set the slit width of the emission monochromator. Choose from 1.5, 2.5, 5, 10 or 20 nm or 10 R.

**Collect timing**

**Total decay time**

Enter the time, in milliseconds, over which the decay will be measured. You should enter a time that will allow for the phosphorescence signal to decay to approximately zero. The lamp will not flash until the total decay time has elapsed.

**No. of cycles**

Enter the number of cycles (or decay curves) over which the run will be collected. This number will be used to average the data. Entering a number of cycles value enables you to obtain better signal to noise ratios as multiple traces can be averaged. Once more than two traces have been collected, the average is applied at the end of each decay curve (a cycle).

**X Mode**

This enables you to set the units of time for the abscissa (or X axis).

Choose your desired abscissa mode by selecting one of the following buttons:

- **ms**
  
  Select this to time the run in milliseconds.

- **Sec**
  
  Select this to time the run in seconds.

**Status Display**

Select this check box to display the **Status Display** window, where you can view the status of various parameters during the data collection.

See also:

- **Accessories**
- **Status Display**

**Understanding Gate Time and Average Time—Lifetimes**

For Phosphorescence scans, there is a relationship between Average Time (the time for which data is averaged) and Gate Time (the overall reading time of the emission signal).
Averaging is calculated as follows:

The intensity readings are taken from different decays, where:

**Number of Decays = Averaging Time/Gate Time**

**Note:**

**Gate Time** is not used if you select **Fluorescence** as the **Data Mode**.

**Average Time** is set on the **Cary** page of the **Setup** dialog. **Gate Time** is set on **Options** page, which is accessible from the **Cary** page.

The following diagram highlights the relationship between **Average Time** and **Gate Time**. In this example, the Average Time is set to five times the Gate Time:

<table>
<thead>
<tr>
<th>Average Time/Gate Time</th>
<th>5</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intensity</td>
<td>( \frac{A_1 + A_2 + A_3}{3} )</td>
</tr>
</tbody>
</table>
where:

- Total Decay Time: 25 ms
- Delay Time: 1 ms
- No. of cycles: 3
- Average Time: 5 ms
- Gate Time: 25 ms

See also:

Lifetimes Application
How many flashes should I perform?—Lifetimes

The **Number of Flashes** option is set in the **Instrument Setup** group on the **Cary** page.

**Example of the Instrument Setup Options**

![Diagram showing intensity and time for setting the number of flashes](image)

**Setting the number of flashes**

If you are measuring phosphorescence samples that have a relatively long decay time (that is, a number of seconds), you can setup the Cary Eclipse to pulse the lamp multiple times, as this will increase the intensity of the phosphorescent signal (refer to Example 1, below). For short lived phosphorescent samples, multiple flashes will not impact on the intensity of the signal, and therefore only one flash is required (refer to Example 2, below).

**Example 1**

The above example shows that the sample does not decay before the instrument lamp flashes again. Therefore setting multiple flashes will result in an increase in the intensity of the signal.
Example 2
The above example shows that the sample decays in less time than the time it takes the instrument to flash the lamp again. Therefore, setting more than 1 flash, in the No. of Flashes field will not increase the intensity of the signal.

Options—Lifetimes

Display Options | Excitation Filter | Emission Filter | PMT Detector Voltage | Status Display

Use the Options page to set up your filters and set the detector voltage. You can also use this page to select the way the collected data is displayed in the Graphics area, and the type of smoothing applied (if any).

From the Setup menu, select Options to access the Options page. You can also access the page from the Setup dialog by selecting the Options tab. To access the Setup dialog, select the Setup button from the Lifetimes Application window.

**Display Options**

**Overlay traces**
Select this option to superimpose the collected data of each sample in the run in one graph box. If you select this option, a graph box is created at the start of the collect in the Graphics area and each sample’s data is overlaid in that box as the data is collected. Traces will always be overlaid in the same graph, regardless of what multicell/well accessory is used. Clear this option to display all of the traces in individual graphs.

**Y minimum/Y maximum**
Enter the minimum and maximum Y values to be displayed on the graph axis. The software will update the Y axes to reflect these settings during the lifetimes run.

**Excitation filter**
Set the excitation filter for the Lifetimes measurement. To do this, select the down arrow to the right of the field and select an option from the list.

A range of filter settings are available:

**Auto**
If you select the Auto option, the filter wheel will be automatically moved to the appropriate position for the selected excitation wavelength. It is recommended that you set the filter to Auto (which is the default setting).

**Open**
If you select Open no filter is used.

**Closed**
If you select Closed, a blank filter is used to prevent light from getting through.
If you are looking at a wavelength range that is within one of these filter ranges, you can select your filter manually. Otherwise it is recommended that you set the filter to **Auto**.

### Emission filter

Set the emission filter for the Lifetimes measurement. To do this, select the down arrow to the right of the field and select an option from the list.

A range of filter settings are available:

- **Auto**
  
  If you select the **Auto** option, the filter wheel will be automatically moved to the appropriate position for the selected emission wavelength. It is recommended that you set the filter to **Auto** (which is the default setting).

- **Open**
  
  If you select **Open** no filter is used.

- **Closed**
  
  If you select **Closed**, a blank filter is used to prevent light from getting through.

If you are looking at a wavelength range that is within one of these filter ranges, you can select your filter manually. Otherwise it is recommended that you set the filter to **Auto**.

### 3%T Attenuator

Select **3%T Attenuator** to decrease the fluorescence intensity and thus the amount of light going to the emission PMT. This setting is useful for highly fluorescent samples when you are using wide slits.

### PMT Detector Voltage

Set the EHT or sample detector PMT. To do this, select one of the following:

- **Low**
  
  Sets the PMT Detector voltage to a pre-defined value of 400 v. Typically you would select this setting if you are reading highly emitting samples.

- **Medium**
  
  Sets the PMT Detector voltage to a pre-defined value of 600 v. Typically you would select this setting if you are reading moderately emitting samples.

- **High**
  
  Sets the PMT Detector voltage to a pre-defined value of 800 v. Typically you would select this setting if you are reading low emitting samples.

- **Manual**
  
  Enables you to enter any PMT voltage in the adjacent Volts field.

- **Volts**
  
  Enter the PMT voltage in the adjacent field. Note that this option is only available if you have selected Manual as the PMT Detector voltage.

### Tip

Increasing or decreasing the PMT Detector Voltage by 10% will double or halve the fluorescence intensity observed.
**Status Display**  Select this check box to display the **Status Display** window. This window allows you to view the status of various parameters during the data collection.

- **See also:**
  - **Setup**
  - **Status Display**

**Accessories—Lifetimes**

Use the **Accessories** page to set up the accessory parameters associated with the data collection.

From the **Setup** menu, select **Accessories** to access the **Accessories** page. You can also access the page from the **Setup** dialog by selecting the **Accessories** tab. To access the **Setup** dialog, select the **Setup** button from the Lifetimes Application window.

The page comprises the following:

**Multicell Holder**

Select this option to use the Multicell Holder with the Cary Eclipse.

- **Note**
  By selecting this accessory you will prohibit the use of some combinations of other accessories that are also controlled by the Accessory Controller Board.

  **Multi-zero**  If you select **Multi-zero**, a Multi-zero will be performed when the **Zero** button is selected in the Lifetimes Application window. A reading will be taken at the set **Averaging Time** at the specified wavelength(s) of one or both monochromators and at every selected cell position. These readings will be offset to zero and subsequent readings will have the corresponding offset value subtracted from each cell.

  **Reset**  Select **Reset** to drive the Multicell Holder to cell position 1, which is its fully retracted or zero position.

  **Go to cell…**  Select **Go to cell** to open the **Multi Cell Changer** dialog. You can use this dialog to manually move the Multicell Holder accessory to a new cell. To do this, simply select the corresponding button of the cell you require in the Multi Cell Changer dialog. The Multicell Holder will move the selected cell to the measurement position in the sample compartment.

  The **Multi Cell Changer** dialog is ‘modeless’ which means that it can remain open even once you have cleared the **Setup** dialog. This enables you to use the **Rapid Read** command to perform a reading at the cell specified in the Cell Changer.

  If you move to a new cell after you have performed a Multi-zero scan, the Cary Eclipse will display the intensity reading corrected by the determined zero-offset amount for that cell.

  - **Note**
    You can use this option to check the intensity of a cell or perform a lifetimes run at a particular cell.

**Temperature Control**

Select this option to specify the temperature parameters for your analysis.

- **Note**
The **Temperature Control** option does not control temperature accessories that are not listed in the **Device** field. For example, if you have selected the **Temperature Control** option and set the **Device** to **None** because you have a different temperature control accessory, the Cary Eclipse software will not control that accessory. However, it will monitor the temperature and prompt the software to start when the accessory reaches the temperature listed in the **Temperature** field.

**Device**

Select the device that will be used to control the water temperature.

The options available will depend upon whether or not you have selected the **Multicell holder** option.

If you *have* selected the Multicell holder, the following **Device** options will be available for selection:

- Temperature controller
- PCB 150 water bath
- None

If you *have not* selected the Multicell holder, the following **Device** options will be available for selection:

- Single cell peltier
- PCB 150 Water bath
- None

**None**

Select this option if you are using a Temperature Control device not listed in this field (for example, a third party water bath). Note that the Cary Eclipse software will not control the accessory. The software will simply monitor the temperature and prompt the software to start when the accessory reaches the temperature listed in the **Temperature** field.

**PCB 150 water bath**

Select this option if you are using the PCB 150 water bath.

**Temperature controller**

Select this option if you are using the Temperature controller.

**Temperature**

Set the temperature at which the analysis will be performed. The default value is 20.00° C.

When you select **OK** to close the **Setup** dialog, all buttons in the Advanced Reads Application window will be disabled until the temperature recorded by the Monitor device is within ±0.5°C of the temperature range.

If **Zero** or **Start** is selected, and the temperature is not within ±0.5°C of the temperature range, then the system will wait for the temperature to fall within the temperature range before continuing with the measurement. Whilst waiting for the temperature, there will be a display on the status line at the bottom of the Advanced Reads Application window giving the current temperature and the elapsed time waiting to reach the set temperature.

**Port**

Select from COM1, COM2, COM3 or COM4.

This option is only available if you have selected **PCB 150 Water bath** as the **Device**.

**Monitor**

Select which device will be used to monitor the reaction temperature. The options in this field will change depending on the **Device** selected.

<table>
<thead>
<tr>
<th>Device selected</th>
<th>Monitor options available</th>
</tr>
</thead>
<tbody>
<tr>
<td>None</td>
<td>Probe 1, Probe 2, Probe 3, Probe 4</td>
</tr>
<tr>
<td>PCB 150 water bath</td>
<td>PCB 150, Probe 1, Probe 2, Probe 3, Probe 4</td>
</tr>
<tr>
<td>Temperature controller</td>
<td>Block, Probe 1, Probe 2, Probe 3, Probe 4</td>
</tr>
</tbody>
</table>

**Temperature Display**

Select this option to view Temperature Display information in the **Status Display** window.
The **Temperature Display** option is only available if you have selected the **Temperature Control** option.

**Block/PCB 150**

Select this option to view the temperature of the Block or the PCB 150 in the Status Display window. This option is not available if you have selected **None** as the **Device** option.

Note that the PCB 150 option is only displayed if you have selected the **PCB 150 Water Bath** as the **Device** option without the **Multicell holder**.

**Probes**

Select one or more of these options to view the temperature of the Probes in the Status Display window.

**Status Display**

Select this check box to display the **Status Display** window. Use this window to view the status of various parameters during the data collection.

See also:

- Lifetimes Application
- Multi Cell Changer
- Cary page
- Status Display window

**Multi Cell Changer—Lifetimes**

Use the **Multi Cell Changer** dialog to manually move the Multicell Holder accessory to a new cell. To access the **Multi Cell Changer** dialog, select the **Go to cell** button from the **Multicell/Temperature Accessories** page.

The dialog comprises the following:

**Multicell changer**

To manually move the Multicell Holder accessory to a new cell, simply select the corresponding button of the cell you require in the Multi Cell Changer dialog. The Multicell Holder will move the selected cell to the measurement position in the sample compartment.

The Multi Cell Changer dialog is ‘modeless’ which means that it can remain open even once you have cleared the **Setup** dialog. This enables you to use the **Rapid Read** command to perform a reading at the cell specified in the Cell Changer.

If you move to a new cell after you have performed a Multi-zero scan, the Cary Eclipse will display the intensity reading corrected by the determined zero-offset amount for that cell.

**Note**

You can use this option to check the intensity of a cell or start a lifetimes run at a particular cell.

See also:

- Multicell/Temperature Accessories
- Rapid Read

**Analyze—Lifetimes**

Use the Analyze page to select from various mathematical equations and set the parameters used to generate
the calculation of reaction rates.

From the Setup menu, select Analyze to access the Analyze page. You can also access the page from the Setup dialog by selecting the Analyze tab. To access the Setup dialog, select the Setup button from the Kinetics Application window.

The page comprises the following:

**Decay time**

**Calculate**  
Select whether to perform a decay time calculation of the rate or the lifetime. The units of ms or s will vary depending upon the X Mode chosen on the Cary page. The Manual guess equations will also vary according to the type of calculation you select.

**Auto Calculate**  
Select this option to automatically perform a rate calculation on collected data at the end of each run. Otherwise you can perform the calculation manually, once the run is completed, by selecting Recalculate from the Lifetimes Application window.

**Decay Time Table**  
Use the Decay Time Table to enter the calculation Start and Stop times (in minutes or seconds) for a rate or lifetime calculation analysis.

**Note**  
This option is only available if you select the Advanced Calculate option.

The table comprises the following:

**Start**  
Use these fields to enter the starting point, in minutes, for a rate calculation analysis.

**Stop**  
Use these fields to enter the end point, in minutes, for a rate calculation analysis.

**Function type**  
Click in this field to access a drop down list where you can select the function to be used for calculating the decay time. The following options are available:

**Single Exp**  
Select this to fit the data with a single exponential decay curve. The form of the equation will vary depending on whether the Rates or Lifetimes option is chosen. The lifetime is defined as the time taken for the intensity to decay to 1/e of it’s initial value.

**Double Exp**  
Select this to fit the data with a double exponential decay curve. The form of the equation will vary depending on whether the Rates or Lifetimes option is chosen. The lifetime is defined as the time taken for the intensity to decay to 1/e of it’s initial value.

**Plot**  
Select the way in which the plot will be displayed from the following options:

**Fit**  
Select Fit to automatically overlay the calculated lines of best fit onto the plotted data.
This option enables you to manually enter the parameters for the decay rate calculation. You will need to have a reasonable estimate of the values for these fit parameters, as they will be used as a first guess in the fitting procedure.

**Note**
If you do not choose Manual Guess then the Cary Eclipse system will automatically calculate the values for Intensity(0), Intensity(∞) and Rate when you select OK to exit the Setup dialog.

**Manual Guess**
Select this option to manually select the parameters with which to calculate a first order or second order reaction rate.

The value of these parameters will be used as a first guess in the fitting procedure.

**Parameters**

- **A1**
  Enter the estimated intensity of the pre-exponential for the first decay process

- **A2**
  Enter the estimated intensity of the pre-exponential for the second decay process. This option is only accessible if you have selected **Double Exp** as the **Function type** in the Decay Time table.

- **k1/τ1**
  Enter the estimated value of the rate or lifetime for the first decay process.

- **K2/τ2**
  Enter the estimated value of the rate or lifetime for the second decay process. This option is only accessible if you have selected **Double Exp** as the **Function type** in the Decay Time table.

- **C**
  Enter the estimated value of the intensity at the end of decay.

**Status Display**
Select this check box to display the **Status Display** window. This window allows you to view the status of various parameters during the data collection.
Recalculate—Lifetimes

Use the pages accessible via the Recalculate option to perform re-calculations on data that has been analyzed. These re-calculations can be made using new Setup parameters. For example you may wish to change the order of a reaction. You can also use the Recalculate button to access Report options.

The Recalculate button is accessible from the Lifetimes Application window. When you select the Recalculate option, the Setup dialog will appear, containing the following tabbed pages:

- **Analyze**
- **Reports**

See also:
- **Lifetimes Application**

Reports—Lifetimes

Use the Reports page to set up the report parameters associated with data collection.

From the Setup menu, select Reports to access the Reports page. You can also access the page from the Setup dialog by selecting the Reports tab. To access the Setup dialog, select the Setup button from the Lifetimes Application window.

**Hot Tip**

You can view your generated report before you print it out using the Print Preview command in the File menu.

If you decide to make changes to the report layout, select (Edit Report) from the Report toolbar. This enables you to directly edit the report.

The page comprises the following:

**Operator**

**Name**

Enter the instrument operator name in this text entry field. Use the arrow to the right of this field to view and select one of the last ten entries in this field.

**Note**

In GLP mode the system will automatically enter the operator’s name and this field will not be editable.

**Comment**

Enter operator details and comments in this text entry field.

**Options**

This group enables you to select the information to include in your report.
**Auto Print**
Select Auto Print to print a report automatically during the data collection. If Auto Print is selected, then the system will send the report information to the specified printer(s) as well as the Report area. However if Auto Print is not selected, the report will only be sent to the Report area and can be viewed by selecting Report in the View menu.

To automatically obtain a copy of the graphics, you will need to select both the Auto Print and Graph options (see below). In this case, all graphical information displayed on screen will be printed with the report. Therefore it is advisable to arrange the graphical information as you require on screen, prior to printing.

For example, if you require a single graph print out, then the Single/Multi Graph option in the Graph menu should be changed to the correct setting.

**Parameters**
Select this option to include the method setup parameters within your report.

**User data form**
Select this option to include in the report any information entered in the User Data Form.

**Graph**
Select Graph to include any graphs or graphic information associated with your report. If you require a printout of the displayed graphics only, you must ensure that only this option is selected. Press the Clear Report button to clear any information in the Report area before you print otherwise this will be included in your printout.

**% Page Height**
Enter the size at which you want the graph to be printed, as a percentage of total page height. For example, if you want graphical information to take up half of the page, enter 50% in this field.

**Company Logo**
Check this option to include your company logo in the report. A company logo will only be included if you have added one on the Company page of the System Information Application.

**Hot Tip**
If you have not included a company logo, select the Header/Footer button to open the System Information Application and use the Add Logo option to add a logo.

**Header/Footer**
Select the Header/Footer button to open the System Information application where you can enter the Header and Footer information to be included in your report. You can also enter any additional comments, and add a logo to your report. Once you have made all your changes, select OK to exit the System Information application and return to the Reports page of the Lifetimes application.

**Focused trace**
Select this option to apply the Report selections to the focused trace only.

**All traces**
Select this option to apply the Report selections to all visible traces.

**X-Y Pairs Table**
Select this if you want to print a table showing the X-Y pairs of the collected data in your report.

**Data Interval**
**Actual**
Select Actual to report the X-Y pairs table for the trace at the actual Data Interval at which the data was collected.

**Interpolated**
Select Interpolated to report the X-Y pairs table for the trace at an interpolated data interval. Enter this data interval in the adjacent field. If no data has been collected at the interval you enter, the Cary Eclipse software will interpolate the data to produce an X-Y result.
Status Display

Select this check box to display the Status Display window, where you can view the status of your lifetimes run during the data collection.

Hot Tip
You can copy a picture, spreadsheet file or document from another application (for example, Microsoft Word or Microsoft Excel) into the report area of the of the Lifetimes application. To edit these objects, simply double click on them to open the associated application in which they were created.

See also:
View menu
User Data Form
Status Display

Reports Toolbar—Lifetimes

Use the Reports toolbar buttons to gain quick access to many of the Edit menu options. You can also use the toolbar to cut, copy and paste information and to change the look of your report.

To display or hide the toolbar, point to the Toolbars option in the View menu and select Reports. To display or hide the toolbar's text labels, point to the Toolbar option in the View menu and select Text Labels.

Hot Tip
To see the name of a button, point to it with the mouse.

The following commands can be accessed from the Reports toolbar:

<table>
<thead>
<tr>
<th>Button</th>
<th>Menu command</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>![Edit Report]</td>
<td>Edit Report</td>
<td>Click this button to edit the report. This command is also accessible by pressing CTRL+R.</td>
</tr>
<tr>
<td>![Undo]</td>
<td>Undo</td>
<td>If available, this undoes the last action. This command is also accessible by pressing CTRL+Z.</td>
</tr>
<tr>
<td>![Cut]</td>
<td>Cut</td>
<td>Click this button to remove the highlighted text and place it on the Windows Clipboard. This command is also accessible by pressing CTRL+X.</td>
</tr>
<tr>
<td>![Copy]</td>
<td>Copy</td>
<td>Click this button to copy the highlighted text onto the Windows Clipboard. This command is also accessible by pressing CTRL+C.</td>
</tr>
<tr>
<td>![Paste]</td>
<td>Paste</td>
<td>Click this button to insert the contents of the Windows Clipboard (text only) at the cursor. Any highlighted text will be replaced. This command is also accessible by pressing CTRL+V.</td>
</tr>
<tr>
<td>![Delete]</td>
<td>Delete</td>
<td>Click this button to remove selected text in the Report area. This command is also accessible by pressing DEL. Note This option is only available once you select Edit Report and there is text highlighted in the Report area.</td>
</tr>
<tr>
<td>![Font]</td>
<td>Font</td>
<td>Click this button to change the font of new or selected text in the report. For existing text, simply highlight the text in the report, select this button and make your changes</td>
</tr>
</tbody>
</table>
Clear Report—Lifetimes

Select **Clear Report** to remove the contents of the Report area. If you have saved the report, selecting this option does not delete the contents of the report, the information is simply removed from the Report Area.

You can select the **Clear Report** button from the Lifetimes Application window or the **Clear Report** command from **Commands** menu.

See also:
- [Lifetimes Application](#)
- [Commands menu](#)

How to use advanced reporting options—Lifetimes

This outlines ways to fully customize your reports. Options available include:

- Including header and footer information
- Including a logo
- Editing the text in the report
- Viewing the report on screen before printing
- Cutting and pasting to a word processor

**Including header and footer information**

You can have text appearing at the top and bottom of every page in your report. To specify what the text should be, do the following:

**Step 1**

On the Windows Taskbar, click the Windows **Start** button, point to **Programs, Cary Eclipse** and then select **System Information**. The System Information application appears with the Company page displayed.

**Step 2**

Enter the header and footer information in the **Header and Footer** fields.

**Step 3**

Click **OK** to implement your changes.

**Including a logo**
You can include a logo on the first page of your report. To specify what the logo should be, do the following:

Step 1

On the Windows Taskbar, click the Windows Start button, point to Programs, Cary Eclipse and then select System Information. The System Information application appears with the Company page displayed.

Step 2

Enter a logo using the Add Logo button. This logo will always be included on the front page of reports.

Step 3

Click OK to implement your changes.

Q. Why isn't the logo I selected included in my report?
A. If you are running the Cary Eclipse software under Windows NT you need to be logged on as an Administrator in order to specify the logo in the System Information application.

Editing the text in the report

You can edit any of the text in the Report area. Just select Edit Report from the Edit menu. You can then add, delete and change text in the Report area.

Q. The Edit Report option is not available, why not?
A. You do not have reporting editing privileges set in the GLP Administration application. The Administrator of the system will have to change your privileges before you can edit reports.

Viewing the report on the screen before printing

You can view a report before printing it. To do this, from the File menu select Print Preview. You can then ensure that the report format is correct before you print it.

Hot Tip
Use the arrow keys at the bottom of the Print Preview screen to change pages.

Cutting and pasting to a word processor

To have complete control over how your report looks you may wish to copy it to a word processor program, such as Microsoft Word. To do this:

Step 1

Open a report.

Step 2

From the Edit menu, choose Edit Report.

Step 3

Click in the Report area and highlight the text you want to export to your word processor.
Step 4

Right-click on the Report area and select **Copy** from the menu.

Step 5

Start up your word processor and use the Paste function (usually under the **Edit** menu) to place the text into a document.

See also:
- How to add a label to a graph
- How to add a picture to a graph
- How to alter the look of your graphs
- How to display a trace that is not appearing

**Auto Store—Lifetimes**

Use the **Auto store** page to specify if and when data will be stored, and in what format. You can also use this page to set up the system to automatically send an email at the end of a read.

From the **Setup** menu, select **Auto store** to access the **Auto store** page. You can also access the page from the **Setup** dialog by selecting the **Auto store** tab. To access the **Setup** dialog, select the **Setup** button from the Lifetimes Application window.

The page comprises the following:

**Storage**

- **Off**
  - Select this if you do not wish to be automatically prompted to save collected data.
  
  **Note**
  - If you wish to store your unsaved data manually, choose **Save Data As…** from the **File** Menu to save your data after the run is complete. You can save your data as a **Batch** or **Data** file.

- **On; Prompt At Start**
  - Select this to display the Windows **Save As** dialog at the start of the collect where you can enter the Batch filename for your data.

- **On; Prompt At End**
  - Select this to display the Windows **Save As** dialog at the end of a collect where you can enter the Batch filename for your data.

**Auto convert**

Use the Auto convert options to convert your data into an ASCII file format that can be opened in spreadsheet programs such as Microsoft Excel.

- **None**
  - Select **None** if you do not need to generate an ASCII file.

- **ASCII (csv)**
  - Select this option to save the data as a comma delimited ASCII Spreadsheet file (*.CSV) file. Only the currently collected data is converted.

  ASCII files are stored in the same folder as the data file from which they were generated and designated the same filename as the method used to collect the data plus the extension
‘CSV’.

**ASCII (csv) with log**

Select this option to save the data as a comma delimited ASCII Spreadsheet file (*.CSV) file. Only the currently collected data is converted. The key method parameters used to collect the data are also saved in this file.

ASCII files are stored in the same folder as the data file from which they were generated and designated the same filename as the method used to collect the data plus the extension ‘CSV’.

**Note**

If you choose to auto-convert the data to an ASCII file but you have set the Storage option to Off, then the ASCII file will be saved with the following format:

DATE TIME. CSV

The date and time will appear in the same form that is set in the Windows Regional Settings Properties dialog that is accessed from the Windows Control Panel.

For example, 22_4_97 1;32;08.CSV

**Q. Why are the Auto Convert options grayed out?**

**A.** The Auto Convert options are only applicable when setting up data collection reports. Therefore, they are only available when you access the Setup dialog by selecting the Setup button or selecting the Setup command from the Setup menu. When you access the Setup dialog by selecting the Recalculate button, the Auto Convert options are grayed out.

**Tell me more about ASCII Spreadsheet Files**

**Email**

Select the Email check box and enter the Email details to send an email to a person once the read is completed. This useful facility can be used to alert you to the completion of the read, particularly if you are working away from the laboratory where the Cary Eclipse instrument is. You can also attach the collected data to the email.

**Note**

You must set up the email Host and Port options on the Email options page of the System Information application in order for the email facility to work correctly.

- **To** Enter the recipient's email address. You can enter more than one recipient in this field.
- **From** Enter your email address (or the email address associated with the computer on which the Cary Eclipse software is installed).
- **Subject** Enter the subject of your email (or leave this field blank if you do not wish to include a subject).
- **Cc** Enter the email address of any recipient you want to copy the email to.
- **Message** Enter your message.
- **Attached…** Use the Attached button include a file attachment in your email. To do this, select Attached and browse your computer to select a file, using the Windows® Open dialog that appears.

**Attach current data** Select this option to attach the data that has been collected during the run to the email.

**How to email your results**

**Status display**
Select this check box to display the **Status Display** window, where you can view the status of your lifetimes run during the data collection.

---

**See also:**
- Save As dialog
- Status Display

### How to email your results—Lifetimes

You may wish to send an email to a person once the read is completed. This useful facility can be used to alert you to the completion of the read, particularly if you are working away from the laboratory where the Cary Eclipse instrument is.

To do this:

**Step 1**

Ensure that you have set up the **Host** and **Port** options on the **Email options** page of the System Information application in order for the email facility to work correctly.

**Step 2**

From the **Setup** menu, select **Auto Store**. The **Auto Store** page is displayed.

**Step 3**

Select the **Email** check box to enable the email options.

**Step 4**

In the **To** field, enter the recipient's email address (for example, *rchandler@varianinc.com*). To enter multiple recipients, simply type a comma ',' between each email address.

**Step 5**

In the **From** field, enter the email address of the computer on which the Cary Eclipse software is installed. This may be your email address, or a communal email address, if the Cary Eclipse software is installed on a computer that has multiple users.

**Step 6**

Enter information in the **Subject** and **Message** fields as required.

**Step 7**

Select **Attach current data**. The data that has been collected during the run will be attached to the email.

---

**Hot Tip**

You can attach other documents to your email. To do this, select the **Attached** button and browse your computer to select a file, using the Windows® **Open** dialog that is displayed.

**Step 8**
Select **OK** to clear the **Auto Store** dialog. Your email will be sent once the run has been completed.

See also:
- [Auto Store](#)

### Performing scans—Lifetimes

- Starting a run
- Sample Name dialog
- Cell Loading Guide
- Start/Stop buttons
- Pre-scan dialog
- Rapid Result dialog
- Zero dialog
- Go To dialog
- Commands menu
- How to perform a temperature controlled run

### Information about starting a collect—Lifetimes

The sequence that occurs when you select **Start** to commence the lifetimes run depends upon your storage and accessory selections.

#### Storage selections

<table>
<thead>
<tr>
<th>If you have selected … as the Storage option on the Auto Store page</th>
<th>The following occurs</th>
</tr>
</thead>
<tbody>
<tr>
<td>On, prompt at start</td>
<td>The Windows <strong>Save As</strong> dialog will appear as soon as you select <strong>Start</strong>. You can save the current method, collected data, report and graph template as a Batch file.</td>
</tr>
<tr>
<td>On, prompt at End</td>
<td>The Windows <strong>Save As</strong> dialog will appear at the end of the data collection. You can save the current method, collected data, report and graph template as a Batch file.</td>
</tr>
<tr>
<td>Off</td>
<td>The Windows <strong>Save As</strong> dialog will not appear.</td>
</tr>
</tbody>
</table>

#### Accessory selections

<table>
<thead>
<tr>
<th>If you have selected … on the Accessories page</th>
<th>The following dialog is displayed</th>
</tr>
</thead>
<tbody>
<tr>
<td>No accessories</td>
<td>The <strong>Sample Name</strong> dialog will appear when you select <strong>Start</strong>, prompting you to load the sample and enter your sample name. Select <strong>OK</strong> to commence the lifetimes run.</td>
</tr>
<tr>
<td>Multicell Holder</td>
<td>The <strong>Cell Loading Guide</strong> dialog will appear. Enter the sample name of each activated cell then select <strong>OK</strong> to commence the</td>
</tr>
</tbody>
</table>
Sample Name—Lifetimes

If you are using a single cell holder, the **Sample Name** dialog appears once you select the **Start** button to commence an analysis.

The page comprises the following:

**Sample Name**  Enter the name of the sample to be used in the lifetimes run. Click on the arrow to the right of the field to view and select the most recently entered sample names.

Once you have entered the **Sample Name**, select **OK** to commence the lifetimes run.

See also:  
Sample Name  
Cell Loading Guide  
Start/Stop buttons  
Setup  
Accessories  
Auto Store  

Cell Loading Guide—Lifetimes

If you are using a Multicell Holder, the Cell Loading Guide dialog appears once you click the **Start** button to commence an analysis.

The Cell Loading Guide dialog prompts you to insert blank samples in the Multicell Holder to collect the Lifetimes scans. You can also use the dialog to name the samples in each selected cell in the Multicell Holder.

The dialog comprises the following:

**Loading Guide/Sample Name entry**  Enter the names of the sample to be used in the lifetimes run in each available field. This sample name will be used as the **Name of Trace** in the **Trace Preferences** dialog.

**Note**  
The Cell Loading Guide dialog appears only if you have selected the **Multicell Holder** option on the **Accessories** page of the **Setup** dialog. Only cells selected in the Multicell Holder diagram on the **Accessories** page will be available in the Loading Guide.

In the adjacent example, cells 1—3 will be available in the **Loading Guide** dialog, as they have been selected in the Multicell Holder diagram.

Once you have entered the **Sample Name**, select **OK** to commence the Lifetimes run.

See also:  
Accessories
The following buttons may be displayed at the top of the Lifetimes Application window. The **Start** and **Connect** buttons will change depending upon the status of the run. For example, if a run has not been started, the **Start** button will be visible to indicate that the system is ready to commence a run.

Select **Start** to commence a lifetimes run using the current Setup parameters. If any unsaved data exists in the Graphics area you will be prompted to save the data. If you choose not to save, any unsaved data, pictures and labels will be deleted when you start the lifetimes run.

If another application is already online with the Cary Eclipse, the **Start** button will change to **Connect**, and you will need to select **Connect** to bring this application online before the **Start** button will re-appear. However, you will not be able to **Connect** if the other application is collecting data.

Select this to end a lifetimes run. If you have selected **Storage On** on the **Auto Store** page of the **Setup** dialog, all data collected is automatically stored in the file specified in the Batch name field of the Windows **Save As** dialog before or after the run commenced.

Before you select **Zero**, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.

When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the Lifetimes Application window.

Select the **Clear Zero** command from the **Commands** menu to remove zero correction and thus remove the subtraction effect.

To access the **Zero** option, select the **Zero** command from the **Commands** menu, or select the **Zero** button from the Lifetimes Application window.

Pre-scan—Lifetimes
Select **Pre-scan** to determine the optimum instrument parameters to be used for your sample. At the end of this collection sequence the software will report the optimum Ex and Em wavelengths and PMT voltage. Once you are satisfied with the parameters, the software will automatically update your method setup to reflect these new parameters.

To access the **Pre-scan** option, select the **Pre-scan** command from the **Graph** menu, or select the **Pre-scan** button from the Lifetimes Application window.

The Pre-scan dialog comprises the following:

### Type of scatter

Select the type of scatter you wish to search for from the following:

- **Rayleigh scatter**
  - Rayleigh scatter refers to the scatter of the excitation light. It is displayed at the peak of the excitation wavelength.

- **Raman scatter**
  - Raman scatter refers to scatter due to the solvent. Its peak is at a fixed energy from the excitation wavelength, and is solvent dependent. Thus, the type of solvent used must be selected from the **Solvent** list.

- **2nd order**
  - Second order scatter refers to the scatter of higher order excitation light. Its peak is a multiple of the excitation wavelength.

See also:

- **Graph menu**
- **Lifetimes Application**

### Go to Wavelength—Lifetimes

Use the **Go to Wavelength** dialog to temporarily change the instrument to a new excitation or emission wavelength. You would mainly use this dialog if you want to quickly go to a wavelength and check the intensity reading of the sample. The lamp will flash continuously in this mode. This option is only available if the instrument is not currently collecting data.

To access the **Go to Wavelength** dialog, select the **Go To Wavelength** command from the **Commands** menu. You can also select F4 to open the dialog.

**Note**

This is not the wavelength used in the application method. If your method uses a wavelength that is different from the 'Go to wavelength' setting you select, then when you select **Start** the system will change to the wavelength selected. (In most applications, this is set in the **Cary** page of the **Setup** dialog.)

The Go to Wavelength page comprises the following:

- **Excitation**
  - **Wavelength**
    - Enter the new wavelength in this numeric entry field. When you select **Go to** the instrument will change to the new value.

- **Zero Order**
  - Select Zero Order to drive the Cary Eclipse fluorescence spectrophotometer to 0 nm (white light). You can then use this light for aligning accessories.

- **Emission**
  - **Wavelength**
Enter the new wavelength in this numeric entry field. When you select **Go to** the instrument will change to the new value.

**Zero Order**
Select **Zero Order** to drive the Cary Eclipse fluorescence spectrophotometer to 0 nm (white light). You can then use this light for aligning accessories.

**Go To**
Click **Go to** once you have entered a new wavelength, to change the instrument to the entered value.

---

**Rapid Result—Lifetimes**

Use the Rapid Result dialog to take a reading of a blank and sample/s, without having to change the current application method. You can use the **Multi Cell Changer** dialog to specify the cell you want to read. This means that between runs, while new samples are being prepared and loaded, other samples can quickly be measured. The result is printed in the Report area.

To access the **Rapid Result** dialog, select **Rapid Result** from the **Commands** menu or press F6.

**Excitation**

**Wavelength**
Enter the wavelength at which you want to take a reading.

**Zero order**
Select this option to set the excitation monochromator to zero order (white light).

**Emission**

**Wavelength**
Enter the wavelength at which you want to take a reading.

**Zero order**
Select this option to set the excitation monochromator to zero order (white light).

**Zero**
Click Zero to change the wavelength to the entered value and measure a blank.

**Result**
Click Result to change the wavelength to the entered value and measure a sample. The X,Y results are corrected for the blank and the results appear in the report window.

---

**Commands menu—Lifetimes**

Use the **Commands** menu to access major operational commands in the Advanced Reads application. The menu is accessible from the menu line at the top of the Lifetimes Application window.

Alternatively you can use function key shortcuts for some of the commands. These appear next to the item in the Commands menu. Note that not all commands appear as both button and menu items.

**Hot Tip**
If you access these commands using the **Commands** menu, rather than from the application buttons, you can remove these buttons from your application window. To do this, clear the **Buttons** command in the **View** menu.
Tell me how to remove or display the command buttons

The **Commands** menu comprises the following commands:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Start</strong></td>
<td>Select this to start a data collection using the currently set up parameters. Note: If the <strong>Connect</strong> command is displayed in place of the <strong>Start</strong> command, this indicates that another Cary Eclipse application is in use and has control of the Cary Eclipse instrument. To make the Advanced Reads application active so that you can perform a read, you will need to select <strong>Connect</strong>. However, you will not be able to select <strong>Connect</strong> if another application is collecting data.</td>
</tr>
<tr>
<td><strong>Stop</strong></td>
<td>Select this to stop the data collection. If you have selected <strong>Storage On</strong> in the <strong>Auto Store</strong> page of the Setup dialog, all data collected is automatically stored in the file specified in the Batch name field of the Windows Save As dialog before or after the run commenced.</td>
</tr>
<tr>
<td><strong>Clear Zero</strong></td>
<td>Select this option to clear the <strong>Zero</strong> command. When you select the <strong>Zero</strong> command (from the <strong>Commands</strong> menu or by pressing F5), the intensity at that time will be reset to zero and all subsequent results will have the intensity value that was reset to zero subtracted from them. Selecting <strong>Clear zero</strong> cancels this zeroing (and thus subtraction) effect.</td>
</tr>
<tr>
<td><strong>Pre-scan</strong></td>
<td>Select <strong>Pre-scan</strong> to quickly determine the optimum instrument parameters to be used for your sample. At the end of this collection sequence the software will report the optimum Ex and Em wavelengths and PMT voltage. Once you are satisfied with the parameters, the software will automatically update your method setup to reflect these new parameters.</td>
</tr>
</tbody>
</table>
| **Zero order** | Select the **Zero order** command to set one of the monochromators to zero order. If you use this command to zero either the excitation or emission monochromators, this will override the **Ex. Wavelength** or **Em. Wavelength** set on the **Cary** page. The Cary Eclipse software will use the zero setting when collecting data. The **Zero order** command is generally used during alignment procedures.  

**Excitation**  
Select this command to set the excitation monochromator to zero order (white light).  

**Emission**  
Select this command to set the emission monochromator to zero order (white light). |
| **Reset**    | Select this command to re-initialize the instrument. This will perform the same initializing procedure that is performed when the instrument is powered on. |
| **Rapid Result** | Select this to display the **Rapid Result** dialog. Use this dialog to take a reading of a blank and multiple sample readings without changing the application method. This means that between runs, while new samples are being prepared and loaded, other samples can quickly be measured. This command can also be accessed by pressing F6. |
| **Zero**     | Select this to zero the current intensity value. The intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before selecting **Start** if you want your data to start at 0 intensity. This command is also accessible by pressing F5. Note |
How to perform a temperature controlled lifetimes run—Lifetimes

This demonstrates how to perform a fluorescence single cell, multi-rate lifetimes run at 37°C using the single cell Peltier accessory.

Follow the steps through, adjusting them to meet the requirements of your particular data collection.

- Start the Lifetimes application
- Set up data collection parameters
- Ensure Multicell Holder accessory is not selected
- Set up accessories for reaction temperature control and temperature display
- Set up instrument parameters
- Set up rate parameters
- Set up collection display options
- Set up rate calculation, rate order an other rate parameters
- Set up reporting and printing requirements
- Set up storage of collected data
- Set up visual system monitoring
- Finish Setup
- Start the lifetimes run

Start the Lifetimes application

Step 1
Click the Windows **Start** button, point to **Programs, Cary Eclipse** and then select **Lifetimes**. The Lifetimes Application is displayed.

**Note**
If you are running a GLP system, you may be prompted to enter a password before accessing the Lifetimes application.

**Set up data collection parameters**

Step 2 [Setup dialog]

Select **Setup** to display the **Setup** dialog and specify the method parameters for a new method.

To do this:

**Ensure Multicell Holder accessory is not selected**

Step 3 [Setup dialog | Accessories page]

Select the **Accessories** tab to display the **Accessories** page. On the **Multicell/Temperature** tab, ensure that the **Multicell Holder** accessory is not selected.

**Set up accessories for reaction temperature control and temperature display**

Step 4 [Setup dialog | Accessories page]

a. Select the **Temperature control** check box to enable the Temperature group.

b. Click on the down arrow adjacent to the **Device** field and select the **Single Cell Peltier** accessory.

c. Set the **Temperature** to 37°C.

d. In the **Temperature Display** group, select **Probe 1** to view the temperature of one temperature probe in the **Status Display** window.

**Set up instrument parameters**

Step 5 [Setup dialog | Cary page]

a. Enter the **Delay Time**, which is the time that elapses between the last flash and the beginning of data collection. Setting a delay time ensures that there is no lamp effects in the phosphorescence decay curve.

b. Enter, in milliseconds, the **Gate Time** for your sample. The gate time is the overall reading time of the emission signal.

c. Enter the **No. of Flashes**, which is how many times the lamp flashes consecutively before the delay time is counted.

d. In the **Ex. Wavelength** field, set the wavelength of the excitation monochromator, which is the wavelength value at which you want to excite your sample.

e. In the **Em. Wavelength** field, set the wavelength of the emission monochromator, which is the wavelength value at which your sample emits.
f. Set the slit width of the excitation monochromator.
   Note that as a general rule, at a fixed emission slit width, increasing the excitation slit width will result in an increase in the fluorescence excitation intensity.

g. Set the slit width of the emission monochromator.
   Note that as a general rule, at a fixed excitation slit width, a two-fold increase in the emission slit width will result in a four-fold increase in the emission intensity. Accordingly, a two-fold decrease in the emission slit width will result in a four-fold decrease in the emission intensity.

**Set up rate parameters**

Step 6 [Setup dialog | Cary tab]

a. Enter the **Total Decay Time**. You should enter a time that will allow for the phosphorescence signal to decay to approximately zero. The lamp will not flash until the total decay time has elapsed.

b. Enter the **No. of Cycles** (or decay curves) over which the run will be collected. This number will be used to average the data. Entering a number of cycles value enables you to obtain better signal to noise ratios as multiple traces can be averaged.

**Set up collection display options**

Step 7 [Setup dialog | Options page]

a. Select the **Options** tab. The **Options** page is displayed.

b. Enter the minimum and maximum Y values to be displayed on the graph axis during the data collection.

c. Set the **Excitation filter** for the concentration measurement. It is recommended that you set the filter to **Auto** (the default setting). This will automatically move the filter wheel to the appropriate position for the selected excitation wavelength.

d. Set the **Excitation filter** and the **Emission filter** for the concentration measurement. In order to minimize steps in the spectra, it is recommended that you set the filter to **Open** (the default setting).

e. Set the **PMT Detector Voltage**. The setting you choose will depend upon your sample. For example, a **Low** setting is recommended for highly emitting samples, while a **High** setting is recommended for low emitting samples.

**Set up rate calculation, rate order and other rate parameters**

Step 8 [Setup dialog | Analyze page]

a. Select the **Analyze** tab. The **Analyze** page is displayed.

b. Select the **Auto Calculate** check box to automatically perform a rate calculation on collected data at the end of each run.

c. In the Decay Time table, enter the calculation **Start** and **Stop** times (in minutes or seconds) for a rate or lifetime calculation analysis and select the **Function Type**.

d. You can select the **Manual Guess** check box and manually enter the parameters: Intensity(0), Intensity
\((\infty)\) and Rate. It is presumed that you have a reasonable idea of the values for these fit parameters, as they will be used as a first guess in the fitting procedure.

**Note**

If you do not choose **Manual Guess**, the Cary Eclipse system will automatically calculate the values Intensity(0), Intensity(\(\infty\)) and Rate when you select **OK** to exit the Setup dialog.

f. Set the **Plot** type to **Fit** to automatically overlay the calculated lines of best fit onto the plotted data.

**Set up reporting and printing requirements**

Step 9 [Setup dialog | Reports page]

a. Select the **Reports** tab to display the **Reports** page where you can specify your reporting requirements for this method.

b. Enter your name in the **Name** field.

c. Enter a comment relating to your experiment in the **Comment** field *(optional)*.

d. Set up your report style by selecting the appropriate check boxes in the **Options** group.

For example, select the **Auto Print** check box to obtain a printout of your report automatically. Select the **Parameters** check box to include your experimental parameters in the report. Select the **Graph** check box to include a calibration graph in the generated report.

**Note**

If **Auto Print** is selected, then the system will send the report information to the specified printer as well as the Report area. However if **Auto Print** is not selected, the report will only be sent to the Report area and can be viewed by selecting the **Report** command from the **View** menu.

e. Select the **X-Y Pairs Table** check box to view a list of abscissa values and their corresponding intensity values.

**Set up storage of collected data**

Step 10 [Setup dialog | Auto Store page]

a. Select the **Auto Store** tab to display the **Auto Store** page.

b. Set the **Storage** option to **On; prompt at start**. You will be prompted to save your method (with calibration), collected data and report when you commence the run.

c. Select the **Auto Convert** option you require. If you select **ASCII (csv)** or **ASCII (csv) with log**, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format. The data file will be saved in the current folder.

**Set up visual system monitoring**

Step 11 [Setup dialog]
Select the **Status Display** check box on any of the Setup pages to display various information fields on your current reaction.

**Finish Setup**

Step 11

Once you are satisfied with your method setup select **OK** to confirm any changes you have made and clear the **Setup** dialog.

**Zero the instrument**

Step 13

a. Select the **Zero** button to zero the system. Alternatively, select **Zero** from the **Commands** menu to perform a zero. The **Cell Loading Guide** dialog will be displayed.

b. Place the blank solution in the sample compartment and select **OK**. The system will perform an instrument zero on the blank solution.

**Start the lifetimes run**

Step 14

a. Select the **Start** button to commence a data collection. Alternatively, select **Start** from the **Commands** menu. **Do not add your active reagent at this time.** The system will display the **Save As** dialog.

b. Enter the file name for this lifetimes run in the **File name** field and select **Save**. The system will display the **Cell Loading Guide** dialog.

c. Place the sample solution in the correct cell position and select **OK**. The **Sync Start** dialog will be displayed.

d. Add your active reagent just before the **Count Down** reaches 0:00 or commence the data collection by selecting the **OK** button.

See also:

- [How to perform post run calculations on previously stored data](#)
- [How to export collected data](#)

**Viewing run progress and results—Lifetimes**

- Reports page
- Reports toolbar
- Clear Report
- View menu
- Edit menu
- Status display
- User Data Form
- Print Preview dialog
- Print
## View menu—Lifetimes

The View menu is accessible from the menu line at the top of the Lifetimes Application window.

**Hot Tip**

Highlight a method or data file in Explorer and simply drag and drop it onto the application to open it. Any data will be displayed in the Graphics area.

The following commands can be accessed from the View menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Toolbar</strong></td>
<td>Select this option to toggle the Graphics and Reports toolbar on and off. You can also select to display or hide the toolbar Text Labels.</td>
</tr>
<tr>
<td><strong>Buttons</strong></td>
<td>Select this option to toggle the left-hand button display (button area) on and off.</td>
</tr>
<tr>
<td><strong>Graphics</strong></td>
<td>Select this option to toggle the Graphics area on and off.</td>
</tr>
<tr>
<td><strong>Report</strong></td>
<td>Select this option to toggle the Report area on and off.</td>
</tr>
<tr>
<td><strong>Report Vertical</strong></td>
<td>If the Graphics option is selected, select Report Vertical to display the Graphics area, Report area and/or ADL Editor side by side vertically.</td>
</tr>
<tr>
<td><strong>ADL</strong></td>
<td>Select this option to toggle the ADL Editor on and off.</td>
</tr>
<tr>
<td><strong>ADL Vertical</strong></td>
<td>If the ADL and Report options are selected, select ADL Vertical to display the ADL Editor vertically on the right hand side of the Report area.</td>
</tr>
<tr>
<td><strong>ADL Quick Reference</strong></td>
<td>This displays the Quick Reference window where you can easily add a command in the correct syntax to your ADL program. A detailed description of all the commands in the Quick Reference is available by selecting the Help button.</td>
</tr>
</tbody>
</table>
| **ADL Hooks** | Select this to activate a link (or hook) to an ADL program at a specific point in the application, and then run that ADL program. For example, ADL hooks have been supplied to run specific programs:  
  - on start-up of the Cary Eclipse software; 
  - when the system goes online or offline; 
  - when the Start button is selected; 
  - when a run finishes; 
  - when a zero is performed etc.  
  
  For the hook to work, ADL Hooks must be selected, and the appropriate ADL file for the application must be present in the ADL folder in the directory. |
<p>| <strong>Hints</strong>     | Select this option to toggle the Hints bubble text on and off. The bubble text appears when you place the arrow cursor over a field, button or text entry field for a short period. It offers a quick explanation of the item’s function. |</p>
<table>
<thead>
<tr>
<th>Command</th>
<th>Button</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Undo</td>
<td><img src="image" alt="undo" /></td>
<td>If available, this undoes the last action. This command is also accessible by pressing CTRL+Z.</td>
</tr>
<tr>
<td>Cut</td>
<td><img src="image" alt="cut" /></td>
<td>Select this command to remove the highlighted text and place it on the Windows Clipboard. This command is also accessible by pressing CTRL+X.</td>
</tr>
<tr>
<td>Copy</td>
<td><img src="image" alt="copy" /></td>
<td>Select this command to copy the highlighted text onto the Windows Clipboard. This command is also accessible by pressing CTRL+C.</td>
</tr>
<tr>
<td>Paste</td>
<td><img src="image" alt="paste" /></td>
<td>Select this command to insert the contents of the Windows Clipboard (text only) at the cursor. Any highlighted text will be replaced. This command is also accessible by pressing CTRL+V.</td>
</tr>
<tr>
<td>Clear/Delete</td>
<td><img src="image" alt="clear" /></td>
<td>Select this command to remove selected text in the Report area. This option is only available once you select <strong>Edit Report</strong> and there is text highlighted in the Report area. This command is also accessible by pressing DEL.</td>
</tr>
<tr>
<td>Select All</td>
<td><img src="image" alt="select_all" /></td>
<td>Select this option to highlight all text in the Report area. This command is also accessible by pressing CTRL+A.</td>
</tr>
<tr>
<td>Font</td>
<td><img src="image" alt="font" /></td>
<td>This enables you to change the font of new or selected text in the report. For existing text, simply highlight the text in the report, select <strong>Font</strong> and make your changes in the <strong>Font</strong> dialog. When you select <strong>OK</strong>, the appearance of the highlighted text in the report will alter to reflect your font changes. You do not have to select <strong>Edit Report</strong> to use this command. This command is also accessible by pressing CTRL+F.</td>
</tr>
<tr>
<td>Edit Report</td>
<td><img src="image" alt="edit_report" /></td>
<td>Select this option to edit the report. This command is also accessible by pressing CTRL+R.</td>
</tr>
<tr>
<td>Clear Report</td>
<td><img src="image" alt="clear_report" /></td>
<td>Select this command to remove the contents of the Report area. If you have saved the report, selecting this option does not delete the contents of the report, the information is simply removed from the Report area.</td>
</tr>
<tr>
<td>Recalculate</td>
<td><img src="image" alt="recalculate" /></td>
<td>Use the pages accessible from the <strong>Recalculate</strong> option to perform re-calculations on data that has been analyzed.</td>
</tr>
</tbody>
</table>

**Edit menu—Lifetimes**

Use the **Edit** menu to copy, cut, paste and delete information. You can also use the menu to access the Report editing commands. The menu is accessible from the menu line at the top of the Lifetimes Application window.

The following commands can be accessed from the **Edit** menu:

See also:
- Lifetimes Application
- Graphics area
- Status Display
- ADL Hooks
- How to insert ADL commands using the Quick Reference
Status Display—Lifetimes

Use the Status Display window to view the status of your lifetimes run during the data collection. Depending upon settings you have for the lifetimes run, the following parameters may be displayed:

- Current Excitation Wavelength (nm)
- Current Emission Wavelength (nm)
- Excitation Slit Width (nm)
- Emission Slit Width
- PMT Voltage (V)

To access the Status Display, select Status Display from the View menu, or from any page of the Setup dialog. You can choose to view the Status Display at any time during a lifetimes run.

User Data Form—Lifetimes

Use the User Data Form to enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data.

To access the User Data Form, select the User Data Form command from the Graph menu, or select the User Data Form button on the Graphics toolbar. The User Data Form command is only available if you have selected a graph box.

The dialog comprises the following:

<table>
<thead>
<tr>
<th>Data Name</th>
<th>This column lists the names of each visible trace in the highlighted graph box.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Other Columns</td>
<td>Use this to record the name of the parameter in which you are interested as a column heading, (for example, pH, ionic concentration, substrate concentration etc.).</td>
</tr>
<tr>
<td>Note</td>
<td>You must enter a column heading, or else the data in that column will not be saved.</td>
</tr>
</tbody>
</table>

Print Preview—Lifetimes

Use the Print Preview dialog to view the current report before you print it. From the File menu, select Print Preview to access the dialog.
The contents of the Graphics area appear first in the preview area followed by the contents of the Report area. You can reduce or enlarge the print preview on screen. To do this, click the left mouse button in the preview area to toggle through the five available views. Click the right mouse button in the preview area to return to the full page view.

The page comprises the following commands:

- **First arrow**
  - Use this arrow to view the first page of the report.
- **Previous arrow**
  - Use this arrow to view the previous page of the report.
- **Next arrow**
  - Use this arrow to view the next page of the report.
- **Last arrow**
  - Use this arrow to view the last page of the report.
- **Print**
  - Use this to display the Windows Print dialog where you can choose the printer and pages to print as well as the number of copies you wish to print.

**Q.** Why are the arrow buttons grayed out?

**A.** The buttons will be grayed out if the report has only one page of data.

**Note**

The options on the Report page are used to generate the contents of the Report area during a run or on a recalculation. If **Auto Print** has been selected the Cary Eclipse will automatically print a report at the end of a data collection.

See also:
- File menu
- Print

**Print—Lifetimes**

Use the **Print** dialog to select the printer and pages to print of the currently displayed report as well as the number of copies you wish to print.

To access the **Print** option, select the **Print** command from the **File** menu, or select the **Print** button from the Lifetimes Application window.

All graphical information displayed on screen will be printed with the report, therefore it is advisable to edit the report on the screen prior to printing. You can also preview the report prior to printing by using the **Print Preview** command.

If you require a printout of the graphics in the Graphics area only, select the **Clear Report** button to erase the contents of the Report area before you print.

**Performing Calculations—Lifetimes—Lifetimes**
How to perform post run calculations on previously stored data—Lifetimes

Retrieve the data

Step 1

From the File menu, select Open to display the Windows Open dialog.

Step 2

A list of previously stored Batch files will appear. Select the Batch file you require then select Open.

The system will automatically retrieve the collected data into the Graphics area. The method that was used to collect the data will also be retrieved.

Note
You can also retrieve data from Data (*.FDLT), Method (*FMLT) and Spreadsheet ASCII (*.CSV) file types.

Set up the recalculation

Step 3

Select the Recalculate button to display the Setup dialog. The Analyze and Reports pages will be displayed to enable you to set up the recalculation parameters.

Step 4

Select the Analyze tab to access the Analyze page where you can set the analysis parameters you require.

For example, select the Auto Calculate option to automatically perform a rate calculation on collected data at the end of each run.

Step 5

a. Select the Reports tab to display the Reports page where you can specify your reporting requirements for this method.
b. Enter your name in the **Name** field.

c. Enter a comment relating to your experiment in the **Comment** field (*optional*).

d. Set up your report style by selecting the appropriate check boxes in the **Options** group.

   For example, select the **Auto Print** check box to obtain a printout of your report automatically. Select the **Parameters** check box to include your experimental parameters in the report. Select the **Graph** check box to include a calibration graph in the generated report.

**Note**

If **Auto Print** is selected, then the system will send the report information to the specified printer as well as the Report area. However if **Auto Print** is not selected, the report will only be sent to the Report area and can be viewed by selecting the **Report** command from the **View** menu.

Step 6

Select **OK** to clear the **Setup** dialog and start the recalculations. The results will be displayed in the Report area.

**Print the report**

Step 7

   a. From the **File** menu, select **Print**. The Windows **Print** dialog is displayed.

   b. Set up your printing parameters and select **OK** to print your report.

**See also:**

[How to generate a report from previously collected data](#)

**How to generate a report from previously stored data—Lifetimes**

Step 1

Select **Open**… from the **File** menu to display the Windows **Open** dialog.

Step 2

A list of previously stored files will appear. If this is not a list of batch files, select the down arrow at the right of the **Files of type** list box and select 'Batch' to list all the batch files.

Step 3

Select the Batch file you require then select **Open**.

The system will automatically retrieve the collected data into the Graphics area. The method that was used to collect the data will also be retrieved.

**Note**

You can also retrieve data from Methods (*.FMLTN), Data (*.FDLT), Report (*.FRLT), Spreadsheet ASCII (*.CSV), GRAMS (*.SPC) and Rich Text Format (*.RTF) file types.
Step 4

Select the **Recalculate** button to display the **Setup** dialog.

Step 5

Select the **Reports** tab and on the **Reports** page specify the information you would like to include in the report. Select **Auto Print** if you require the new report to print to the Reports area and to a printer.

Step 6

Click **OK** to display the results in the Report area (and send to the printer if **Auto Print** is selected).

---

**Note**

If you require a printout of only the current graphics in the Graphics area of a previously stored data file:

- Select the **Clear Report** option in the Commands menu to clear any text that may be displayed in the Report area.
- Press the **Print** button to obtain a printout of the graphics.

Alternatively tick only the **Graph** check box in the **Reports** page to obtain a printout of graphics only.

---

**See also:**

[How to use advanced reporting options](#)

---

**Maths window**

Use the **Maths** window to perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator.

To access the **Maths** window, select the **Calculator** command from the **Graph** menu, or select the **Calculator** button on the **Graphics** toolbar.

The **Maths** window is only available in the Kinetics, Lifetimes, Scan and Validate applications.

The **Maths** window comprises the following:

**Calculation display field**

View the current calculation, including the trace/s being used, the trace operation being carried out and any functions that have been applied to the trace.

**Hot Tip**

Click on the down arrow adjacent to this field to view the last ten unique calculations that have taken place. Use this list to keep track of previous manipulations and repeat calculations quickly by simply selecting and then applying the desired calculation.

**Selected Trace/Focused Trace**

Choosing either of these options selects the highlighted (red) trace in the current Graph box for manipulation. When choosing between these options, use this simple rule:

- Use the **Selected Trace** option when the selected calculation is only to be applied to one trace.
- Use the **Focused Trace** option when the same calculation is to be applied to a number of traces.
The following example illustrates the difference between these options.

To perform the same manipulation on several traces:

a. Highlight a trace by clicking on it with the mouse. The trace will become red.
b. Select **Selected Trace**, **Apply** and then = to plot the result of the calculation
c. Highlight the next trace and repeat step b.

OR

a. Highlight the first trace.
b. Select **Focused Trace**, **Apply** and then = to plot the result of the calculation;
c. Highlight the next trace and select the = button to perform the same calculation on the newly selected trace.

**Trace**
Use the Trace group options to select the operation to be performed on the traces, the filter size and the interval (where applicable).
Click **Apply** to insert the selected operation into the equation
Click **=** (equals) to display the result of the calculations.

**Operation**
Click on an operation to find out more about it

- Anisotropy
- Polarization
- Sum Function
- Mean
- Boxcar Averaging
- Moving Average
- Savitzky-Golay
- Spline
- Deriv 1
- Deriv 2
- Deriv 3
- Deriv 4

**Display Options**
Select whether the results of the operation performed on the trace will be viewed in a New Graph box or in the Selected Graph box. If you choose the Selected Graph option, you will be able to directly compare the results with existing traces.

**Functions**
This group contains number and function buttons that can be used as a standard calculator in addition to being applied to traces.

To use the functions:

1. select a trace or value
2. select either a second trace or number
3. select the equals (=) button to display the result

All the basic mathematical functions are available (addition, subtraction, multiplication and division), as well as logarithm (**Log**) and square root (**Sqrt**) functions. When used as a calculator, the results of equations appear in the Report area.

**Additional buttons**

- **C**
- **CE**
Click **CE** to clear the last entry entered in the calculation display field.

=  
Click **=** (equals) to start the operation shown in the calculation display field.

See also:
[Maths Home Page](#)
[Maths How To](#)

**Maths Operations**

The following operations can be selected on the **Maths** window. To access the **Maths** window, select the **Calculator** command from the **Graph** menu, or select the **Calculator** button on the **Graphics** toolbar.

**Anisotropy, Polarization and Sum Function**

The term Anisotropy refers to an unequal angular distribution about a point in space. Anisotropy also refers specifically to a simple expression for measuring the degree of polarization of the fluorescence. It is almost identical to the expression for polarization apart from having a factor of two in the denominator. Anisotropy is usually represented by the lowercase letter "r", while polarization by an uppercase "P".

The two expressions are listed below:

\[
Anisotropy \\
r = \frac{(I_{vv} - GI_{vh})}{(I_{vv} + 2GI_{vh})}
\]

\[
Polarisation \\
P = \frac{(I_{vv} - GI_{vh})}{(I_{vv} + GI_{vh})}
\]

where

- \(I_{vv}\) equals the intensity of fluorescence with excitation polarizer = vertical, emission polarizer = vertical
- \(I_{vh}\) equals the intensity of fluorescence with excitation polarizer = vertical, emission polarizer = horizontal
- \(I_{hv}\) equals the intensity of fluorescence with excitation polarizer = horizontal, emission polarizer = vertical
- \(I_{hh}\) equals the intensity of fluorescence with excitation polarizer = horizontal, emission polarizer = horizontal

\(G\) is a factor that accounts for the polarization bias of the instrument. In particular it accounts for the wavelength-dependent polarization bias of the emission side of the instrument (dominated by the polarization bias of the emission grating).

The \(G\) factor is given by:

\[
G = \frac{I_{hv}}{I_{hh}}
\]

where

- \(I_{hv}\) equals the intensity of fluorescence with excitation polarizer = horizontal, emission polarizer = vertical
- \(I_{hh}\) equals the intensity of fluorescence with excitation polarizer = horizontal, emission polarizer = horizontal

The expression for the anisotropy is a quotient. The denominator of the expression, as given by \(I_{vv} + 2GI_{vh}\) is the ‘sum’ function, and actually represents the total emission unbiased by any sample polarization effects.
**Mean**
The Mean operation allows the average of two or more traces to be taken and the resulting trace to be plotted. The mean of the traces can also be used in conjunction with other functions.

The equation used in mean calculations is:

\[
\frac{\text{scan 1} + \text{scan 2} + \ldots + \text{scan x}}{x}
\]

The calculation is performed over the common X range of the traces and the trace with the most dense data is used to determine the data points. The other traces are interpolated at the relevant data points.

**Boxcar Averaging**
This algorithm will take the number of points as defined by the Filter size, and average these, displaying the average at the middle X-point of the filter.

For example, for the following:

Data: \((x_1, y_1), (x_2, y_2), (x_3, y_3) \ldots (x_n, y_n)\)

Filter size: 5

The first smoothed data point will be \((x_3, \text{sum}(Y_1:y_5)/5)\), the next \((x_8, \text{sum}(y_6:y_{10})/5)\) \ldots \((x_{n-2}, \text{sum}(y_{n-4}:y_{n})/5)\).

If there are not enough points at the end of the data set to complete the filter size, then these points will be ignored, hence, the gap at the end may be larger than the gap in the beginning. There will be a gap before the first point and after the last point of the smoothed continuum, with respect to the original unsmoothed data. This algorithm is only valid for evenly x-spaced data points.

Within a time scan, (e.g., Advanced collect in kinetics) the data points may be unevenly spaced between different stages. Each stage will be treated as a new collect, hence, there may be large gaps between stages. If there are not enough points at the end of a stage to complete the filter size, then these points are ignored, hence, there may be large gaps at the end of a stage. The filter size will be limited to odd numbers, with the minimum of 3 to a maximum of 101.

**Moving Average**
This algorithm will take the number of points as defined by the Filter size, and average these, displaying the average at the middle X-point of the filter.

For example, for the following:

Data: \((x_1, y_1), (x_2, y_2), (x_3, y_3) \ldots (x_n, y_n)\)

Filter size: 5

The first smoothed data point will be \((x_3, \text{sum}(Y_1:y_5)/5)\), the next \((x_4, \text{sum}(y_2:y_6)/5)\) \ldots \((x_{n-2}, \text{sum}(y_{n-4}:y_{n})/5)\). There will be a gap before the first point and after the last point of the smoothed continuum, with respect to the original unsmoothed data. This algorithm is only valid for evenly x-spaced data points. Within a time scan, (e.g., Advanced collect in kinetics), the data points may be unevenly spaced between different stages. This will be treated by interpolation of data from the previous stage so as to give evenly spaced data points into the next stage. If there is not enough data for interpolation, then the next stage will be treated as a new collect, hence, there may be gaps and further delays in displaying the point. The filter size will be limited to odd numbers, with the minimum of 3 to a maximum
The Savitzky-Golay technique enables you to reduce the noise in the currently selected Maths scan by using smoothing.

When you select this operation you will be prompted to enter a Filter Size from 5 to 101. The larger the Filter Size, the more data points that will be smoothed.

Q. I cannot enter a particular value; it keeps changing to another number — why?  
A. The Savitzky-Golay technique requires an odd number to be entered, not an even number.

You will also need to enter an Interval. The Interval refers to how many of the averaged points will be plotted on the smoothed trace. The smaller the interval, the more detail there will be in the trace.

Consider selecting an Interval of 1. This would mean that when the smoothed continuum is plotted, every smoothed point will be displayed.

Q. Why can't I see the Filter Size and Interval fields?  
A. These will be displayed when you select Smooth in the Operation field.

The Deriv operations allow you to obtain derivatives of a selected scan. The derivative can be of the orders 1 to 4. The resulting trace is then plotted. The Savitzky-Golay technique is used for derivative calculations.

On selection of the Deriv operation you will be prompted to enter a Filter Size from 5 to 101. The Filter Size refers to the number of points around a particular point that will be averaged to produce a new derived Y value. The lower the Filter Size, the more data points that will be used in the derivative trace.

Q. I cannot enter a particular value; it keeps changing to another number — why?  
A. The Savitzky-Golay technique requires an odd number to be entered, not an even number.

You will also need to enter an Interval. The Interval refers to how many of the derived points will be plotted on the resulting trace. The smaller the Interval, the more detail there will be in the trace.

Q. Why can't I see the Filter Size and Interval fields?  
A. These will be displayed when you select one of the ‘Derivs’ in the Operation field.

See also:  
About smoothing

Displaying data graphically—Lifetimes

- Graphics area
- Graphics How To
- Graph menu
- Graphics toolbar
- Graph Labels
- Add Label dialog
Graphics area—Lifetimes

The Graphics area of the Lifetimes application appears in the top half of the Lifetimes Application window and is used to display traces of collected data. In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area surrounding the graph is referred to as the graph box. A graph will appear in the Graphics area when you select Start to commence a Lifetimes run.

To display or hide the Graphics area, select the Graphics command on the View menu. Double-clicking on the Graphics area enables you to toggle between half or full size display sizes.

You can access the functions associated with the Graphics area by doing the following:

- selecting one of the Graphics toolbar buttons;
- selecting a command from the Graph menu; or
- clicking the right mouse button in the Graphics area and select a command from the menu

Note
Not all functions are available on every menu.
You can customize the graphics area to suit your requirements in the following ways:

Note
All of the commands and dialogs listed are accessible from the Graph menu.

<table>
<thead>
<tr>
<th>To…</th>
<th>Use the…</th>
<th>Tell me how…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Change the axes scaling automatically</td>
<td>Autoscale graph command</td>
<td>How to scale your graph automatically</td>
</tr>
<tr>
<td>Change the axes scaling manually</td>
<td>Axes Scales dialog</td>
<td>How to scale your graph manually</td>
</tr>
<tr>
<td>Modify the graph appearance (axes, trace)</td>
<td>Graph Preferences dialog</td>
<td>How to alter the look of your graphs</td>
</tr>
<tr>
<td>Add a new graph to the display</td>
<td>Add graph command</td>
<td></td>
</tr>
<tr>
<td>Remove a graph from the display</td>
<td>Remove graph command</td>
<td></td>
</tr>
<tr>
<td>View single or multiple graphs</td>
<td>Single/multiple graph command</td>
<td></td>
</tr>
<tr>
<td>Resize a graph box</td>
<td>Cursor</td>
<td>How to resize graph boxes</td>
</tr>
<tr>
<td>Track the focused trace</td>
<td>Cursor Mode dialog</td>
<td>How to track the focused trace</td>
</tr>
<tr>
<td>Select the focused trace</td>
<td>Cursor</td>
<td>How to select the focused trace</td>
</tr>
<tr>
<td>Display a trace in the graph</td>
<td>Trace Preferences dialog</td>
<td>How to display a trace that is not appearing in a graph box</td>
</tr>
<tr>
<td>Remove a trace from a graph</td>
<td>Trace Preferences dialog</td>
<td>How to remove a trace from a graph</td>
</tr>
<tr>
<td>Remove a trace from the trace list</td>
<td>Trace Preferences dialog</td>
<td>How to remove a trace from the trace List</td>
</tr>
<tr>
<td>Zoom in and out on the graph</td>
<td>Zoom In and Zoom Out commands</td>
<td>How to zoom a graph</td>
</tr>
<tr>
<td>Add a label to the graph box</td>
<td>Add Label dialog</td>
<td>How to add a label to a graph</td>
</tr>
<tr>
<td>Edit labels in the graph box</td>
<td>Edit Label dialog</td>
<td>How to edit a label</td>
</tr>
<tr>
<td>Moving labels in the graph box</td>
<td>Cursor</td>
<td>How to move a label</td>
</tr>
<tr>
<td>Delete labels from the graph box</td>
<td>Delete Annotation command</td>
<td>How to delete a label</td>
</tr>
<tr>
<td>Add a picture to the graph box</td>
<td>Add Picture dialog</td>
<td>How to add a picture to a graph</td>
</tr>
<tr>
<td>Move pictures in the graph box</td>
<td>Cursor</td>
<td>How to move a picture</td>
</tr>
<tr>
<td>Replace pictures in the graph box</td>
<td>Edit Picture dialog</td>
<td>How to replace a picture</td>
</tr>
<tr>
<td>Delete pictures from the graph box</td>
<td>Delete annotation command</td>
<td>How to delete a picture</td>
</tr>
<tr>
<td>Save the Graphics area you have set up as a graphics template</td>
<td>Save As dialog (from the File menu)</td>
<td>How to create a graphics template</td>
</tr>
</tbody>
</table>
Note
The red trace in any graph box is the one on which all information appears and the one used for the cursor tracking. It is called the focused trace.

See also: Graphics toolbar Graphics How To

Graphics How To—Lifetimes

Traces

- How to track the focused trace
- How to select the focused trace
- How to display a trace that is not appearing in a graph box
- How to remove a trace from a graph
- How to remove a trace from the trace List

Sizing and scale

- How to resize graph boxes
- How to scale your graph manually
- How to scale your graph automatically
- How to alter the look of your graphs
- How to zoom a graph

Labels

- How to add a label to a graph
- How to edit a label
- How to delete a label
- How to move a label

Pictures

- How to add a picture to a graph
- How to move a picture
- How to delete a picture
- How to replace a picture

Graph menu—Lifetimes

Introduction

Use the Graph menu to view data in various graphical formats or in multiple graphs in the Graphics area. You can double-click on the Graphics area to toggle the display through graph and report to graph only.

In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area surrounding the graph is referred to as the graph box. A graph will appear in the Graphics area when you select Start to commence a Lifetimes run.

The Graph menu can be accessed in the following ways:
• from the main menu at the top of the Lifetimes Application window
• by clicking the right mouse button in a graph box or the Graphics area

**Note**
The red trace in any graph box is the one on which all information appears and the one used for the cursor tracking. It is called the focused trace.

### Menu commands

The following commands can be accessed from the Graph menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>Button</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Print XY point</td>
<td></td>
<td>Select this to print the cursor's current X and Y coordinates in the report area.</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Note</strong> This option is only available from the Graph menu that appears when you right-click in a graph box.</td>
</tr>
<tr>
<td>Copy Graph</td>
<td></td>
<td>Select <strong>Copy Graph</strong> to copy the entire Graphics area to the Windows Clipboard. All of the graphs in the Graphics area will be copied to the Clipboard.</td>
</tr>
<tr>
<td>Paste to Graph</td>
<td></td>
<td>Select <strong>Paste to Graph</strong> to paste the contents of the Windows Clipboard into the Graphics area. Text is pasted as a label, graphics are pasted as pictures.</td>
</tr>
<tr>
<td>Add Graph</td>
<td></td>
<td>Select <strong>Add Graph</strong> to add a new graph box to the Graphics area. You may want to do this if you have, for example, a graph containing existing data is displayed and you want to collect some new data in a new graph.</td>
</tr>
<tr>
<td>Remove Graph</td>
<td></td>
<td>Select <strong>Remove Graph</strong> to delete the selected graph box from the Graphics area. The graph box is removed, but the data is not deleted and is still available from the Trace Preferences window. You may want to use remove graph if you have a large number of graph boxes open but only want to focus on some of these.</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Note</strong> You can remove multiple graph boxes by first holding down the CTRL key and selecting the graphs to be removed and then selecting the Remove Graph button or menu item.</td>
</tr>
<tr>
<td>Clear All Traces</td>
<td></td>
<td>Select <strong>Clear All Traces</strong> to delete all traces from the graph/s. Note that selecting this option simply turns off the Visible property for the traces in the Trace Preferences window. You can redisplay the traces by making the traces Visible again in the Trace Preferences window.</td>
</tr>
<tr>
<td>Add Label</td>
<td></td>
<td>Select <strong>Add Label</strong> to display the Add Label dialog where you can enter a text label to be placed on a graph or in the background of the Graphics area.</td>
</tr>
<tr>
<td>Add Picture</td>
<td></td>
<td>Select <strong>Add Picture</strong> to display the Add Picture dialog where you can select a picture, bitmap or chemical structure to be placed on a graph or in the background of the Graphics area.</td>
</tr>
<tr>
<td>Edit Annotation</td>
<td></td>
<td>Select <strong>Edit Annotation</strong> to alter the content or look of the selected annotation, (either a label or a picture) in the Graphics area. This option can only be activated if an annotation is selected.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>If the selected annotation is a label, selecting Edit Annotation will open the <strong>Edit Label</strong> dialog where you can change the text of the selected label. If the selected annotation is a label, selecting Edit Annotation will open the <strong>Edit</strong>...</td>
</tr>
<tr>
<td><strong>Picture</strong> dialog where you can replace the selected picture with a new picture.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>---</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Delete Annotation</strong></td>
<td>Select <strong>Delete Annotation</strong> to remove an annotation, (such as a label or a picture) from the Graphics area. This option can only be activated if an annotation is selected. You can use this menu option to remove highlighted annotations in the Graphics area such as labels, pictures and ruler/fit information.</td>
<td></td>
</tr>
<tr>
<td><strong>Trace Preferences</strong></td>
<td>Select this command to display the <strong>Trace Preferences</strong> window where you can select the trace(s) to display in the selected graph.</td>
<td></td>
</tr>
<tr>
<td><strong>Graph Preferences</strong></td>
<td>Select this command to display the <strong>Graph Preferences</strong> dialog where you can change the appearance of your graph. Any changes you make to the appearance of the graph will be applied to all graphs.</td>
<td></td>
</tr>
<tr>
<td><strong>Cursor Mode...</strong></td>
<td>Select this command to display the <strong>Cursor Modes</strong> dialog where you can choose the type of cursor to be used on screen. For example, you can select to use a free cursor that can move anywhere in the graph or exactly trace the focused scan (red scan). Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the Least Squares method or the Point to Point method.</td>
<td></td>
</tr>
<tr>
<td><strong>Axes Scales...</strong></td>
<td>Select this command to display the <strong>Axes Scales</strong> dialog where you can set the scale range for the X and Y axes.</td>
<td></td>
</tr>
</tbody>
</table>
| **Auto scale graph** | To automatically scale the graph, point to one of the following options:  

**Autoscale[XY]**  
Select Autoscale[XY] to automatically scale collected data by scaling the ordinate (Y) height and abscissa (X) width to full screen.  

**Autoscale[X]**  
Select Autoscale[X] to automatically scale collected data by scaling the abscissa (X) width to full screen.  

**Autoscale[Y]**  
Select Autoscale[Y] to automatically scale collected data by scaling the ordinate (Y) height to full screen. |
| **Zoom Out** | Select **Zoom Out** to zoom back to the original X and Y ranges which were used to collect the data (i.e. the method setup conditions of the focused scan). For example, the system will revert back to the X and Y ranges set in the method if you zoom in and out during a data collection.  

**Tell me How to zoom a graph** |
| **Single/Multi Graph** | Select **Single/Multi Graph** to toggle between displaying one or all graphs.  

**Note**  
The graph highlighted in the Multi Graph view will be displayed when you toggle the Single Graph option. |
| **Auto Arrange Graphs** | Select **Auto Arrange Graphs** to view and arrange all graphs simultaneously.  

**Note**  
You can move graph boxes manually by clicking the left mouse button outside the graph axes and dragging the graph box to a new position. You can also resize graphs. |
| **User Data Form** | Select **User Data Form** to display the **User Data Form** where you can enter extra information about the traces in the highlighted graph box. For example, you might |
want to record information about the experimental conditions you used to collect the data, such as the pH at which data was collected.

### Calculator

Select **Calculator** to display the **Maths** window where you can perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator.

### History List

This displays a list of the current graphs.

---

**Graphics Toolbar—Lifetimes**

Use the Graphics toolbar buttons to gain quick access to many of the Graph menu options. You can also use the toolbar to select the graph you want to view and to change the look of your graphs.

To display or hide the toolbar, point to the Toolbars option in the View menu and select Graphics. To display or hide the toolbar's text labels, point to the Toolbars option in the View menu and select Text Labels.

**Hot Tip**

To see the name of a button, point to it with the mouse.

The following commands can be accessed from the Graphics Toolbar:

<table>
<thead>
<tr>
<th>Button</th>
<th>Menun command</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image" alt="Trace Preferences" /></td>
<td>Trace Preferences</td>
<td>Select this button to display the <strong>Trace Preferences</strong> window where you can select the trace(s) to display in the selected graph.</td>
</tr>
<tr>
<td><img src="image" alt="Graph Preferences" /></td>
<td>Graph Preferences</td>
<td>Select this button to display the <strong>Graph Preferences</strong> dialog where you can change the appearance of your graph. Any changes you make to the appearance of the graph will be applied to all graphs.</td>
</tr>
<tr>
<td><img src="image" alt="Cursor Mode" /></td>
<td>Cursor Mode</td>
<td>Select this button to display the <strong>Cursor Modes</strong> dialog where you can choose the type of cursor to be used screen. For example, you can select to use a free cursor that can move anywhere in the graph or exactly trace the focused scan (red scan). Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the Least Squares method or the Point to Point method.</td>
</tr>
<tr>
<td><img src="image" alt="Axes Scales..." /></td>
<td>Axes Scales...</td>
<td>Select this button to display the <strong>Axes Scales</strong> dialog where you can set the scale range for the X and Y axes.</td>
</tr>
</tbody>
</table>
Autoscale[XY]  
Select the Autoscale[XY] button to automatically scale collected data by scaling the ordinate (Y) height and abscissa (X) width to full screen.

Autoscale[X]  
Select the Autoscale[X] button to automatically scale collected data by scaling the abscissa (X) width to full screen.

Autoscale[Y]  
Select the Autoscale[Y] button to automatically scale collected data by scaling the ordinate (Y) height to full screen.

Zoom Out  
Select the Zoom Out button to Zoom back to the original X and Y ranges which were used to collect the data (i.e. the method setup conditions of the focused scan). For example, the system will revert back to the X and Y ranges set in the method if you zoom in and out during a data collection.

Add Label  
Select this button to display the Add Label dialog where you can enter a text label to be placed on a graph or in the background of the Graphics area.

Add Picture  
Select this button to display the Add Picture dialog where you can select a picture, bitmap or chemical structure to be placed on a graph or in the background of the Graphics area.

Single/Multi Graph  
Select Single/Multi Graph to toggle between displaying one or all graphs.

Note  
The graph highlighted in the Multi Graph view will be displayed when you toggle the Single Graph option.

Auto Arrange Graphs  
Select the Auto Arrange Graphs button to view and arrange all graphs simultaneously.

You can move graph boxes manually by clicking the left mouse button outside the graph axes and dragging the graph box to a new position. You can also resize graphs.

Add Graph  
Select this button to add a new graph box to the Graphics area. You may want to do this if you have, for example, a graph containing existing data is displayed and you want to collect some new data in a new graph.

Remove Graph  
Select this button to delete the selected graph box from the Graphics area. The graph box is removed, but the data is not deleted and is still available from the Trace Preferences dialog.

You may want to use remove graph if you have a large number of graph boxes open but only want to focus on some of these.

Note  
You can remove multiple graph boxes by first holding down the CTRL key and selecting the graphs to be removed and then selecting the Remove Graph button or menu item.

Clear All Graphs  
Select this button to delete all traces from the graph/s. Note that selecting this option simply turns off the Visible property for the traces in the Trace Preferences window. You can redisplay the traces by making the traces Visible again in the Trace Preferences window.

User Data Form  
Select this button to display the User Data Form where you can enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data, such as the pH at which data was collected.

Calculator  
Select this button to display the Maths window where you can perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator.
Graph Labels—Lifetimes

- Add Label dialog
- Edit Label dialog
- How to add a label to a graph
- How to edit a label
- How to move a label
- How to resize a label
- How to delete a label

Add Label—Lifetimes

Use the **Add Label** dialog to place a label on your graph or into the background of the Graphics area.

To add a label to a graph box, you must have at least one trace Visible in the graph box. (You can select which traces you want to make visible in the **Trace Preferences** dialog.) If you are viewing multiple graphs, the label will be placed in the graph box that is currently selected. However, you can add a label outside of a graph box in the background of the Graphics area at any time.

**Q.** Can I add a label that will print with all my graphs.
**A.** Yes, you can add your label to the background of the Graphics area. To do this, right-click on a section of the background (gray area) and select **Add Label** from the menu. (You will notice that this menu has less options than the menu that appears when you right-click on a graph.) In the **Add Label** dialog, enter the label to be added to all graphs and select OK. The label will appear in the background of the Graphics area.

To access the **Add Label** dialog, select the **Add Label** command from the **Graph** menu, or select the **Add Label** button on the Graphics toolbar.

The **Add Label** dialog comprises the following:

<table>
<thead>
<tr>
<th>To selected trace</th>
<th>Select this option to associate a label with the selected trace. When you associate a label with a trace and make the trace visible in the Graph box, the label will also be displayed with the trace.</th>
</tr>
</thead>
<tbody>
<tr>
<td>To selected graph</td>
<td>Select this option to associate the label with the selected graph. When you associate a label with a graph, the label will always be displayed in the Graph box, regardless of what traces are visible.</td>
</tr>
</tbody>
</table>
**Selected trace**  This will identify the trace that is currently selected and display the name of the trace.

**Font…**  Click **Font** to open the Windows Font dialog. In this dialog you can specify the font type, style and size to be used for the label.

**Label**  Use the **Label** field to enter the text for the label. You can enter as many lines of text as you require.

**Hot Tip**
If you wish to include certain XY co-ordinates on the graph as a label, position your cursor at the appropriate place, right-click and select **Add Label** from the menu. When the Add Label dialog appears, the co-ordinate positions will be added to the **Label** field.

**Note**
When you save the data (as a Batch or Data file) the label is saved with its associated trace or graph. Labels outside graph boxes can be saved in a graphics template. To do this, choose **Graph Template** as the **Save As type** in the **Save As** dialog.

**See also:**
- Trace Preferences
- Graph menu
- Graphics toolbar
- How to add a label to a graph
- How to move a label
- How to edit a label
- How to resize a label
- How to delete a label

**Edit Label—Lifetimes**

Use the **Edit Label** dialog to alter the content or look of your label.

To access the **Edit Label** dialog, right-click on the label you wish to change and select **Edit Annotation** from the menu.

The dialog comprises the following:

**Font**  Select **Font** to open the **Font** dialog, where you can specify the font type, style and size to be used for the label.

**Label**  Use the **Label** field to alter the text for the label.

**See also:**
- How to edit a label
- How to add a label to a graph
- How to move a label

**How to add a label to a graph—Lifetimes**

You can add a label to your graph or Graphics area background. To do this:

**Step 1**

Click on the graph box you require or click outside the graph box(es) to choose the Graphics area background.

**Note**
There must be at least one trace Visible in the graph box before you can add a label to it.
Step 2

Click the Add Label button on the toolbar or right-click on the graph and select Add Label from the menu. The Add Label dialog will appear. The Add Label command is also accessible from the Graph menu.

Step 3

Click the Font button. The Windows Font dialog appears. Select the font type, size and style you require for your label. A sample of the font style appears in the Sample field. When you are satisfied with your selections, select OK to return to the Add Label dialog.

Step 4

Enter the label in the Label field and select OK. The label will be displayed on the selected graph. If necessary, you can move a label so that it is more appropriately placed on your graph.

See also:
How to move a label
How to edit a label
How to resize a label

How to edit a label—Lifetimes

Step 1

Place the cursor on the label and click the left mouse button to select the label.

Hot Tip
If you are having trouble selecting the label it may be that a trace is interfering with the label selection. Try zooming in and clicking in a different part of the label.

Step 2*

Click the right mouse button and select Edit Annotation from the drop-down menu. Alternatively you can select the Edit Annotation command from the Graph menu. The Edit Label dialog is displayed.

Step 3

Use the Edit Label dialog to change the label.

For example, if you want to change the font size of the label, select the Font button. In the Size field, scroll through the available sizes for the font and select the size you require. Alternatively, highlight the currently selected size in the Size entry field and enter the size you require. Click the OK button in this dialog to return to the Edit Label dialog.

Step 4

Click OK to clear the Edit Label dialog. Your label will appear with the new font size.

See also:
How to add a label to a graph
How to move a label—Lifetimes

To move a label so that it is more appropriately placed on your graph or Graphics area background, do the following:

Step 1

Place the cursor on the label.

Step 2

Click and hold down the left mouse button.

**Hot Tip**
If you are having trouble selecting the label, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 3

Drag the label to the correct position and release the mouse button. The label will be displayed at the new position.

**Note**
Labels are transparent so as not to interfere with any traces on graphs.

See also:
- How to add a label to a graph
- How to edit a label
- How to resize a label
- How to delete a label

How to resize a label—Lifetimes

To resize a label so that it is more appropriately sized on your graph or Graphics area background, do the following:

Step 1

Select the label by clicking on it with the mouse. A border will be displayed around the label to indicate it is selected.

**Hot Tip**
If you are having trouble selecting the label, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Position the cursor over one of the corner edges of the label until the cursor becomes a diagonal double-headed arrow. Click and hold down the left mouse button.
Example of label showing the border that indicates it is selected and the arrow to be used for re-sizing

Step 3

Increase or decrease the size of the label by dragging the mouse. The label text will also be resized. Release the mouse button when the label and text are the correct size.

**Note**
Labels are transparent so as not to interfere with any traces on graphs.

See also:
[How to add a label to a graph](#)
[How to edit a label](#)
[How to delete a label](#)

**How to delete a label—Lifetimes**

To delete a label from a graph or Graphics area background, do the following:

Step 1

Position the cursor on the label and click the left mouse button to select it.

**Hot Tip**
If you are having trouble selecting the label it may be that a trace is interfering with the label selection. Try zooming in and clicking in a different part of the label.

Step 2

Click with the right mouse button and select **Delete Annotation** from the drop-down menu displayed. Alternatively, you can select the **Delete Annotation** command from the **Graph** menu. The label will be deleted from the graph or Graphics area.

See also:
[How to add a label to a graph](#)
[How to move a label](#)
[How to edit a label](#)
[How to resize a label](#)

**Graph Pictures—Lifetimes**

- [Add Picture dialog](#)
- [Edit Picture dialog](#)
- [How to add a picture](#)
- [How to replace a picture](#)
- [How to move a picture](#)
- [How to resize a picture](#)
- [How to delete a picture](#)
Add Picture—Lifetimes

Use the Add Picture dialog to insert a picture or bitmap into the Graphics area. The picture can be added as a background picture or to a particular graph box.

To add a picture to a graph box you must have at least one trace Visible in the graph box. (You can select which traces you want to make visible in the Trace Preferences dialog.) If you are viewing multiple graphs, the picture will be placed in the graph box that is currently selected. However, you can add a picture outside of a graph box in the Graphics area at any time.

Q. Can I add a picture that will print with all my graphs.
A. Yes, you can add your picture to the background of the Graphics area. To do this, right-click on a section of the background and select Add Picture from the menu. (You will notice that this menu has less options than the menu that appears when you right-click on a graph.) In the Add Picture dialog, load the picture to be added to all graphs and select OK. The picture will appear in the background of the Graphics area.

To access the Add Picture dialog, select the Add Picture command from the Graph menu, or select the Add Picture button on the Graphics toolbar.

The Add Picture dialog comprises the following commands:

- **To selected trace**: Select this option to associate a picture with the selected trace. When you associate a picture with a trace and make the trace visible in the Graph box, the label will also be displayed with the trace.
- **To selected graph**: Select this option to associate the picture with the selected graph. When you associate a picture with a graph, the label will always be displayed in the Graph box, regardless of what traces are visible.
- **Load**: Click Load to open the Windows Open dialog. Use this dialog to browse your computer for the picture you want to insert. The picture must be one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.

If you are viewing multiple graphs the picture will be placed on the graph that is currently selected. If no graph is selected, the picture will be placed on the Graphics area background.

**Note**
When you save the data (as a Batch or Data file) the label is saved with its associated trace or graph. Labels outside graph boxes can be saved in a graphics template. To do this, choose Graph Template as the Save As type in the Save As dialog.

See also:
- Trace Preferences
- Graph menu
- Graphics toolbar
- How to add a picture to a graph
- How to move a picture
- How to delete a picture
- How to resize a picture
- How to replace a picture

Edit Picture—Lifetimes
Use the **Edit Picture** dialog to change a picture you have inserted into the Graphics area.

To access the **Edit Picture** dialog, right-click on the picture you wish to change and select **Edit Annotation** from the menu.

The **Edit Picture** dialog comprises the following commands:

- **To selected trace/graph** Displays whether you originally selected to add the picture to a trace or a graph. This option is not editable.
- **Load** Click **Load** to open the Windows **Open** dialog. Use this dialog to browse your computer for the picture you want to insert. The selected picture will replace the current picture. The picture must be one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.

See also:
- How to replace a picture
- How to add a picture
- How to move a picture
- How to delete a picture

**How to add a picture to a graph—Lifetimes**

You can add a picture to your graph or the Graphics area background. To do this:

**Step 1**

Click on the graph box you require or click outside the graph box(es) to choose the Graphics area background.

**Note**

There must be at least one trace Visible in the graph box before you can add a picture to it.

**Step 2**

Click the **Add Picture** button on the toolbar or right-click on the graph and select **Add Picture** from the menu. The **Add Picture** dialog will appear. The **Add Picture** command is also accessible from the **Graph** menu.

**Step 3**

Click the **Load** button. The Windows **Open** dialog appears.

**Step 4**

Browse through your directories to find the picture you want to insert. All pictures must be in the bitmap format and will have the extension .BMP.

**Step 5**

Highlight the .BMP that you want to insert and select **Open**.

**Step 6**

The **Add Picture** dialog will appear showing a preview of the bitmap. If the bitmap is the correct one select the
OK button. The bitmap will be placed onto the graph.

Note
You should take care not to add a bitmap that is too big as it will obscure part or all of your graph.

You can move a picture so that it is more appropriately placed on your graph.

See also:
How to move a picture to a graph
How to delete a picture
How to replace a picture
How to resize a picture

How to replace a picture—Lifetimes

To change a picture that you have added to a graph or Graphics area background do the following:

Step 1
Move the cursor to the picture and select the left mouse button to select it.

Hot Tip
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2
Click the right mouse button and select Edit Annotation from the drop-down menu or select Edit Annotation from the Graph menu. The Edit Picture dialog is displayed.

Step 3
From the Edit Picture dialog, select the Load button. The Windows Open dialog is displayed.

Step 4
Browse through your directories to find the picture to replace the current bitmap. Picture files must be in one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.

Step 5
Highlight the file that you want to insert and select Open to return to the Edit Picture dialog.

Step 6
The Edit Picture dialog will display a preview of the bitmap. If the bitmap is the correct one select the OK button. The bitmap will be placed onto the graph. If you need to, move the picture so that it is more appropriately placed on your graph.

Note
Do not to add a bitmap that is too big as it will obscure part or all of your graph.
See also:
How to add a picture to a graph
How to move a picture
How to delete a picture
How to resize a picture

How to move a picture—Lifetimes

To move a picture so that it is more appropriately placed on your graph or Graphics area background, do the following:

Step 1

Place the cursor on the picture.

Step 2

Click and hold down the left mouse button.

Hot Tip

If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 3

Drag the picture to the correct position and release the mouse button. The picture will be displayed at the new position.

See also:
How to add a picture to a graph
How to delete a picture
How to replace a picture
How to resize a picture

How to resize a picture—Lifetimes

To resize a picture so that it is more appropriately sized on your graph or Graphics area background, do the following:

Step 1

Select the picture by clicking on it with the mouse. A border will be displayed around the picture to indicate it is selected.

Hot Tip

If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Position the cursor over one of the corner edges of the picture until the cursor becomes a diagonal double-headed arrow. Click and hold down the left mouse button.
Example of picture showing the border that indicates it is selected and the arrow to be used for re-sizing

Step 3

Increase or decrease the size of the picture by dragging the mouse. The picture will also be resized. Release the mouse button when the picture is the correct size.

See also:
How to add a picture to a graph
How to move a picture to a graph
How to delete a picture
How to replace a picture
How to resize a picture

How to delete a picture—Lifetimes

To delete a picture from a graph or Graphics area background, do the following:

Step 1

Position the cursor on the picture and click the left mouse button to select it.

Hot Tip
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Click with the right mouse button and select Delete Annotation from the drop-down menu displayed. Alternatively, you can select the Delete Annotation command from the Graph menu. The picture will be deleted from the graph or Graphics area.

See also:
How to add a picture to a graph
How to move a picture
How to zoom a graph
How to resize a picture

Trace Preferences—Lifetimes

A single graph box can display many traces. This window enables you to select the trace(s) to display (make Visible) in the selected graph box.

To access the Trace Preferences dialog, select the Trace Preferences command from the Graph menu,
The dialog comprises the following:

**Hot Tip**
The Trace Preferences menu is also available on clicking the right mouse button anywhere in the trace listing.

### Trace Preferences table

#### Visible
Use the Visible field to select which traces will appear on the selected graph. To select or clear the Visible field, simply click the left mouse button in this column. A tick will appear next to traces that are to be displayed in the selected graph.

If you are in single graph view, all selected traces will appear on the one graph. If you are in Multi Graph view the traces will appear on the graph that is currently selected.

**Hot Tip**
To make all traces Visible, right-click on the table and select the **Select All** command from the menu. Then right-click on the table again and select the **Show Traces** command from the menu.

#### Color
The Color field enables you to view the color of each trace that appears on the selected graph. To change the color of a trace, highlight it, click the right mouse button in the Trace Preferences table and select Change Trace Color. You can then select a color from the Color dialog that is displayed.

The red trace in any graph box is the one on which all information appears. This trace, which is used for the cursor tracking, is called the focused trace and it is always red. When you select a trace in the Color column it will become the focused trace in the selected graph box.

**Note**
When you change the trace line width in the **Graph Preferences** dialog it also will change the width of the color trace line in this dialog.

#### Name of trace
The Name of Trace field contains a description of the traces currently available for display in the selected graph box.

#### Filename
This field displays the filename and path for the trace. If the name is too long to appear in the field you can use the keyboard arrow keys to scroll to the end of the filename or you can resize the Trace Preferences window. If the file path for a trace shows 'not saved' then the trace is unsaved data and is only stored in memory.

### Trace Audit Log

The Trace Audit Log displays information about the highlighted trace. This includes instrument parameters and data form information relating to the trace that has been selected. It also logs all information about data manipulation. When more than one trace is highlighted, the Trace Audit Log will also display information about the trace that was last selected.

**See also:**
- [Graph menu](#)
- [Graphics toolbar](#)
Graph Preferences
Trace Preferences menu
How to remove a trace from the trace list
How to remove a trace from a graph

Trace Preferences menu—Lifetimes

The Trace Preferences menu is displayed when you click the right mouse anywhere on the Trace Preferences table. The menu contains the following commands:

**Select All** This selects or highlights all the traces in the listed on the Trace Preferences page.
**Select None** This de-selects all the traces in the listing.
**Show Traces** Select Show Traces to make all of the traces currently highlighted Visible, meaning that they will be displayed in the current graph box.
**Hide Traces** Select Hide Traces to remove the Visible property from all the traces currently highlighted, which will remove them from the current graph box.
**Delete Traces** Select Delete Traces to delete all the highlighted traces in all graphs. Note that if you save the data file before you select Delete Traces, the traces are not deleted from the saved file, and they can be retrieved at a later date. Selecting Delete Traces will delete the traces from the application memory, and it is recommended that you select this option if you need to free up memory on your computer.

**Note**
If the application is controlled by the GLP Administration application, you will be prompted to save an unsaved data file before it is removed from the application.

**Change Trace Color** Select Change Trace Color to display the Windows Color dialog where you can change the color of the currently highlighted trace(s). The focused trace will remain red while it retains the focus, and will revert to the selected color when the focus is moved.

How to select the focused trace—Lifetimes

To select the scan that you want to be the focused (red) trace, do one the following:

- From the Graph, click the left mouse button on the trace that you want to make the focused trace.
- From the Graph Legend (if displayed), click the left mouse button on the trace that you want to make the focused trace.
- From the Trace Preferences dialog, click in the Color column of the trace that you want to make the focused trace.

**Note**
Select the Show Legend check box in the Graph Preferences dialog to display the Graph Legend.

See also:
- How to track the focused trace
- How to view a particular trace
How to display a trace that is not appearing in the graph box—Lifetimes

Step 1

Click on the graph in the Graphics area.

Step 2

Select the Trace Preferences button [Trace Preferences] from the Graphics toolbar, or select Trace Preferences from the Graph menu. The Trace Preferences dialog is displayed.

Step 3

Find that trace that you wish to see and select the Visible option. A tick will appear in the Visible column to indicate that it will now be displayed.

If you wish to make multiple traces visible, do the following:

a. Hold down the CTRL key select all the traces that you want to display.

b. Click the right mouse button and select the Show Traces command from the drop-down menu. Ticks will appear next to the selected traces to indicate that they will now be displayed.

Step 4

Press OK to clear the Trace Preferences dialog. The selected trace(s) will be displayed.

See also:
How to remove a trace from a graph
How to remove a trace from the trace list

How to remove a trace from a graph—Lifetimes

Step 1

Select a graph in the Graphics area.

Step 2

Click the Trace Preferences button [Trace Preferences] on the Graphics toolbar or select Trace Preferences from the Graph menu. The Trace Preferences dialog is displayed.

Step 3

Find that trace that you no longer wish to see and select the Visible option. The tick will be removed from the Visible column to indicate that it will not be displayed.

If you wish to make multiple traces visible, do the following:

a. Hold down the CTRL key select all the traces that you no longer want to display.
b. Click the right mouse button and select the **Hide Traces** command from the drop-down menu. The ticks will be removed from the selected traces to indicate that they will not be displayed.

**Step 4**

Select **OK** to clear the **Trace Preferences** dialog. The trace will be removed from the graph.

See also:
- [How to display a trace that is not appearing in the graph box](#)
- [How to remove a trace from the trace list](#)

**How to remove a trace from the trace list—Lifetimes**

If your graphs contain unwanted traces, you may wish to delete them (rather than simply making them invisible) in order to free up application memory and aid system performance.

**Step 1**

Select a graph in the Graphics area.

**Step 2**

Click the **Trace Preferences** button on the Graphics toolbar or select **Trace Preferences** from the **Graph** menu. The **Trace Preferences** dialog is displayed.

**Step 3**

Select the trace to be removed by clicking on it.

**Step 4**

Click the right mouse button to display the **Trace Preferences** menu.

**Step 5**

Select **Delete Traces** to delete the sample from the trace list. You will be prompted to confirm this action.

**Step 6**

Select **OK** to clear the **Trace Preferences** dialog. The trace will be removed from all graphs.

See also:
- [How to remove a trace from a graph](#)
- [How to display a trace that is not appearing in the graph box](#)

**Graph Preferences—Lifetimes**

See also:
- [Axes Preferences](#)
- [Advanced Options](#)
- [Trace Style](#)
- [Show Legend](#)

**Introduction**

Use the **Graph Preferences** dialog to change the appearance of your graph.
All graphs will appear labeled with the X axis label being horizontal and the Y axis vertical. You cannot change the orientation of these labels. However you can alter the font and size of the axes and their labels.

Any changes you make to the appearance of the graph will apply to all graphs until the settings are changed again.

To access the Graph Preferences dialog, select the Graph Preferences command from the Graph menu, or select the Graph Preferences button on the Graphics toolbar.

Dialog controls

The Graph Preferences dialog comprises the following:

Axes Preferences

Axes Color  Select the color that you want to assign to the axes of the graph/s from the drop down list.
Axes Width  Select the width of the axes from the drop down list. The width, which is in pixels, ranges between one and five pixels.
Grid Style  Select the style of grid for the graph from the drop down list. If you do not wish to include a grid on the graph, select None.
Plot Area Color  Select the color to be used as the background of the graph from the drop down list. If you do not want a background color on the graph, select White.
Trace Width  Select the width of the trace from the drop down list. The width is given in pixels and ranges between one and five pixels.
Font  Select the Font button to display the Windows Font dialog where you can define the font style for the axes of the graph/s. The style and size of the font will be applied to all graph. The style, name and point size of the font appears on the font button.
Preview Box  Preview any changes you make to the axes preferences in the preview box. The changes shown in this box will not be made until you select OK.

Advanced Options

Auto tick marks  The Auto tick marks option enables you to approximately set how many tick marks (or points) will be included on the graph axes. The tick marks generated will be whole, rounded, even numbers. The Auto tick marks option is not available if you have selected Major ticks.

Y-axis  Use the Y-axis slider bar to increase or decrease the number of tick marks (or points) to be included on the Y axis.

X-axis  Use the X-axis slider bar to increase or decrease the number of tick marks (or points) to be included on the X axis.

Major ticks  Use the Major Ticks option to enable you to set exactly how many ticks (or points) will be added to the X and Y axes. The number of marks you specify will be divided by the axis range, which can result in odd numbers on the axis. Note that any changes are not reflected
in the graph bitmap at the top of the screen.

**Decimals**

Use the **Decimals** option to set the number of X and Y decimal places to be displayed on the axes. To set the number of decimal places, select the **Decimal** option and enter the number in the Y-axis and X-axis combo boxes. Note that any changes are not reflected in the graph bitmap at the top of the screen.

**Trace Style**

**Points**

Select **Points** to display each individual data point collected by the Cary Eclipse on the graph.

**Points and solid lines**

Select **Points and Solid Lines** to display each individual data point collected by the Cary Eclipse on the graph. Additionally, an interpolated line that connects the points will be drawn.

**Solid lines**

If you select **Solid Lines**, the Cary Eclipse system will interpolate the data between the actual, collected data points and show the data as a continuous line.

**Style cycles automatically**

If you select **Style Cycles Automatically**, the Cary Eclipse system will display each of the above three trace styles. This makes it easier to differentiate between traces, especially on black-and-white printers.

**Show Legend**

**Show Legend**

Select **Show Legend** to display a legend in the right side of each graph box. A single left click on a sample name or example line within the legend will make it the focused or red scan. Information about the trace will be displayed below the X axis.

**Hot Tip**

Double-clicking on the legend will automatically display the **Trace Preferences** window, enabling you to view more detailed information about your trace (e.g. audit log, File name, etc.).

See also:
- Graph menu
- Graphics toolbar
- Graphics area
- Graphics How To

**How to alter the look of your graphs—Lifetimes**

**Step 1**

Click the **Graph Preferences** button on the Graphics toolbar, or select **Graph Preferences** from the **Graph** menu. The **Graph Preferences** dialog is displayed.

**Step 2**

Use the available options available to choose your preferred appearance for the graphs. A preview box in the top right-hand corner will demonstrate how the graphs will appear.

**Step 3**
Select OK to clear the **Graph Preferences** dialog and update all graphs to reflect your selections.

**Note**
You can also add labels and pictures to your graphs to alter their appearance.

**Step 4**
To re-use your graphics setup at a later date, save the setup as a graphics template.

**See also:**
- [How to add a label to a graph](#)
- [How to add a picture to a graph](#)
- [How to move a label](#)
- [How to move a picture](#)
- [How to resize graph boxes](#)
- [How to make a graphics template](#)

**Cursor Modes—Lifetimes**

Use the **Cursor Modes** dialog to choose the type of cursor to be used screen. For example, you can select to use a free cursor that can move anywhere in the graph or exactly trace the focused scan (red scan). Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the Least Squares method or the Point to Point method.

To access the **Cursor Modes** dialog, select the **Cursor Mode** command from the **Graph** menu, or select the **Cursor Mode** button on the Graphics toolbar.

The dialog comprises the following:

**Cursor Modes**

**None**
Select this for no cursor tracking.

**Free**
The Free cursor is not confined to a particular trace on the graph. It can move anywhere in a graph box, and its ‘X,Y’ position is shown in the bottom right-hand side of the Graphics area. You can use the Free cursor to zoom in on a particular part of a graph.

**Note**
The Free cursor moves from pixel point to pixel point in the selected graph box. Hence you need to be careful when positioning the cursor over a scan to obtain an accurate (X,Y) reading. It is recommended that you use the Track cursor mode for this.

**Track**
Use the **Track** cursor to exactly trace the focused (red) scan in a graph box. The cursor position is displayed in the X and Y at the bottom right-hand side of the Graphics area.

**Hot Tip**
If you wish to fix the cursor on a particular position on the curve, move the cursor to the required position. Click the left mouse button, press ALT on the keyboard, then release the mouse button.

**Ruler-Least Squares**
This uses a least-squares technique to determine the slope and thus the reaction rate between the start and end points.
To track the focused (red) trace in the graph:

Step 1

Click the **Cursor Modes** button from the Graphics toolbar, or select **Cursor Mode** from the **Graph** menu. The **Cursor Modes** dialog will appear.

Step 2

Select **Track** and select the **OK** button.

Step 3

Place the blue tracking cursor in the graph box you require.

Step 4

Move the cursor left and right across the graph box to trace the focused (red) scan. The cursor coordinates change (in the X, Y field at the bottom left hand corner of the Graphics area) as you move along the curve.

**Hot Tip**

If you wish to fix the cursor on a particular position on the curve, move the cursor to the required position. Press 'Alt' on the keyboard, then release the mouse button.

**Axes Scales—Lifetimes**

Use the **Axes Scales** dialog to manually change the axes range. This can assist in zooming in on a particular
part of the graph.

To access the Axes Scales dialog, select the Axes Scales command from the Graph menu, or select the Axes Scales button on the Graphics toolbar.

The dialog comprises the following:

**X Axis**

<table>
<thead>
<tr>
<th>Mode</th>
<th>View the current units for the X axis.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Scale</td>
<td>Use the Scale field to enter a new range for the X axis, in the format n1 - n2 (where n1 is the start of the scale and n2 is the end of the scale).</td>
</tr>
</tbody>
</table>

**Hot Tip**
The Cary Eclipse will store the ten most recently entered ranges, which you can access by selecting the down arrow adjacent to the Scale field.

**Y Axis**

<table>
<thead>
<tr>
<th>Mode</th>
<th>View the current units for the X axis.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Scale</td>
<td>Use the Scale field to enter a new range for the Y axis, in the format n1 - n2 (where n1 is the start of the scale and n2 is the end of the scale).</td>
</tr>
</tbody>
</table>

**Hot Tip**
The Cary Eclipse will store the ten most recently entered ranges, which you can access by selecting the down arrow adjacent to the Scale field.

See also:
- Graph menu
- Graphics toolbar
- How to scale your graph manually
- How to scale your graph automatically
- How to zoom a graph

**How to scale your graph manually—Lifetimes**

Step 1

Click on the graph whose axes you want to change to make it the focused trace.

Step 2

Click the Axes Scales button on the toolbar, or select Axes Scales from the Graph menu. The Axes Scales dialog is displayed.

Step 3

Type in the new ranges for the X and Y axes in the format ‘n-n’, or choose an option for each from the drop-down list.

Step 4

Select OK. The axes on the selected graph will change to reflect your chosen settings.
How to scale your graph automatically—Lifetimes

Step 1
Click on the graph whose axes you want to change to make it the focused trace.

Step 2
To autoscale the graph, select one of the following:

- Click the **Autoscale XY** button on the toolbar to autoscale both X and Y axes.
- Click the **Autoscale X** button on the toolbar to autoscale the X axis only.
- Click the **Autoscale Y** button on the toolbar to autoscale the Y axis only.

The focused trace will be scaled according to the option selected.

Single/Multi Graph—Lifetimes

Select this option to toggle between a display of either one or multiple graph boxes in the Graphics area.

Example of a single graph box display
Example of a multi graph box display

**Note**
Up to 100 graphs can be displayed in the Graphics area at the one time.

See also:
- Graph menu
- Graphics toolbar
- Graphics How To

**Example of a Graphics Template—Lifetimes**

By using the Add Label command in the Cary Eclipse graphics system, you can customize a graphics template such as the one shown below:

The above is created as follows:

- The graph on the left is the collected data.
- The text on the top right is a multi-line label (added using the Add Label command).
- The logo is an inserted picture (added using the Add Picture command).
How to create a graphics template—Lifetimes

A graphics template lets you set up and store the style of a graphics report for future use. You can do this by adding labels and pictures to the background of the Graphics area and altering any axes styles etc. One advantage of this over the application's built-in reporting features is that you have more flexibility as to where you can position graph boxes and any text that you may want to appear (e.g. operator name, batch number etc.) and also where you add pictures, e.g. company logos etc.

Step 1

Start the application and open the data file from which you want to make a graphics template. Alternatively, collect some new data.

Step 2

Make as many graph boxes as you require in your template by using the Add Graph function.

Step 3

Adjust the graph boxes to the correct size and position them as you want them to appear in the template.

Step 4

Do any of the following, if required:

- Change the appearance of the graph boxes in the Graph Preferences dialog
- Use the Add Label function to append text to the Graphics area
- Use the Add Picture function to add pictures, such as your company's logo to the Graphics area

Step 5

From the File menu, select Print Preview. Use the Print Preview dialog displayed to check the template that you have created.

Step 6

When you are satisfied with the appearance of the graphics, select the Save As… command from the File menu. The Save As dialog is displayed.

Step 7

A list of stored data files will appear. Select the down arrow at the right of the Files of type field and select 'Graph Template' to list all the template files. Ensure that the Save only focused trace check box is cleared.

Step 8

Enter a name for the template in the File name field.
Select **OK** to save the graphic template. The template will be used each time you create a new file.

**Note**
You can also make a graphic template in a similar manner from existing data. No data will be saved in the template.

See also:
- How to add a label to a graph
- How to add a picture to a graph
- How to move a label
- How to move a picture

**Ruler Result—Lifetimes**

Use the ‘ruler’ function in the Lifetimes Application to measure zero order reaction rates in lifetimes traces. The ruler can be used after lifetimes data is collected to manually select the range over which to calculate the reaction rate. From this data, the slope the rate of reaction is determined.

To access the **Ruler Result** dialog, do the following:

1. From the Graph menu select **Cursor Modes**. In the Cursor Modes dialog, select **Ruler-Least Squares** or **Ruler-Point to Point** option as the cursor mode. See More Information on Least Squares and Point to Point calculations.
2. Select a region of the trace that is relatively straight and place the cursor on the point of the trace where you wish to start drawing the ruler. Click and hold down the left mouse button and drag the cursor to the second point on the trace. A ruler line will draw as you move the cursor.
3. Release the mouse button. The **Ruler Result** dialog is displayed.

The **Ruler Result** dialog comprises the following:

- **Slope** Displays the slope of the line of best fit in the region you selected.
- **X Start**/ **X Stop** Displays the beginning and end X co-ordinates of the region you selected.
- **Y Start**/ **Y Stop** Displays the beginning and end Y co-ordinates of the region you selected.
- **Retry** Select **Retry** to re-select a region of the trace.
- **Add Label** Select **Add Label** to add a label to the graph that displays the slope of the line you have selected.

**More information on Least Squares and Point to Point calculations**

**Least squares slope calculation**

You can calculate the rate of a zero order reaction by manually selecting the range over which to calculate the reaction rate of a lifetimes plot using the ruler on a particular graph.

A linear equation is fitted to the data using a Ruler-Least Squares technique. The slope of the line of best fit is reported as the lifetimes rate in the Ruler Result dialog using the previous zero order equation.

**Point to point slope calculation**

You can choose to calculate the point to point slope instead of the least squares slope value.
The point to point slope is calculated from the slope of a line drawn between two ordinate values at two, specified abscissa points.

If you choose OK or Add Label in the Ruler Result dialog then a corresponding Results table will appear in the Report area that includes the manually selected abscissa, ordinate range, calculated reaction rate (Slope) and Standard Deviation (SD).

The SD results means that the system looks at the difference between the actual Y value and the fitted Y value after the linear least squares fit or point to point fit has been applied. The system takes the difference of each point over the selected region and calculates the SD using the following equation:

\[ S = \sqrt{\frac{1}{n - 2} \sum_{i=1}^{n} (f_{m,i} - f_{c,i})^2} \]

where,

- \( f_{m,i} \) are measured values
- \( f_{c,i} \) are calculated values
- \( n \) is the number of points

The Factor* Slope result is also displayed. This is the product of the Factor multiplied by the calculated lifetimes ruler slope. The Factor value (entered in the Analyze tab) also appears above the result table in the report.

**Note**
You can only perform a manual zero order rate calculation after the data is collected. Once the data is collected, select **Cursor modes** in the **Graph** menu and choose the lifetimes ruler type you require. You can then select the range over which to calculate the reaction rate.

**See also:**
- **Cursor modes**
- **How to use the lifetimes ruler**

### How to use the lifetimes ruler—Lifetimes

The ‘ruler’ function can be used for measuring zero order reaction rates in lifetimes traces. You can use the ruler after lifetimes data is collected to manually select the range over which to calculate the reaction rate. The slope is then determined and thus the rate of reaction.

To use the ruler:

**Step 1**

From the **Graph** menu, select **Cursor Modes**. Alternatively, select the button on the toolbar. The **Cursor Modes** dialog is displayed

**Step 2**

Select **Ruler—Least Squares** to use a least-squares technique to determine the slope and thus the rate of the
reaction over the selected range. Alternatively, select **Ruler—Point to Point** to use a line drawn between two selected points to determine the point to point slope and thus the reaction rate.

**Step 3**

Select **OK** to clear the **Cursor Modes** dialog and return to the Lifetimes Application window.

**Step 4**

Select a region of the trace that is relatively straight and then place the cursor on the first point on the trace where you wish to start drawing the ruler. Press and hold down the left mouse button.

**Step 5**

Move the cursor to the second point on the trace (you will see the ruler line drawing as you move the cursor) and release the mouse button.

The **Ruler Rate** dialog will be displayed to enable you to view the start and end positions of the calculation range that you selected. The value of the calculated slope is also displayed.

If you are not satisfied with the rate result, select the **Retry** button to repeat Steps 3 to 5, this time selecting another range.

**Note**

If you wish to exit the lifetimes ruler mode, select **Cancel**. The lifetimes ruler trace will be removed from the graph.

**Step 7**

Select **Add Label** to add the determined slope value of the ruler trace to the graph. The **Ruler Rate** dialog will be cleared.

If you do not wish to add a label, simply select **OK** to clear the **Ruler Rate** dialog.

The results of the manual determination of the reaction rate will appear in a table in the Report area. The ordinate and abscissa start and end values are reported as well as the calculated slope. The result table will also include the calculated standard deviation as well as the Factor*slope result.

**See also:**

[Cursor modes](#)

**File Management—Lifetimes**

- [File menu](#)
- [Open dialog](#)
- [Save As dialog](#)
- [Filename extensions](#)
- [ASCII spreadsheet file](#)
- [Auto Store page](#)
- [How to export collected data](#)
- [How to combine data files into a batch file](#)
File menu—Lifetimes

Use the File menu to open or save a method or data file, print or preview a report, or exit the Cary Eclipse application.

Hot Tip
Highlight a method or data file in Explorer and simply drag and drop it onto the application to open it. Any data will be displayed in the Graphics area.

The following commands can be accessed from the File menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>New</td>
<td>Select New to create a new file. The Save As dialog will be displayed for you to enter the name of the new file and select a location.</td>
</tr>
<tr>
<td>Open Method…</td>
<td>Select Open Method to display the Open dialog where you can search for and load a stored method.</td>
</tr>
<tr>
<td>Open…</td>
<td>Select Open to display the Open dialog where you can search for and load method, data, report, graph template and batch files, as well as other file types compatible with the Cary Eclipse software. When you load a batch file, the method which collected the data is loaded as well as the associated report and graphics layout.</td>
</tr>
<tr>
<td>Save Method As…</td>
<td>Select this option to display the Save As dialog and store the current method. You can use the Save As dialog to store the method, data, report, or graphics template separately or together as a Batch file. The data can also be saved as a comma delimited ASCII Spreadsheet file.</td>
</tr>
<tr>
<td>Save As…</td>
<td>Select this option to display the Save As dialog and store the collected data. You can use the Save As dialog to store the method, data, report, or graphics template separately or together as a Batch file. The data can also be saved as a comma delimited ASCII file.</td>
</tr>
<tr>
<td>Print Setup…</td>
<td>Select this option to display the Windows Print Setup dialog where you can select the printer, printer page size and orientation.</td>
</tr>
<tr>
<td>Print Preview…</td>
<td>Select this option to display the Print Preview dialog where you can view the current report as it will be printed.</td>
</tr>
<tr>
<td>Print…</td>
<td>Select this option to display the Windows Print dialog where you can choose the printer and pages to print as well as the number of copies you wish to print.</td>
</tr>
<tr>
<td>Explorer…</td>
<td>Select this option to run Windows Explorer. This may help you locate method and data files.</td>
</tr>
<tr>
<td>History List</td>
<td>This displays a list of the four most recently opened files.</td>
</tr>
<tr>
<td>Exit</td>
<td>Select this option to exit the Lifetimes application.</td>
</tr>
</tbody>
</table>

See also:
- Open
- Save As
- Print Preview
- Filename Extensions
- ASCII Spreadsheet File

Open—Lifetimes
Use the dialog to open a file previously saved by the application. To access the Open dialog, select **Open** or **Open Method** from the File menu.

**Look in**
The Look In field lists the available folders and files. Click the down arrow on the right of this field to view the location of the current folder in your computer's hierarchy. You can also select a new location simply by selecting a different folder. The folders and files in the selected folder are displayed the list box below the Look In field.

**Note**
To open the folder in the level that is one higher than the current folder, click the toolbar.

**Look in list box**
The Look In List Box, which is directly below the Look In field, displays the folders and files in the selected location. To view the contents of a folder, double-click it.

- Click to create a new folder. Type the name of the new folder in the highlighted text entry field that appears and press Enter. Then double-click on the new folder to open it.
- Click to display the directory structure of the drive.
- Click to display the files as a list.
- Click to display files as a detailed list including the file size, file type and the date the file was last modified.
- Click to preview the selected trace before you load it into the application.

**File name**
Enter the name of the file you want to open in this field. The easiest way to do this is to find the file in the Look in list box and then select the file. However, you can type the full path of a file and then select Open to open the file.

**Files of type**
Click on the arrow on the right of this field to view a list of the different types of application files that can be opened in the list box.

*Methods (.*,FMLT)*
Choose this file type to open a Method file. Only the method is loaded. No data is loaded.

*Data (.*,FDLT)*
Choose this file type to open a Data file. Only the data is loaded. The method that was used to collect the data is not loaded. The last used method or default method will be displayed.

*Report (.*,FRLT)*
Choose this file type to open a Report file. You can view the report in the Report area.

*Graph Template (.*,FGLT)*
Choose this file to open a Graph Template file. The file will set up the layout of the Graphics area including any bitmaps as well as the range and style of the graph axes.

*Batch (.*,FBLT)*
Choose this file type to open a Batch file. A Batch file includes data, the method used to collect the data, report and graphics setup.

*Spreadsheet Ascii (.*,CSV)*
This will open ASCII files that contain single comma delimited files with two or more columns.

*All files (.*,)*
This enables you to view all files in the directory so that you can open any compatible file.

**Overlay Data**
Select this check box to overlay individual data files in one graph box. If this is not checked
the data from each file will appear in individual graph boxes. This option is only available if the Open dialog has been accessed by selecting Open… from the File menu.

**Hot Tip**
Use the SHIFT or Control key to select multiple data files.

**Open**
This button opens the highlighted file/s or the files listed in the File name field.

**Note**
This dialog appears when you select either the Save Method As… or Save Data As… menu option in the File menu. When you select Save Method As… this dialog appears with 'Methods' displayed in the Files of type field. When you select Save Data As… this dialog appears with 'Batch' displayed in the Files of type field.

Traces (or data) that are currently Visible in the Graphics area are saved, regardless of how they are displayed (full screen graph box or tile).

For example, if six graph boxes are displayed in the Graphics area and each graph box has one Visible trace then all six traces will be saved.

If you do not wish to save particular traces, make them not Visible in the Trace Preferences dialog.

The Save As dialog comprises the following:

**Look In**
The Look In field lists the available folders and files. Click the down arrow on the right of this field to view the location of the current folder in your computer's hierarchy. You can also select a new location simply by selecting a different folder. The folders and files in the selected folder are displayed the list box below the Look In field.

**Note**
To open the folder in the level that is one higher than the current folder, select on the toolbar.

**Look in list box**
The Look In List Box, which is directly below the Look In field, displays the folders and files in the selected location. To view the contents of a folder, double-click it.

Click to create a new folder. Type the name of the new folder in the highlighted text entry field that appears and press Enter. Then double-click on the new folder to open it.

Click to display the directory structure of the drive.

Click to display the files as a list.

Click to display files as a detailed list including the file size, file type and the date the
The Cary Eclipse system uses the filename extension to distinguish between data types (e.g. method or data) and also to differentiate applications (e.g. Concentration or Kinetics).

The Syntax for file extensions is:

### File Name
Enter the name of the file you want to save in this field. You can also type the full path of a file then select the Save button to save the file.

### Files of Type
Click on the arrow on the right of this field to view a list of the different types of application files that can be opened in the list box.

- **Methods (*.FMLT)**: Choose this file type to only save the current Method. Any collected data is not stored.
- **Data (*.FDLT)**: Choose this file type to only save the data currently Visible in the Trace Preferences dialog (for any graph box). Note that the method used to collect the data is not stored with the data.
- **Report (*.FRLT)**: Choose this file type to only save the contents currently shown in the Report area.
- **Graph Template (*.FGLT)**: Choose this file type to save the layout of the Graphics area as a template. For example, any bitmaps you have added to your Graphics area will be saved as well as the range and style of the graph axes.
- **Batch (*.FBLT)**: Choose this file type to save the data that is currently Visible in the Trace Preferences dialog (for any graph box), the method used to collect the data and the currently displayed report all together as a Batch file. The graphics setup is also stored.
- **Spreadsheet Ascii (*.CSV)**: Choose this file type to save the raw intensity data currently Visible in the Trace Preferences dialog (for any graph box) as a comma delimited ASCII Spreadsheet File. The data is saved as XY pairs delimited by a comma.
  
  **Note**
  This file type can be easily imported into a spreadsheet program. For example, Microsoft Excel. You can also double-click on the stored *.CSV file from Microsoft Explorer to load Excel automatically with the ASCII XY data displayed.

- **GRAMS (*.SPC)**: Choose this file type to only save the data currently Visible in the Trace Preferences dialog (for any graph box) as a Cary Eclipse GRAMS data file.

### Save
Click this button to save the file currently present in the File name field in the directory specified in the Look in field.

---

**Tell me more about Filename Extensions**

See also:

- File menu

### Filename extensions—Lifetimes

The Cary Eclipse system uses the filename extension to distinguish between data types (e.g. method or data) and also to differentiate applications (e.g. Concentration or Kinetics).

The Syntax for file extensions is:
The following files may be opened and saved in the Lifetimes Application:

<table>
<thead>
<tr>
<th>Extension</th>
<th>Available action</th>
<th>File type</th>
</tr>
</thead>
<tbody>
<tr>
<td>*.FMLT</td>
<td>Open/Save</td>
<td>A method file (*.FMLT) consists of the software setup by which data is collected.</td>
</tr>
<tr>
<td>*.FDLT</td>
<td>Open/Save</td>
<td>A data file (*.FDLT) contains data collected that is currently visible in the Trace Preferences dialog (for any graph box).</td>
</tr>
<tr>
<td>*.FRLT</td>
<td>Open/Save</td>
<td>A report file (*.FRLT) contains the report generated by the Cary Eclipse about the data.</td>
</tr>
<tr>
<td>*.FGLT</td>
<td>Open/Save</td>
<td>A graph template file (*.FGLT) contains information that sets up the layout of the Graphics area including any bitmaps as well as the range and style of the graph axes.</td>
</tr>
<tr>
<td>*.FBLT</td>
<td>Open/Save</td>
<td>A batch file (*.FBLT) contains the collected data that was Visible in the Trace Preferences dialog (for any graph box), the method by which the data was collected and the report and graphics template.</td>
</tr>
<tr>
<td>*.CSV</td>
<td>Open/Save</td>
<td>An ASCII spreadsheet file (*.FCSV) has the same extension for all applications. It consists of raw intensity data (that was currently Visible in the Trace Preferences dialog (for any graph box) saved as XY comma-delimited pairs. This file can be imported into a spreadsheet program such as Microsoft Excel.</td>
</tr>
<tr>
<td>Data.*</td>
<td>Open</td>
<td>A Cary DOS (Data.*) file is the equivalent of a Cary Windows Data file—it does not contain any Batch information.</td>
</tr>
<tr>
<td>*.ADL</td>
<td>Open</td>
<td>An ADL file (*.ADL) is a program file generated in the Applications Development Language (ADL).</td>
</tr>
</tbody>
</table>
An ASCII Spreadsheet file consists of raw intensity data saved as XY comma-delimited pairs. You can choose to import or export Cary Eclipse files to ASCII files, which can then be imported in spreadsheet programs such as Microsoft Excel.

The format is as follows:

- Line 1 contains the sample name plus a comma
- Line 2 contains X and Y labels. The name of the X label must be the same as that used to label the graph. For example, in Lifetimes, the graph label is Time (ms).
- Lines 3 and onward contain the X,Y data
- After the last line of X,Y pairs there must be a blank line separating the data from any text audit strings or the end of the file.

For example, if a lifetime file was collected as:
ms = 0.0083, intensity = 0.0255
ms = 0.01667, intensity = 0.0500
ms = 0.02450, intensity = 0.0750
etc.

It would appear as follows in the ASCII file:

```
Lifetimes,
Time (ms); Intensity (a.u.)
0.0083, 0.0255,
0.01667, 0.0500,
0.02450, 0.0750,

Lifetimes,
Collection time;

```

Microsoft’s Excel generates a CSV file with the above format.
There is also an alternative format for CSV files. For example,

<table>
<thead>
<tr>
<th>Lifetimes</th>
<th>Time (ms); Intensity (a.u.)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0.0083,0.0255</td>
</tr>
<tr>
<td></td>
<td>0.01667,0.0500</td>
</tr>
<tr>
<td></td>
<td>0.02450,0.0750</td>
</tr>
</tbody>
</table>

| Lifetimes, |
|           |
| Collection time; |

See also:
- Save As
- Open

How to export collected data—Lifetimes

If you want to export data from a Cary Eclipse application into a third-party application, then you simply need to save your data as an Spreadsheet ASCII (*.CSV) file or a Rich Text Format (*.RTF) file. Spreadsheet ASCII files are recommended if you are planning to use the data in a spreadsheet program such as Microsoft® Excel. Rich Text Format files are recommended if you are planning to open the data in a word processor program such as Microsoft® Word.

To export collected data:

Step 1

From the File menu, select Save As. The Save As dialog is displayed.

Step 2

Click on the down arrow next to the Files of type list box and select the Spreadsheet ASCII (*.CSV) or Rich Text Format (*.RTF) option.

Step 4

Type the name of your new file in the File name field.

Step 5

Select Save to export your data to this new file.

How to combine data files into a batch file—Lifetimes

To combine a number of data files into a batch file, do the following:

Step 1

Select Open… from the File menu to display the Windows Open dialog.
Step 2
A list of stored batch files will appear. Select the down arrow to the right of the Files of type field and select 'Data' to list all the data files.

Step 3
Select the Overlay Data check box.

Step 4
Highlight the data files that you require. Hold down the CTRL key to select multiple files.

Step 5
Select Open. The highlighted data files will load into the application and appear in the same graph box.

Step 6
Select Save As… from the File menu to display the Windows Save As dialog.

Step 7
A list of stored data files will appear. Select the down arrow at the right of the Files of type field and select 'Batch' to list all the batch files. Ensure that the Save only focused trace check box is cleared.

Step 8
In the File name field, type in the file name for your new batch file.

Step 9
Select Save to create the new batch file. The current method will be stored with the batch file.

All the data files are now combined into the one batch file.

Software and Screen features—Lifetimes

- Menu line
- Status display
- Status line
- Buttons
- Function keys
- Right mouse menu
- How to remove or display the command buttons

Menu line—Lifetimes

- File menu
- Edit menu
- View menu
- Commands menu
Status Line—Lifetimes

The Status Line is displayed at the very bottom of the Lifetimes Application window. The line will display the following information (from left to right):

- **Hardware Status**: This field displays status messages indicating the current instrument activity.
- **Sequence Information**: This field displays status messages indicating the current system activity.
- **Current Results**: This field displays information on the last reading.

See also: Lifetimes Application window

Buttons—Lifetimes

The major commands in the Lifetimes application can be accessed both from the menu items at the top of the main window and from the application buttons. These buttons, which appear in the left side of the Lifetimes Application window, comprise the following:

**Hot Tip**

If you wish to access these commands from the main menus, rather than from these buttons, you can remove the **Command** buttons from your application window. To do this, clear the **Buttons** command in the **View** menu. This will enlarge the Graphics area, making viewing and selection of scans easier.

<table>
<thead>
<tr>
<th>Button</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Setup</strong></td>
<td>Click this button to display the <strong>Setup</strong> dialog where you can set up your Cary Eclipse for the collection, storage and analysis of the lifetimes run. You can also set up any accessories that you may have and define the reporting options.</td>
</tr>
<tr>
<td><strong>Zero</strong></td>
<td>Select this to <strong>Zero</strong> the current ordinate value. This command can also be accessed by pressing F5.</td>
</tr>
<tr>
<td><strong>Note</strong></td>
<td>Before you select <strong>Zero</strong>, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.</td>
</tr>
<tr>
<td><strong>Pre-scan</strong></td>
<td>Select <strong>Pre-scan</strong> to quickly determine the optimum instrument parameters to be used for your sample. At the end of this collection sequence the software will report the optimum Ex and Em wavelengths and PMT voltage. Once you are satisfied with the parameters, the software will automatically update your method setup to reflect these new parameters.</td>
</tr>
<tr>
<td><strong>Clear Report</strong></td>
<td>Select <strong>Clear Report</strong> to remove all of the text that appears in the Report area. If you have saved the report, you can view the information again. To do this, simply open the report file using the Open… command in the Report menu.</td>
</tr>
<tr>
<td><strong>Recalculate</strong></td>
<td>Select <strong>Recalculate</strong> to display the <strong>Analyze</strong> and <strong>Reports</strong> pages of the <strong>Setup</strong> dialog. You can use the <strong>Analyze</strong> page to recalculate the rate, using the current data. The <strong>Reports</strong> page is used to enter and select information that will appear in the report.</td>
</tr>
<tr>
<td><strong>Print</strong></td>
<td>Select <strong>Print</strong> to display the Windows <strong>Print</strong> dialog where you can choose what printer the report will be printed to. You can also select what pages to print as well as the number of copies you wish to print.</td>
</tr>
</tbody>
</table>
Function keys—Lifetimes

The function keys at the top of your keyboard perform basic Cary Eclipse operations simply and quickly.

<table>
<thead>
<tr>
<th>Function key</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>This displays the Help for the current dialog or window.</td>
</tr>
<tr>
<td>F4</td>
<td>Press F4 to display the Go to Wavelength dialog.</td>
</tr>
<tr>
<td>F5</td>
<td>Press F5 to perform an instrument Zero. The current intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before selecting Start if you want your data to start at 0 intensity. This command is also accessible from the Commands menu or by selecting the Zero button.</td>
</tr>
<tr>
<td>F6</td>
<td>Press F6 to display the Rapid Result dialog where you can read a blank and multiple sample readings without changing the application method. This command is also accessible from the Commands menu.</td>
</tr>
<tr>
<td>F9</td>
<td>If the application is online, press F9 to start a data collection. This is equivalent to selecting the Start button. If the application is not online, you will need to press F9 to first connect to the Cary Eclipse. Then press F9 to start the data collection. This command is also accessible from the Commands menu or by selecting the Start button.</td>
</tr>
<tr>
<td>F12</td>
<td>Select this to stop the current measurement. This is equivalent to selecting the Stop button. This command is also accessible from the Commands menu or by selecting the Stop button.</td>
</tr>
</tbody>
</table>

Note
The options on the Report page are used to generate the contents of the Report area during a run or on a recalculation. If Auto Print has been selected the Cary Eclipse will automatically print a report at the end of a data collection.

See also:
- Commands menu
- Setup
- Zero
- Analyze
- Reports
- How to remove or display the command buttons

See also:
- Go to Wavelength
- Zero
- Rapid Result
- Start/Stop buttons
Right mouse menu—Lifetimes

The following commands are accessible by clicking the right mouse button on editable fields in the Cary Eclipse software:

- **Undo**: If available, this reverts to the previous value before the Edit on the field.
- **Cut**: If available, this cuts the highlighted text to the clipboard.
- **Copy**: If available, this copies the highlighted text to the clipboard.
- **Paste**: If available, this pastes the text from the clipboard to the cursor position.
- **Delete**: If available, this deletes the highlighted text/number.
- **Select All**: If available, this selects all entries.

How to remove or display the command buttons—Lifetimes

The command buttons (Setup, Zero, Pre-scan, Clear Report, Recalculate and Print) appear on the left side of the Lifetimes Application window.

To remove the command buttons

Step 1

From the Lifetimes Application window, select the View menu.

Step 2

Clear the Buttons menu item.

The buttons will disappear from the application screen, giving you more room to display reports.

To show the command buttons

Step 1

From the Lifetimes Application window, select the View menu.

Step 2

Select the Buttons menu item.

The buttons will re-appear in the application screen.

ADL—Lifetimes

- [ADL Command Line](#)
- [ADL Hooks](#)
- [How to insert ADL commands](#)

ADL Command Line—Lifetimes

Introduction to ADL
Use the ADL Command Line to run Single-line or Multi-line Applications Development Language (ADL) commands that enable you to perform functions over and above that of the existing program functionality.

ADL is not a general purpose programming language. It has been developed specifically by Varian to meet the needs of spectroscopists. It can be used to handle all the data collected from the Cary Eclipse—both photometric values and entire data collections. ADL programs that you develop should be stored with the extension 'ADL'.

To access the ADL Command Line, select the ADL Command item from the Commands menu. You can also press ALT+Y to open the dialog.

**Note**
Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.

---

**Software controls**

**Single Line**
Selecting this lets you enter ADL commands on a single line into the Command Line entry box.

**Q. Can I enter more than one ADL command on a single line?**
**A. Yes, provided that the commands are separated by a semicolon ';'.**

**Multi-line**
The Multi-line capability enlarges the Command Line entry box. Use the Multi-line capability to create a more complex ADL program by adding a series of commands over multiple lines. When Multi-line mode is selected, the Quick Reference option becomes available.

**Quick Reference**
Select Quick Reference to open the Quick Reference dialog where you can easily add a command in the correct syntax to your ADL program. This option is only available for Multi-line commands.

**Show me How to insert ADL commands using the Quick Reference**
A detailed description of all the commands in the Quick Reference is available by selecting the Help button on the Quick Reference dialog.

**Browse**
Click Browse to browse your release CD for ADL programming examples that you can load and run. Alternatively, you can select and load ADL programs that you may have created.

**Command Line (entry box)**
This entry box enables you to use ADL commands. You can enter single ADL commands into this field, or you can enter a series of ADL commands. If you have the entry type set to Single Line, you can review previous ADL entries by selecting the down arrow to the right of the list box.

The ADL commands in the Command Line are executed when you select OK.

**Note**
On entering this window, the Cary Eclipse will recall the last command or program executed from the Command Line entry box. The ADL commands entered in the last executed program are stored in the Adl folder (accessed from the Varian/Cary Eclipse software folder) as CaryEquation.ADL.
ADL Hooks—Lifetimes

The **ADL Hooks** command is accessible from the **View** menu.

Instead of writing a full ADL program you can use ADL hooks to link into an existing application. This is particularly useful if you want to make small changes to the way an application works, e.g. you want to pop up a dialog for the user to enter some specific data before the run starts. ADL hooks are links at specific points in an application at which you can run an ADL program. For example, ADL hooks have been supplied to run the following ADL programs:

**Note**
Full documentation on ADL programming is available in the ADL Online Help.
Click the button below to view this Help.

<table>
<thead>
<tr>
<th>Command</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>LT_Startup.ADL</td>
<td>This is run when the Lifetimes application starts. For this to to work the application must be started from a batch or method file that has ADL Hooks on.</td>
</tr>
<tr>
<td>LT_PreSeq.ADL</td>
<td>This runs when the <strong>Start</strong> button is selected.</td>
</tr>
<tr>
<td>LT_PostSeq.ADL</td>
<td>This runs at the end of the <strong>Start</strong> button sequence.</td>
</tr>
<tr>
<td>LT_Online.ADL</td>
<td>This runs when Cary Eclipse is detected online.</td>
</tr>
<tr>
<td>LT_OffLine.ADL</td>
<td>This runs when Cary Eclipse ‘goes’ offline.</td>
</tr>
<tr>
<td>LT_Zero.ADL</td>
<td>This runs when the <strong>Zero</strong> button is selected.</td>
</tr>
<tr>
<td>LT_Recalc.ADL</td>
<td>This runs when the <strong>Recalculate</strong> button is selected.</td>
</tr>
<tr>
<td>LT_Shutdown.ADL</td>
<td>This runs when the application is shutting down.</td>
</tr>
<tr>
<td>LT_PreRead.ADL</td>
<td>This runs just before a read is taken.</td>
</tr>
<tr>
<td>LT_PostRead.ADL</td>
<td>This runs just after a read is taken (It has the reading result stored in the UserResult variable.)</td>
</tr>
<tr>
<td>LT_PreSetup.ADL</td>
<td>This runs when the <strong>Setup</strong> button is selected.</td>
</tr>
<tr>
<td>LT_PostSetup.ADL</td>
<td>This runs when the OK button on the <strong>Setup</strong> dialog is selected.</td>
</tr>
<tr>
<td>LT_CellChange.ADL</td>
<td>This runs instead of the Cary Eclipse driving the Multicell Holder. It enables you to add your own instructions for the Cary Eclipse Multicell Holder or to use your own cell changing accessory.</td>
</tr>
<tr>
<td>LT_PreStage.ADL</td>
<td>This runs prior to each collection stage. It determines the stage by checking the UserResult variable as UserResult = stage number.</td>
</tr>
</tbody>
</table>
How to insert ADL commands using the Quick Reference—Lifetimes

The Quick Reference window is accessed from the ADL Command window. It enables you to easily add a command in the correct syntax to your ADL program. To do this:

Step 1
Press Alt-Y to open the ADL Command Line window.

Step 2
Select the Multi-line option. The Command Line entry box will expand to take multiple lines of ADL commands and the Quick Reference check box is displayed.

Step 3
Select the Quick Reference check box. The Quick Reference window is displayed.

Step 4
If necessary, use the Go To field to jump to the appropriate part of the Quick Reference listing.

Step 5
Highlight the command you require in the Quick Reference list.

Step 6
Click the Insert in ADL button. The selected ADL command will appear in the Command Line entry box.

Step 7
Where necessary, alter the defaults supplied by the inserted Quick Reference command in the Command Line entry box to suit your program.

Step 8
Repeat steps 4–7 to add more commands as required.

See also:

<table>
<thead>
<tr>
<th>Command</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>LT_PreUserRecalc</td>
<td>This runs just after the Recalculate button has been selected and performs any pre-processing required prior to a recalculation. If the ADL Variable UserBoolean is set to FALSE, it can also replace the normal recalculations.</td>
</tr>
<tr>
<td>LT_UserRecalc</td>
<td>Runs just prior to the calculations. Can also be used to replace the calculations</td>
</tr>
<tr>
<td>LT_PostUserRecalc</td>
<td>Runs after the Recalculate sequence has been completed</td>
</tr>
<tr>
<td>LT_TraceName</td>
<td>Allows the name of the sample to be obtained from an ADL file, instead of being entered by typing into the sample loading dialog. This only works for single samples.</td>
</tr>
<tr>
<td>LT_UserPreCalc</td>
<td>This runs just prior to the calculations. Any preprocessing of data should be done here.</td>
</tr>
<tr>
<td>LT_UserPostCalc</td>
<td>This runs after the calculations. Any post processing of data should be done here.</td>
</tr>
</tbody>
</table>
Help and Safety information—Lifetimes

- Help menu
- Safety home page

Help menu—Lifetimes

Use the **Help** menu to view the online help and software information for this application.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lifetimes Help</td>
<td>Select this to display the Lifetimes Home Page. This contains a list of online Help topics specific to the Lifetimes Application.</td>
</tr>
<tr>
<td>Help Topics</td>
<td>Select this to display the Cary Eclipse Windows Online Help Home Page.</td>
</tr>
<tr>
<td>Tip of the day</td>
<td>View useful tips about using your Cary Eclipse instrument and software.</td>
</tr>
<tr>
<td>Eclipse Tour</td>
<td>Select this to display the Cary Eclipse Multimedia Online Help. This will give you an animated introduction to the Cary Eclipse software and hardware.</td>
</tr>
<tr>
<td>About…</td>
<td>Select <strong>About</strong> to access a dialog that displays information about the application as well as Varian email and Internet contact information. To clear the <strong>About</strong> dialog, select <strong>OK</strong>.</td>
</tr>
</tbody>
</table>

See also: [Lifetimes home page](#)

Safety Home Page

Click on the desired index entry to view the Help.

- About Safety
- Lamp module
- Ultraviolet radiation
- Electrical hazards — Cary Eclipse
- Panels, covers and modules
- Other precautions
- Warnings and cautions
- Warning symbols
- Color coding
- Information symbols
- FCC advisory
- CE compliant products

Lifetimes How To—Lifetimes

Select the procedure you require below for a step-by-step informative guide on running the application software.

Lifetimes How To
Calculation and reporting How To

- How to perform post run calculations on previously stored data
- How to use the lifetimes ruler
- How to generate a report from previously collected data
- How to use advanced reporting options

Other How To

- How to email your results
- How to export collected data
- How to combine data files into a batch file
- How to remove or display the command buttons
- How to insert ADL commands using the Quick Reference

See also:
- Lifetimes Application Home Page
- Graphics How To

How to resize graph boxes—Lifetimes

Step 1

Click on the graph that you want to resize so that the highlight box appears around it.

Step 2

Move the cursor to the edge of the highlight box and place it over one of the square marks (e.g. ■) in the box until it turns into a double headed arrow (↔).

Step 3

Click and hold the left mouse button and drag the box to the desired size.

Step 4

Release the left mouse button.

See also:
- How to alter the look of your graphs

How to zoom a graph—Lifetimes

To zoom a graph, do the following:

Step 1

Click on the button on the toolbar, or select Cursor Mode from the Graph menu. The Cursor Modes dialog will appear.

Step 2

Select Free and select the OK button.
Step 3

Click and hold down left mouse button in the highlighted graph box.

Step 4

Drag the mouse sideways and up or down. You will see a zoom box made up of a dotted line appear on the screen. (See picture below.)

![Dotted line is a zoom box](image)

*The dotted line is a zoom box.*

**Hot Tip**

To move the box on the screen, hold down the left and right mouse buttons together. You can then position the box anywhere in the graph box. Releasing the right mouse button will still allow you to resize the zoom box.

Step 5

Release the left mouse button. The area defined by the zoom box will be redrawn to fill the entire graph box, and the scans within the box will be enlarged accordingly. The abscissa (X) and ordinate (Y) coordinates displayed below the graph will also be changed to reflect the positions of the corners of the zoom box.

You can repeat this process if you need to zoom in even further.

**Note**

The system may limit the extent to which you can zoom.

See also:

- [How to scale your graph manually](#)
- [How to scale your graph automatically](#)

**Maths How To...**

- [How to perform a spectral subtraction](#)
- [How to perform a mean calculation](#)
- [How to smooth a trace](#)
- [How to derive a trace](#)

See also:

- [Maths Home Page](#)

**How to take the derivative of a trace**

Some uses of derivatives are:

- To resolve partly overlapping peaks
- To quantitatively determine trace compounds
- To characterize pure compounds
- Purity testing
To take the derivative of a collected trace and plot the result, follow these steps:

Step 1
Click on the trace to be smoothed. It will become red (highlighted).

Step 2
Click the Selected Trace or Focused Trace button. The name of the highlighted trace will appear in the calculation display field.

Step 3
In the Trace group, select your derivative order in the Operation field. Choose from derivatives 1 to 4 (Deriv1, Deriv2, Deriv3, Deriv4). You may have to scroll to find the derivative you require.

Step 4
Enter a Filter Size and an Interval in the corresponding fields.

Step 5
In the Display Options group, select New Graph to plot the result in a new graph box, or Selected Graph to plot the result in the current graph box.

Step 6
Click the Apply button in the Trace group. Your Deriv selection will be displayed before the trace name in the calculation display field.

Step 7
Click on the equals (=) button. The result will be displayed in the Graphics area.

See also:
About derivatives

How to smooth a trace

To smooth a collected trace and plot the result, follow these steps:

Step 1
Click on the trace to be smoothed. It will become red (highlighted).

Step 2
Click on the Maths button in the toolbar (it's on the far right) or select the Maths menu.

Step 3
Click the Selected Trace or Focused Trace button. The name of the highlighted trace will appear in the calculation display field.
Step 4
In the Trace group, select Smooth in the Operation field. (You may have to scroll to find it.)

Step 5
Enter a Filter Size and an Interval in the corresponding fields.

Step 6
In the Display Options group select New Graph to plot the result in a new graph box, or Selected Graph to plot the result in the current graph box.

Step 7
Click the Apply button in the Trace group. The word ‘Smooth’ will appear before the trace name in the calculation display field.

Step 8
Click on the equals (=) button. The result will be displayed in the Graphics area.
# Ratio Home Page

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<th>Show me...</th>
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<td></td>
<td>• by entering the values manually</td>
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<tr>
<td></td>
<td></td>
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<td>• ADL Command Line, ADL Hooks</td>
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## Ratio Application

### Introduction

The Ratio application is used to measure a change in the intensity at two different wavelengths. It is used
primarily to measure wavelength shifting dyes which shift their wavelength maxima when they bind to ions.

Use the Ratio application to measure excitation or emission shifting dyes at two different emission or excitation wavelengths as a function of time. From this you can obtain an intensity versus time plot. The features of the application include:

- Calculation of Zero order, First order and Second order reaction rates from intensity versus time data.
- Selectable time window for calculation.
- Overlay of the best-fit line on raw data.
- Automatic or manual estimates for the first order and second order Marquardt fitting.

**Hot Tip:**
The Ratio application enables you to monitor the influx of intracellular ions by ratioing two wavelengths changing over time. If you wish to collect scans as a function of time in order to determine whether the spectrum is changing, use the **Cycle mode** option in the Scan application. The **Cycle mode** option is available from the **Options** page of the **Setup** dialog.

To access the Ratio Application, click the Windows **Start** button, point to **Programs, Cary Eclipse** and then select **Ratio**.

**Software controls**

The application comprises the following:

**Instrument Status Readings**
The Instrument Status Readings, which display the X and Y values of the graph axes are displayed in the top left and right corners respectively of the Ratio Application Window.

- **Y value**
  Displays the current Y value, intensity. This value is continually updated during data collection.

- **X value**
  Displays the current X value, which is the current reading for Time. On the Cary page of the Setup dialog, choose whether to display the time value in minutes or seconds.

**Start and Stop buttons**
The **Start** and **Stop** buttons at the top of the Ratio Application window are used to start and stop instrument analyses. These buttons can also be accessed from the **Commands** menu.

**Tell me more about the Start and Stop buttons**

**Graphics area**
The Graphics area refers to the plotting area of the application. It can contain traces of collected data, labels and pictures. In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area defined by the axes is referred to as a graph box.

To display or hide the Graphics area, select the Graphics command on the View menu. Double-clicking on the Graphics area enables you to toggle between half or full size display sizes. To manually size the Graphics area, place the mouse cursor over the border between the Graph and Report areas until the cursor changes to a double arrow (↕). Then hold down the mouse and drag it up or down to resize the areas.

**Tell me more about the Graphics area**
**Report area**

Use the Ratio Report area to view the current report. The report may include the results of your data collection, the method parameters used to collect the results and any graphics you have included. You can select the information to include in your report in the Reports tab of the Setup dialog.

To display or hide the Report area, select the Report command from the View menu. Double-clicking on the Report area enables you to toggle between half or full size display sizes. To manually size the Report area, place the mouse cursor over the border between the Graph and Report areas until the cursor changes to a double arrow (➡️). Then hold down the mouse and drag it up or down to resize the areas.

You can copy the generated report onto the Windows clipboard. To do this, highlight the report (or a relevant section of it) and press CTRL+C. You can then paste the report into a word processor such as Microsoft® Word.

**Hot Tip**

You can edit the Ratio Report by pressing CTRL+R or selecting the **Edit Report** command from the **Edit** menu.

**Commands**

The Ratio Application window contains the following commands, all of which can also be accessed from the various menus in the application.

*Click on the buttons to view information about them:*

- Setup ...
- Zero
- Set temp ...
- Stop time ...
- Clear report
- Recalculate
- Print ...

**Hot Tip**

By becoming familiar with these commands (Setup, Zero, Set Temp, etc.), you can remove these buttons from the Ratio window. This will enlarge the Report area and make it easier to view results. To do this, clear the Buttons item in the View menu.

**Menus**

The Ratio Application contains the following menus:

- File
- Edit
- View
- Commands
- Setup
- Graph
- Help

See also:

- Status Display
- Status Line
- Keyboard function keys
- Ratio home page

**Validation of your Cary Eclipse for the Ratio application**

Two files are shipped with the Ratio application to enable you to validate the software. The files are stored on your CD in a Validation subdirectory.

To validate your software, you should Open each file in turn and select the Recalculate button. The results calculated by the Cary Eclipse should match those given in the validation files listed below. These results were determined by exporting the data to a spreadsheet program and performing the same calculation as that used in the Ratio application.

- Calcium Fura 2 final results.FBRT
- Calcium single wavelength final results.FBRT

**Getting ready for a run—Ratio**
Setup—Ratio

Use the Setup dialog to set up your instrument and accessories for a Ratio run. You can also use the dialog to set up the Analysis, Report and Storage parameters associated with the run.

To access the Setup dialog select the Setup button from the Ratio Application window or the Setup command from the Setup menu.

The Setup dialog comprises the following tabbed pages:

- **Cary**
- **Options**
- **Accessories**
- **Analyze**
- **Reports**
- **Auto Store**

See also:
- Ratio Application

### Setup menu—Ratio

Use the Setup menu to access the parameters for setting up a Ratio read.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Setup</strong></td>
<td>Choose Setup to display the Setup dialog where you can set up your instrument for a Ratio measurement. The Setup dialog can also be accessed from the Setup command button.</td>
</tr>
<tr>
<td><strong>Cary</strong></td>
<td>Choose Cary to display the Cary page. You can use this page to set up the instrument parameters associated with the data collection.</td>
</tr>
</tbody>
</table>
Use the Cary page to set up the instrument parameters associated with the data collection.

From the Setup menu, select Cary to access the Cary page. You can also access the page from the Setup dialog by selecting the Cary tab. To access the Setup dialog, select the Setup button from the Ratio Application window.

**Note**
The Ratios application operates in Fluorescence mode only.

### Instrument setup

**Data mode**  
**Fluorescence**  
Fluorescence refers to the emission of light from an excited single state. The fluorescence signal is measured when the lamp flashes.

### Wavelength setup

**Collection type**  
Choose from the following:

*Single wavelength*  
Select this option to monitor a single wavelength in your reaction, reading the intensity of one wavelength over time. Select this option for non wavelength shifting dyes.

*Multi wavelength*
Select this option to monitor two wavelengths in your reaction. Select this option if you are using wavelength shifting dyes whose wavelength maxima shift in the presence of the dye they measure. If you select **Multi wavelength**, the Multi wavelength table will be displayed.

**Multiwavelength table**
Use the Multi wavelength table to enter the excitation and emission wavelengths to be monitored for each wavelength.

**Excitation fast filter**
Select this option to use the fast filter accessory to collect excitation wavelengths. This option can be used to measure the excitation of wavelength shifting dyes when the ion response rate is very fast.

**Note**
You will need to install the fast filter accessory if you wish to select this option.

**Emission fast filter**
Select this option to use the fast filter accessory to collect emission wavelengths. This option can be used to measure the emission of wavelength shifting dyes when the ion response rate is very fast.

**Note**
You will need to install the fast filter accessory if you wish to select this option.

**Ex. Wavelength**
Set the wavelength of the excitation monochromator, which is the wavelength value at which you want to excite your sample. Click the arrow to the right of the field to quickly select the most recent wavelength entries. The **Ex. wavelength** option is not available if you have selected the **Multi wavelength** or **Excitation fast filter** options.

Alternatively, to set the excitation monochromator to zero order (white light), from the **Commands** menu, point to **Zero Order** and select **Excitation**. A tick next to this option indicates that it is selected. Clear this option to return to monochromator light and use the value set in the **Ex. Wavelength** field.

**Em. Wavelength**
Set the wavelength of the emission monochromator, which is the wavelength value at which your sample emits. Click the arrow to the right of the field to quickly select the most recent wavelength entries. The **Em. wavelength** option is not available if you have selected the **Multi wavelength** or **Emission fast filter** options.

Alternatively, to set the emission monochromator to zero order (white light), from the **Commands** menu, point to **Zero Order** and select **Emission**. A tick next to this option indicates that it is selected. Clear this option to return to monochromator light and use the value set in the **Em. Wavelength** field.

**Ex. Slit (nm)**
Set the slit width of the excitation monochromator.

*As a general rule:*
At a fixed emission slit width, increasing the excitation slit width will result in an increase in the fluorescence excitation intensity.

**Em. Slit (nm)**
Set the slit width of the emission monochromator.
As a general rule:
At a fixed excitation slit width, a two-fold increase in the emission slit width will result in a four-fold increase in the emission intensity. Accordingly, a two-fold decrease in the emission slit width will result in a four-fold decrease in the emission intensity.

Hot Tip
Increase your slit width and decrease your PMT to get an increase in the signal intensity and a decrease in noise. Be careful not to start with too high an intensity as this may lead to an over range.

Collect timing

Simple collect  Select this option to set the Number of Stages to '1'. Use one stage if you require only one collection rate for the entire ratio run.

Advanced collect  Check this option to enable the Number of Stages entry field and specify the collect time profile of a run, enabling you to vary the rate at which data is collected over the period of the run. Enter the times required in the fields in the Cycles table.

Q. When do I choose an Advanced Collect?
A. The Advanced Collection option allows you to divide the length of the run into different sections or ‘stages’. You can nominate a different data collection rate for each stage. This means that you can collect lots of data points during the stage when the reaction is happening quickly and then reduce the number of data points collected when the intensity change is slower.

No. of stages
Use this to set the number of time segments into which you wish to divide the total data collection time, each having a different data collection rate. The Cycles table will expand to fit the number you enter.

Q. Why isn't the Number of Stages entry field active?
A. You have selected Simple Collect. On selecting Advanced Collect, the field will be activated.

X Mode
This enables you to set the units of time for the abscissa (or X axis).
Choose your desired abscissa mode by selecting one of the following buttons:

Min
Select this to time the run in minutes.

Sec
Select this to time the run in seconds.

Ave time (s)
Set the amount of time, in seconds, for which data is averaged. To do this, enter the amount in the Ave. time field, or use the up/down arrows to the right of the field to increase or decrease the time. The averaged result is displayed in the report.

When setting the Ave Time, consider the following:

- The longer the Ave Time, the longer the signal will be averaged
- A longer Ave Time will result in more precise results as the averaging of the signal reduces any contribution of background ‘noise’.
Averaging is calculated as follows:
the number of readings averaged are taken from consecutive flashes at 80 Hz where:
number of consecutive flashes = averaging time/0.0125.

**Hot Tip**
By increasing the averaging time it is possible to collect more data points, up to 80 points/sec. This is useful if you have a fast run.

**Cycles Table**
This table enables you to vary the data collection rates within the one ratio run. This feature is particularly useful when analyzing slow reactions and multi-step reactions.

**Stage**
This column displays the stage number/s.

**Cycle**
Enter a number in this numeric field to set the rate at which the system will cycle the specified cell setup.

**Stop**
Type a number in this numeric field to set the **Stop Time** to establish how long you require the ratio analysis to continue.

**Note**
**Stop Time** overrides **Cycle Time** so make sure that the **Stop Time** is long enough to allow the measurement of all Cycles.

**Status Display**
Select this check box to display the **Status Display** window, where you can view the status of various parameters during the data collection.

Use the Options page to set up your filters and set the detector voltage. You can also use this page to select the way the collected data is displayed in the Graphics area, and the type of smoothing applied (if any).

From the **Setup** menu, select **Options** to access the **Options** page. You can also access the page from the **Setup** dialog by selecting the **Options** tab. To access the **Setup** dialog, select the **Setup** button from the Ratio Application window.

**Display Options**
Select this option to superimpose the collected data of each sample in the run in one graph box. If you select this option, a graph box is created at the start of the collect in the Graphics area and each sample’s data is overlaid in that box as the data is collected. Traces will always
be overlaid in the same graph, regardless of what multicell/well accessory is used. Clear this option to display all of the traces in individual graphs.

**Note**
When using the Wellplate reader it is possible to define how you want to overlay the traces. This is done on the **Accessories** tab when the Wellplate reader has been selected, using the Display options button.

_Y minimum/Y maximum_
Enter the minimum and maximum Y values to be displayed on the graph axis. The software will update the Y axes to reflect these settings during the ratio run.

**Smoothing**
Select **Smoothing** to reduce the noise of your collected data. When you select this option, smoothing will be applied to the ratioed intensity signal using the algorithms defined below. You can select one of the three types of smoothing that can be performed during a run. Only the smoothed trace is displayed during data collection, however the ratioed unsmoothed data is also saved with the experiment.

**Type**

**Savitzky-Golay**
This algorithm will take the number of Filter points, apply the Savitzky and Golay smoothing algorithm and display the smoothed value at the middle X-point of the filter.
The naming of the smoothed trace will be 'Sample name_SG'.

[Tell me more about Savitzky-Golay smoothing](#)

**Boxcar average**
This algorithm will take the number of points as defined by the Filter size, and average these, displaying the average at the middle X-point of the filter.
The naming of the smoothed trace will be 'Sample name_Box'.

[Tell me more about Boxcar averaging](#)

**Moving average**
This algorithm will take the number of points as defined by the Filter size, and average these, displaying the average at the middle X-point of the filter.
The naming of the smoothed trace will be 'Sample name_Mov'.

[Tell me more about Moving average](#)

**Filter size**
Enter the **Filter size**, which determines the number of points that will be used in the smoothing procedures. The higher the filter size (and thus the number of points used), the greater the amount of smoothing that will be performed.

Filter sizes must be odd values. For Boxcar and Moving averages, the range is 3–101. For Savitzky-Golay smoothing, the range is 5–101.

**Excitation filter**
Set the excitation filter for the Ratio measurement. To do this, select the down arrow to the right of the field and select an option from the list.

A range of filter settings are available:

**Auto**
If you select the **Auto** option, the filter wheel will be automatically moved to the appropriate position for the selected excitation wavelength. It is recommended that you set the filter to **Auto** (which is the default setting).

**Open**
If you select **Open** no filter is used.

**Closed**
If you select **Closed**, a blank filter is used to prevent light from getting through.

**Emission filter**
Set the emission filter for the Ratio measurement. To do this, select the down arrow to the right of the field and select an option from the list.

A range of filter settings are available:

**Auto**
If you select the **Auto** option, the filter wheel will be automatically moved to the appropriate position for the selected emission wavelength. It is recommended that you set the filter to **Auto** (which is the default setting).

**Open**
If you select **Open** no filter is used.

**Closed**
If you select **Closed**, a blank filter is used to prevent light from getting through.

**250–395, 335–620, 550–1110, 695–1110**
If you are looking at a wavelength range that is within one of these filter ranges, you can select your filter manually. Otherwise it is recommended that you set the filter to **Auto**.

**3%T Attenuator**
Select **3%T Attenuator** to decrease the fluorescence intensity and thus the amount of light going to the emission PMT. This setting is useful for highly fluorescent samples when you are using wide slits.

**PMT Detector Voltage**
Set the EHT or sample detector PMT. To do this, select one of the following:

**Low**
Sets the PMT Detector voltage to a pre-defined value of 400 v. Typically you would select this setting if you are reading highly emitting samples.

**Medium**
Sets the PMT Detector voltage to a pre-defined value of 600 v. Typically you would select this setting if you are reading moderately emitting samples.

**High**
Sets the PMT Detector voltage to a pre-defined value of 800 v. Typically you would select this setting if you are reading low emitting samples.

Manual
Enables you to enter any PMT voltage in the adjacent Volts field.

Volts
Enter the PMT voltage in the adjacent field. Note that this option is only available if you have selected Manual as the PMT Detector voltage.

Tip
Increasing or decreasing the PMT Detector Voltage by 10% will double or halve the fluorescence intensity observed.

Reagent list
Select this option to display the Reagents List. To set the list of reagents:
Enter your solutions to be used during the reaction in the Reagent Name list, typing the reagent name in the available entry fields. Be sure to set the reagent number to five even if five reagents are not used.

To display the Reagent List once you have exited the Setup dialog simply select the Reagent List button. If you wish to time stamp the point at which a particular reagent was added simply click on the reagent entry field in the open Reagent List and this will note on the graph at what time the selected reagent was added to the reaction.

Status Display
Select this check box to display the Status Display window. This window allows you to view the status of various parameters during the data collection.

See also:
Setup
Status Display window

About Smoothing—Ratio

Savitzky-Golay
Savitzky-Golay smoothing is used to obtain good noise reduction with sharp peaks. The equation constants are specific to Filter Size. The constants for each filter size are published in the Savitzky-Golay paper (A. Savitzky, M.J.E. Golay, Anal. Chem., 36 (1964) 1627). The equation also requires that Filter Sizes entered must be odd values, between 5 and 101.

Example

Data       (x1, y1), (x2, y2), (x3, y3) … (xn,yn)
Filter size 5

Result

The first smoothed data point will be:
(x3, (Y1*a1+Y2*a2+Y3*a3+Y4*a4+y5*a5)/b),

the next:
(x4, (y2*a1+y3*a2+y4*a3+y5*a4+y6*a5)/b) …
(xn-2, sum(yn-4*a1+yn-3*a2+yn-2*a3+yn-1*a4+yn*a5)/b),

where a1..a5, b are constants defined by Savitzky and Golay.

Each smoothed point is calculated using the previous and next data points. For this reason, calculations cannot be performed on the very first and very last data points in a data set. Hence, smoothed traces will lack the first and last X values of the original trace.

**Boxcar average**

Boxcar averaging can be used when the change in intensity is minimal.

**Example**

Data  \((x_1, y_1), (x_2, y_2), (x_3, y_3) \ldots (x_n, y_n)\)

Filter size  5

**Result**

The first smoothed data point will be:
\((x_3, \text{sum}(Y_1:y_5)/5),\)

the next:
\((x_8, \text{sum}(y_6:y_{10})/5) \ldots (x_{n-2}, \text{sum}(y_{n-4}:y_{n})/5).\)

If there are not enough points at the end of the data set to complete the filter size, then these points will be
ignored, hence, the gap at the end may be larger than the gap in the beginning.

**Moving average**

Example

Data: (x1, y1), (x2, y2), (x3, y3) … (xn, yn)
Filter size: 5

Result

The first smoothed data point will be: (x3, sum(Y1:y5)/5),

the next: (x4, sum(y2:y6)/5) … (xn-2, sum(yn-4:yn)/5).

**Accessories—Ratio**

Use the Accessories page to set up the accessory parameters associated with the data collection.

From the Setup menu, select Accessories to access the Accessories page. You can also access the page from the Setup dialog by selecting the Accessories tab. To access the Setup dialog, select the Setup button from the Ratio Application window.

The Accessories page will vary, depending upon what accessory button you have selected from the following:
Multicell and Temperature accessories—Ratio

The Multicell and Temperature accessory page is displayed when you select the **Multicell/Temperature** tab on the **Accessories** page.

The page comprises the following:

**Multicell Holder**

Select this option to use the Multicell Holder and cells with the Cary Eclipse.

**Note**

By selecting this accessory you will prohibit the use of some combinations of other accessories that are also controlled by the Accessory Controller Board.

**Multi-zero**

If you select **Multi-zero**, a Multi-zero will be performed when the **Zero** button is selected in the Ratio Application window. Because cuvettes can have different intensities, and samples vary, this field is used when you require all blank readings to consistently start at zero intensity. Please refer to the read me file for Multi-zero.

A reading will be taken at the set **Averaging Time** at the set Ex. and Em. wavelengths and at every selected cell position. These readings will be offset to zero and subsequent readings will have the corresponding offset value subtracted from each cell.

**Reset**

Select **Reset** to drive the Multicell Holder to cell position 1, which is its fully retracted or zero position.

**Go to cell...**

Select **Go to cell** to open the **Multicell Changer** dialog. You can use this dialog to manually move the Multicell Holder accessory to a new cell. To do this, simply select the corresponding button of the cell you require in the Multi Cell Changer dialog. The Multicell Holder will move the selected cell to the measurement position in the sample compartment.

The **Multicell Changer** dialog is ‘modeless’ which means that it can remain open even once you have cleared the Setup dialog. This enables you to use the **Rapid Read** command to perform a reading at the cell specified in the Cell Changer.

If you move to a new cell after you have performed a Multi-zero scan, the Cary Eclipse will display the intensity reading corrected by the determined zero-offset amount for that cell.

**Note**

You can use this option to check the intensity of a cell or start a ratio run at a particular cell.

**Temperature Control**

Select this option to use the Temperature Controller with the Cary Eclipse.

**Note**
The **Temperature Control** option does not control temperature accessories that are not listed in the **Device** field. For example, if you have selected the **Temperature Control** option and set the **Device** to **None** because you have a different temperature control accessory, the Cary Eclipse software will not control that accessory. However, it will monitor the temperature and prompt the software to start when the accessory reaches the temperature listed in the **Temperature** field.

**Device**

Select the device that will be used to control the solution temperature.

The options available will depend upon whether or not you have selected the **Multicell holder** option.

If you *have* selected the Multicell holder, the following **Device** options will be available for selection:

- Temperature controller
- PCB 150 water bath
- None

If you *have not* selected the Multicell holder, the following **Device** options will be available for selection:

- Single cell peltier
- PCB 150 Water bath
- None

**None**

Select this option if you are using a Temperature Control device not listed in this field (for example, a third party water bath). Note that the Cary Eclipse software will not control the accessory. The software will simply monitor the temperature and prompt the software to start when the accessory reaches the temperature listed in the **Temperature** field.

**PCB 150 Water Bath**

Select this option if you are using the PCB 150 water bath.

**Single cell peltier**

Select this option if you are using the Single cell peltier.

**Temperature controller**

Select this option if you are using the Temperature controller.

**Temperature**

Set the temperature at which the reaction will be performed. The default value is 20.00°C.

When you select **OK** to close the **Setup** dialog, all buttons including the Start button in the Ratio Application window will be disabled until the temperature recorded by the **Monitor** device is within ± 0.5°C of the temperature range.

If **Zero** or **Start** is selected, and the temperature is not within ± 0.5°C of the temperature range, then the system will wait for the temperature to be within the set range before continuing with the measurement. Whilst waiting for the temperature, there will be a display on the status line at the bottom of the Ratio Application window giving the current temperature and the elapsed time waiting to reach the set temperature.

**Port**

Select from **COM1**, **COM2**, **COM3** or **COM4**.

This option is not available if you have selected **None** as the **Device**.

**Monitor**

Select which device will be used to monitor the reaction temperature. Select from **Block**, **Probe 1**, **Probe 2**, **Probe 3** or **Probe 4**.

Note that the the options in this field may change depending on the **Device** selected.

**Temperature Display**

Select this option to view Temperature Display information in the **Status Display** window.
The Temperature Display option is only available if you have selected the Temperature Control option.

- **Block/PCB 150**: Select this option to view the temperature of the Block (if you have selected the Single Cell Peltier device) or the PCB 150 (if you have selected the PCB 150 Water Bath) in the Status Display window.

- **Probes**: Select one or more of these options to view the temperature of the Probes in the Status Display window.

### Status Display

Select this check box to display the Status Display window. Use this window to view the status of various parameters during the data collection.

- **Accessories**
  - Multi Cell Changer
  - Fast Filter Accessory
  - Status Display

### Multi Cell Changer—Ratio

Use the Multi Cell Changer dialog to manually move the Multicell Holder accessory to a new cell. To access the Multi Cell Changer dialog, select the Go to cell button from the Multicell/Temperature Accessories page.

The dialog comprises the following:

- **Multicell changer**: To manually move the Multicell Holder accessory to a new cell, simply select the corresponding button of the cell you require in the Multi Cell Changer dialog. The Multicell Holder will move the selected cell to the measurement position in the sample compartment.

  The Multi Cell Changer dialog is ‘modeless’ which means that it can remain open even once you have cleared the Setup dialog. This enables you to use the Rapid Read command to perform a reading at the cell specified in the Cell Changer.

  If you move to a new cell after you have performed a Multi-zero scan, the Cary Eclipse will display the intensity reading corrected by the determined zero-offset amount for that cell.

  **Note**: You can use this option to check the intensity of a cell or perform a scan at a particular cell.

- **See also**:
  - Multicell/Temperature Accessories
  - Rapid Read

### Wellplate accessory—Ratio

The Wellplate accessory page is displayed when you select the Wellplate tab on the Accessories page.

The page comprises the following:

- **Wellplate reader**
The Wellplate accessory page is displayed when you select the Wellplate tab on the Accessories page.

The page comprises the following:

**Wellplate reader**

Select this option to use the Wellplate reader with the Cary Eclipse.

Q. Why is the Wellplate reader option unavailable?
A. The multicell holder, temperature control and/or polarization accessory has been selected. As you cannot place any of these accessories and the Wellplate reader in the Cary Eclipse sample compartment at the same time, you must clear other accessory selections before selecting the Wellplate reader.

**Plate format**
Select the type of Wellplate to be used. The default options are 96 wells, 192 wells or 384 wells. The graphical representation of the Wellplate will change according to the number of wells chosen.

**Auto-zero**
Select this option to perform an auto-zero when the Zero button is selected from the Ratio Application window. A zero reading will be taken at the selected well position using the current method setup. To select the well position to be zeroed, use the Select Well option.

These readings will be offset to zero and subsequent readings will have this single value subtracted from each well in the sequence. The reading is taken only once, stored, and subtracted from the ratioed intensity reading. Refer to the Fluorescence Late Breaking news for additional information on ratioed intensities.

**Select well**
Choose Select well to open the Go To well dialog where you can select the well position to be zeroed. This option is only available if you have selected Auto-zero.

**Well**
Displays the well position of the well selected for auto-zeroing. This field is not editable.

**Display options**
Select this option to open the Display Options dialog where you can set up the display parameters associated with the Wellplate reader.

**Sample names**
Select Sample Names to open the Wellplate Sample List dialog where you can enter or modify the sample names. Highlight one or more wells on the Wellplate reader diagram to view and change their sample information.

**Go to well**
Select this option to open the Go To Well dialog. Select a well on the Wellplate reader diagram and a Go to well dialog will be displayed. This dialog will remain open once the Setup dialog is closed allowing you to select individual wells before each run. This option is only available if you have disabled the Wellplate reader check box.

**Well position**
Displays the well position that the mouse cursor is pointing to. This field is not editable.

**Reset**
Select Reset to move the Wellplate reader to the first well position defined by the read sequence.

**Park**
Select Park to move the Wellplate reader to the Park position. The Wellplate reader accessory is moved clear of the sample compartment to enable you to easily access all wells.

**Wellplate reader diagram**
The Wellplate reader diagram represents the number of wells present for the Wellplate configuration you have chosen.

The labels for the rows will be 1 .. 12 and for the columns A .. H (for a 96 Wellplate). Similar labels will occur for different Wellplates according. The sequence for numbering the read order will be from A1 to A12, B12 - B1, C1 - C12…etc for a 96 Wellplate. A similar sequence will be valid for other plates.
Status Display

Select this check box to display the Status Display window. This window allows you to view the status of various parameters during the data collection.

See also:
- Accessories page
- Display Options dialog
- Go To Well dialog
- Wellplate Sample List dialog
- How to import sample names into the Wellplate sample list
- Status Display window

Display Options—Ratio

Use the Display Options page to set up the display parameters associated with the Wellplate Reader.

The Display Options page is accessible from the Wellplate Accessories page by selecting the Display Options button.

The page comprises the following:

Display

Individual graphs
Select this option to display the collected data of each sample in individual graph boxes, rather than overlaid on each other in one box.

At the start of a scan, a separate box is created for each sample. Similarly, the collected data for each sample is displayed in a separate graph box.

Overlay all traces
Select this option to superimpose the collected data of each sample in the run in one graph box.

One box is created at the start of the collect in the Graphics area and each sample’s data is overlaid in that box as the data is collected.

User defined
Select this option if you wish to specify the exact number of traces to be included on one graph.

Number of traces per graph
Enter the number of traces to be displayed per graph.

Overlay rows
Select Overlay rows to view overlaid graphs of the wells you have selected. Wells in the same row of the wellplate reader will be overlaid. If you had selected wells as shown in the adjacent diagram, a total of seven graphs would be displayed (as seven rows contain selected wells). The first graph would contain
Wellplate Sample List—Ratio

Use the Wellplate Sample List dialog to enter or modify the sample names of each well in the wellplate reader.

The Wellplate Sample List is accessible from the Wellplate Accessories page by selecting the Sample Names button.

The page comprises the following:

- **No. of samples** Displays the total number of samples that have been selected from the Wellplate Reader diagram on the Wellplate Accessories page. This field is not editable.
- **Sample table** The sample table lists the Sample Names and Well numbers of the selected samples.
  - **Sample Name** Samples are named numerically, depending upon where they are located on the Wellplate Reader diagram. The sequence for numbering the Sample Name order is to read from A1 to A12, B1 to B12, C1 to C12…etc for a 96 wellplate. Therefore, if the B2 cell is selected, its corresponding Sample Name will be Sample 14.
  
  To modify the name of a sample, highlight the sample name and type in a new name. You can enter up to 20 characters for each name.

- **Well** Displays the position of the well corresponding to the Sample Name. This field is not editable.
- **Import names** Select Import Names to display the Open dialog which you can use to select a text file to import into the Sample Names column.

See also: [Wellplate Accessories](#)

How to import sample names into the Wellplate sample list
Go To Well—Ratio

You can use the Go To Well dialog to manually move the Wellplate Reader accessory to a new well. This enables you to check the intensity of a well or perform a read at a particular well.

The Go To Well page is accessible from the Wellplate Accessories page by selecting the Select well or Go To Well button.

The Go To Well dialog is ‘modeless’ which means that it can remain open even once you have cleared the Setup dialog. This enables you to use the Rapid Read command to perform a reading at the selected well.

If you move to a new well after you have performed a Multi-zero, the Cary Eclipse will display the intensity reading corrected by the determined zero-offset amount for that well.

The page comprises the following:

**Hot Tip**

If your samples are the same for a number of reactions, create a text file containing the name of each sample and simply import the file into the Sample Names Column for each run.

**Increment**

Select Increment to copy and increment the sample names in the Sample Name list. This will save you from entering each sample name individually when the names consist of a descriptor followed by a number.

To use the increment option, highlight the name that you want to increment and select Increment. The Cary Eclipse will increment the names of all the files proceeding the first highlighted name.

**Example:**

In this example, Sample B1 is highlighted when the Increment button is selected.

<table>
<thead>
<tr>
<th>Sample Name</th>
<th>Becomes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Samp A</td>
<td>Samp A</td>
</tr>
<tr>
<td>Sample B</td>
<td>Sample B1</td>
</tr>
<tr>
<td>Sample C</td>
<td>Sample B2</td>
</tr>
<tr>
<td>Sample D</td>
<td>Sample B3</td>
</tr>
<tr>
<td>Sample E</td>
<td>Sample B4</td>
</tr>
</tbody>
</table>

See also:

- Wellplate Accessories page
- About the Wellplate Reader

**Go To Well—Ratio**

You can use the Go To Well dialog to manually move the Wellplate Reader accessory to a new well. This enables you to check the intensity of a well or perform a read at a particular well.

The Go To Well page is accessible from the Wellplate Accessories page by selecting the Select well or Go To Well button.

The Go To Well dialog is ‘modeless’ which means that it can remain open even once you have cleared the Setup dialog. This enables you to use the Rapid Read command to perform a reading at the selected well.

If you move to a new well after you have performed a Multi-zero, the Cary Eclipse will display the intensity reading corrected by the determined zero-offset amount for that well.

The page comprises the following:

**Wellplate reader diagram**

The Wellplate Reader diagram represents the number of wells present for the chosen wellplate configuration.

The labels for the rows will be 1 .. 12 and for the columns A .. H (for a 96 wellplate). Similar labels will occur for different wellplates accordingly. The sequence for numbering the read order will be from A1– A12, B12– B1, C1– C12, etc for a 96 wellplate. A similar sequence will be for other plates.
To move the Wellplate Reader accessory to a new well, simply click on the corresponding well on this diagram. The Wellplate Reader will move the selected well to the measurement position in the sample compartment.

To select or clear a single well, simply click or right-click on it.

To select or clear a block of wells, hold down the cursor and drag around the block required. All wells within the area will be selected or cleared.

| X, Y, Well | Displays the Well Position that the mouse cursor is pointing to. This field is not editable. |
| Reset | Select Reset to move the Wellplate Reader to the first well position, which is well A1. |
| Park | Select Park to move the Wellplate Reader to the Park position. When the accessory is in the park position you have full access to the Wellplate Reader platform. This is useful for inserting or removing wellplates from the accessory. |

See also:
- Wellplate Accessories page
- About the Wellplate Reader

**Fast Filter accessory—Ratio**

If you are using the Fast Filter accessory, this page enables you to setup the filters. The Fast Filter accessory page is displayed when you select the Fast Filter tab on the Accessories page.

The page comprises the following:

**Fast Filter**

<table>
<thead>
<tr>
<th>Result 1 or Result 2</th>
</tr>
</thead>
</table>

**Note**

These options are only available if you have set the Collection type on the Cary page to Excitation fast filter or Emission fast filter.

The Result and Filter options reflect the setup of the fast filter accessory and are used to indicate what positions the filters have been placed in. Once you have set up your fast filter accessory, you must indicate the positions of the filters using these options.

**Note**

When placing filters in the fast filter accessory, you must place them in opposite positions (e.g., Positions 1 and 3 or Positions 2 and 4) so that the fast filter accessory does not become unbalanced. The filters must also be the same dimensions and of similar mass.

Black discs supplied with the accessory must also be placed in the filter positions which are not in use to ensure the filter wheel is adequately balanced. For example if filters are placed in positions 1 and 3, black discs must be placed in positions 2 and 4.

Dots on the accessory indicate the position number (refer adjacent diagram).
If you have filters in positions 1 and 3:

Select the **Result 1** option to calculate and display the intensity of Filter 1 and Filter 3. You can then choose to determine the ratio of Filter 1:Filter 3 or the ratio of Filter 3:Filter 1 by selecting the appropriate filters in the adjacent fields. For example, to determine the ratio of Filter 3:Filter 1, select **Filter 3** in the first field. **Filter 1** will then be automatically selected in the second field.

If you have filters in positions 2 and 4:

Select the **Result 2** option to calculate and display the intensity of Filter 2 and Filter 4. You can then choose to determine the ratio of Filter 2:Filter 4 or the ratio of Filter 4:Filter 2 by selecting the appropriate filters in the adjacent fields. For example, to determine the ratio of Filter 4:Filter 2, select **Filter 4** in the first field. **Filter 2** will then be automatically selected in the second field.

**Position**

*Note*

This option is only available if you have set the Collection type on the Cary page **Excitation fast filter** or **Emission fast filter** AND the **Result 1** and **Result 2** options are not selected.

Select this to specify the filter position at which data collection at a single wavelength (specified on the Cary page) will be performed. The options are: **Park**, **Filter 1**, **Filter 2**, **Filter 3** or **Filter 4**. If you select the **Park** position, the accessory will be rotated 45° from position 1 (to the 'Flag' position shown in the above diagram), ensuring that a beam does not pass through any of the four positions. The fast filter accessory will drive to the selected position when you close the Setup dialog.

**Filter 1/ Filter 2/ Filter 3/ Filter 4**

These fields enable you to enter information about the types of filters you are using in the fast filter accessory. This information is for your reference only. It will not be used in any software calculations. The following information can be entered:

- **Name**
  
  Enter the name of the filter used.

- **Wavelength**

  Enter the wavelength of the filter.

**Status Display**

Select this check box to display the **Status Display window**. Use this window to view the status of various parameters during the data collection.

See also:

- Cary tab
- Accessories
- Multi Cell Changer
- Status Display

**Analyze—Ratio**

Use the Analyze page to select from various mathematical equations and set the parameters used to generate
the calculation of reaction rates.

From the Setup menu, select Analyze to access the Analyze page. You can also access the page from the Setup dialog by selecting the Analyze tab. To access the Setup dialog, select the Setup button from the Ratio Application window.

**Subtract autofluorescence**

Select this option if you wish to remove the intrinsic fluorescence value associated with cells from your ratioed trace.

In the **Enter value to subtract from trace** field, enter the fluorescence value that you wish to be subtracted from your post reaction ratioed trace. Selecting this option disables both the Dual wavelength analysis and Single wavelength analysis options.

**Dual wavelength analysis**

Select this option when performing ratio calculations for wavelength shifting dyes and when you are required to monitor two wavelength pairs.

**Enter Rmin and Rmax manually:**
Select this option if you wish to determine the concentration value by entering previously determined Rmin, Rmax, Sf(λ₂), Sb(λ₂) and Kd values directly into the Ratio software.

**Determine Rmin and Rmax from the graph:**
Select this option if you require the Rmin, Rmax, Sf(λ₂), Sb(λ₂) values to be calculated from the graph.

For more information on the ratio calculations see the Grynkiewitz equation.

**Single wavelength analysis**

Select this option when performing calculations for non-wavelength shifting dyes.

**Enter Fmin and Fmax manually**
Select this option if you wish to manually enter the Fmin and Fmax values for the Grynkiewitz equation.

**Determine Fmin and Fmax from graph**
Select this option if you require the Fmin and Fmax values to be calculated from the graph.

**Q Why are the Subtract autofluorescence, Dual wavelength analysis or Single wavelength analysis options not available?**

**A** If one of the above options are not available deselect the option currently in use to make each of the options available.

**Status Display**

Select this check box to display the Status Display window. This window allows you to view the status of various parameters during the data collection.
How to obtain a ratioed trace—Ratio

To determine the true fluorescence value of a reaction it is necessary to subtract the autofluorescence of the reaction. That is it is necessary to subtract the intrinsic cell fluorescence from the overall calculated fluorescence of the reaction.

Step 1

Using the Trace Preferences option display the two traces with the autofluorescence subtracted as two separate graphs.

Select the individual traces by clicking in the Visible column next to the trace.

Step 2

Return to the main graphics area and select a trace by clicking in the graphics area.

Step 3

Using the calculator button in the toolbar, open the Math's dialog box.

Step 4

Enter your first trace in the Calculation entry field by using the Selected Trace button.

Step 5

Divide the Selected Trace by the Focused Trace by simply selecting the divide option and then using the Focused Trace button.

Step 6

Under Trace select Ratio mode from the operation types. Click Apply and then equals.

Step 7

A ratioed trace is will now be visible in the graphics window and will be named after the two initial traces with _AF1 ending the name.

**Note**
The ratioed trace has not been saved. Save the new trace as a batch file.

To obtain a concentration graph from the ratioed trace it is necessary to use the Grynkiewitz equation.

Step 8

Select the Recalculate button in the bottom left hand corner of the application. On the Analyze tab, select the
Dual Wavelength checkbox.

There are two ways of performing the Grynkiewitz equation:

1. **By manually entering the required data**
   
   **Note**
   
   If the reaction has been performed previously and is relatively stable it is possible to use the Rmin and Rmax values previously calculated.

2. **Gathering the required data from the graph**
   
   **Note**
   
   If you are performing the reaction for the first time, or the reaction is unstable and variable, the Rmin and Rmax should be determined from the graph.

**Status Display**

Select this check box to display the Status Display window. This window allows you to view the status of various parameters during the data collection.

See also:

- How to obtain an autofluorescence value
- How to subtract autofluorescence
- Manually entering data in the Grynkiewitz equation
- Gathering the required data from the graph for the Grynkiewitz equation
- More information on the Grynkiewitz equation

**Reports—Ratio**

Use the Reports page to set up the report parameters associated with data collection.

From the Setup menu, select Reports to access the Reports page. You can also access the page from the Setup dialog by selecting the Reports tab. To access the Setup dialog, select the Setup button from the Ratio Application window.

**Hot Tip**

You can view your generated report before you print it out using the Print Preview command in the File menu.

If you decide to make changes to the report layout, select (Edit Report) from the Report toolbar. This enables you to directly edit the report.

The page comprises the following:

**Operator**

**Name** Enter the instrument operator name in this text entry field. Use the arrow to the right of this field to view and select one of the last ten entries in this field.
Options

This group enables you to select the information to include in your report.

**Auto Print**
Select **Auto Print** to print a report automatically during the data collection. If **Auto Print** is selected, then the system will send the report information to the specified printer(s) as well as the Report area. However if **Auto Print** is not selected, the report will only be sent to the Report area and can be viewed by selecting **Report** in the **View** menu.

To automatically obtain a copy of the graphics, you will need to select both the **Auto Print** and **Graph** options (see below). In this case, all graphical information displayed on screen will be printed with the report. Therefore it is advisable to arrange the graphical information as you require on screen, prior to printing. For example, if you require a single graph print out, then the **Single/Multi Graph** option in the **Graph** menu should be changed to the correct setting.

**Parameters**
Select this option to include the method setup parameters within your report.

**User data form**
Select this option to include in the report any information entered in the **User Data Form**.

**Graph**
Select **Graph** to include any graphs or graphic information associated with your report. If you require a printout of the displayed graphics only, you must ensure that **only** this option is selected. Select the **Clear Report** button to clear any information in the Report area before you print otherwise this will be included in your printout.

**% Page Height**
Enter the size at which you want the graph to be printed, as a percentage of total page height. For example, if you want graphical information to take up half of the page, enter 50% in this field.

**Company Logo**
Check this option to include your company logo in the report. A company logo will only be included if you have added one on the Company page of the System Information Application.

**Hot Tip**
If you have not included a Company logo, select the Header/Footer button to open the System Information Application and use the Add Logo option to add a logo.

**Header/Footer**
Select the **Header/Footer** button to open the System Information application where you can enter the Header and Footer information to be included in your report. You can also enter any additional comments, and add a logo to your report. Once you have made all your changes, select OK to exit the System Information application and return to the Reports page of the Ratio application.

**Focused trace**
Select this option to apply the **Report** selections to the focused trace only.

**All traces**
Select this option to apply the **Report** selections to all visible traces.

**X-Y Pairs Table**
Select this if you want to print a table showing the X-Y pairs of the collected data in your report.

**Data Interval**  
*Actual*  
Select *Actual* to report the X-Y pairs table for the trace at the actual **Data Interval** at which the data was collected.

*Interpolated*  
Select *Interpolated* to report the X-Y pairs table for the trace at an interpolated data interval. Enter this data interval in the adjacent field. If no data has been collected at the interval you enter, the Cary Eclipse software will interpolate the data to produce an X-Y result.

**Status Display**

Select this check box to display the **Status Display** window, where you can view the status of your ratio run during the data collection.

**Hot Tip**  
You can copy a picture, spreadsheet file or document from another application (for example, Microsoft Word or Microsoft Excel) into the report area of the Lifetimes application. To edit these objects, simply double click on them to open the associated application in which they were created.

See also:  
- User Data Form  
- View menu  
- Status Display window

**Report Toolbar—Ratio**

Use the Report toolbar buttons to gain quick access to many of the **Edit** menu options. You can also use the toolbar to cut, copy and paste information and to change the look of your report.

To display or hide the toolbar, point to the **Toolbars** option in the **View** menu and select **Reports**. To display or hide the toolbar's text labels, point to the **Toolbar** option in the **View** menu and select **Text Labels**.

**Hot Tip**  
To see the name of a button, point to it with the mouse.

The following commands can be accessed from the Reports Toolbar:

<table>
<thead>
<tr>
<th>Button</th>
<th>Menu command</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>![ ]</td>
<td><strong>Edit Report</strong></td>
<td>Click this button to edit the report. This command is also accessible by pressing CTRL+R.</td>
</tr>
<tr>
<td>![ ]</td>
<td><strong>Undo</strong></td>
<td>If available, this undoes the last action. This command is also accessible by pressing CTRL+Z.</td>
</tr>
<tr>
<td>![ ]</td>
<td><strong>Cut</strong></td>
<td>Click this button to remove the highlighted text and place it on the Windows Clipboard. This command is also accessible by pressing CTRL+X.</td>
</tr>
<tr>
<td>![ ]</td>
<td><strong>Copy</strong></td>
<td>Click this button to copy the highlighted text onto the Windows Clipboard.</td>
</tr>
</tbody>
</table>
Clear Report—Ratio

Select the Clear Report command to remove the contents of the Report Area in the Ratio Application window. If you have saved the report, selecting this option does not delete the contents of the report, the information is simply removed from the Report Area.

To access the Clear Report option, select the Clear Report command from the Commands menu, or select the Clear Report button from the Ratio Application window.

See also:
  - Ratio Application
  - Commands menu

How to use advanced reporting options—Ratio

This outlines ways to fully customize your reports. Options available include:

- Including header and footer information
- Including a logo
- Editing the text in the report
- Viewing the report on screen before printing
- Cutting and pasting to a word processor

Including header and footer information

You can have text appearing at the top and bottom of every page in your report. To specify what the text should be, do the following:

Step 1

On the Windows Taskbar, click the Windows Start button, point to Programs, Cary Eclipse and then select System Information. The System Information application appears with the Company page displayed.

Step 2
Enter the header and footer information in the **Header and Footer** fields.

Step 3

Click **OK** to implement your changes.

**Including a logo**

You can include a logo on the first page of your report. To specify what the logo should be, do the following:

Step 1

On the Windows Taskbar, click the Windows **Start** button, point to **Programs, Cary Eclipse** and then select **System Information**. The System Information application appears with the Company page displayed.

Step 2

Enter a logo using the **Add Logo** button. This logo will always be included on the front page of reports.

Step 3

Click **OK** to implement your changes.

**Q.** Why isn't the logo I selected included in my report?

**A.** If you are running the Cary Eclipse software under Windows NT you need to be logged on as an Administrator in order to specify the logo in the System Information application.

**Editing the text in the report**

You can edit any of the text in the Report area. Just select **Edit Report** from the **Edit** menu. You can then add, delete and change text in the Report area.

**Q.** The Edit Report option is not available, why not?

**A.** You do not have reporting editing privileges set in the GLP Administration application. The Administrator of the system will have to change your privileges before you can edit reports.

**Viewing the report on the screen before printing**

You can view a report before printing it. To do this, from the **File** menu select **Print Preview**. You can then ensure that the report format is correct before you print it.

**Hot Tip**

Use the arrow keys at the bottom of the Print Preview screen to change pages.

**Cutting and pasting to a word processor**

To have complete control over how your report looks you may wish to copy it to a word processor program, such as Microsoft Word. To do this:

Step 1

Open a report.
Step 2

From the **Edit** menu, choose **Edit Report**.

Step 3

Click in the Report area and highlight the text you want to export to your word processor.

Step 4

Right-click on the Report area and select **Copy** from the menu.

Step 5

Start up your word processor and use the Paste function (usually under the **Edit** menu) to place the text into a document.

**Auto Store—Ratio**

Use the **Auto Store** page to specify if and when data will be stored, and in what format. You can also use this page to set up the system to automatically send an email at the end of a read.

From the **Setup** menu, select **Auto store** to access the **Auto store** page. You can also access the page from the **Setup** dialog by selecting the **Auto store** tab. To access the **Setup** dialog, select the **Setup** button from the Ratio Application window.

The page comprises the following:

**Storage**

- **Off**
  - Select this if you do not wish to be automatically prompted to save collected data.
  - **Note**
    - If you wish to store your unsaved data manually, choose Save Data As… from the File Menu to save your data after the run is complete. You can save your data as a Batch or Data file.

- **On; Prompt At Start**
  - Select this to display the Windows **Save As** dialog at the start of the collect where you can enter the Batch filename for your data.

- **On; Prompt At End**
  - Select this to display the Windows **Save As** dialog at the end of a collect where you can enter the Batch filename for your data.

**Auto convert**

Use the Auto convert options to convert your data into an ASCII file format that can be opened in spreadsheet programs such as Microsoft Excel.

- **None**
  - Select **None** if you do not need to generate an ASCII file.

- **ASCII (csv)**
  - Select this option to save the data as a comma delimited ASCII Spreadsheet file (*.CSV) file. Only the currently collected data is converted.
ASCII files are stored in the same folder as the data file from which they were generated and designated the same filename as the method used to collect the data plus the extension ‘CSV’.

**ASCII (csv) with log**

Select this option to save the data as a comma delimited ASCII Spreadsheet file (*.CSV) file. Only the currently collected data is converted. The key method parameters used to collect the data are also saved in this file.

ASCII files are stored in the same folder as the data file from which they were generated and designated the same filename as the method used to collect the data plus the extension ‘CSV’.

**Note**

If you choose to auto-convert the data to an ASCII file but you have set the Storage option to Off, then the ASCII file will be saved with the following format:

DATE TIME. CSV

The date and time will appear in the same form that is set in the Windows Regional Settings Properties dialog that is accessed from the Windows Control Panel.

For example, 22_4_97 1;32;08.CSV

**Email**

Select the Email check box and enter the Email details to send an email to a person once the read is completed. This useful facility can be used to alert you to the completion of the read, particularly if you are working away from the laboratory where the Cary Eclipse instrument is. You can also attach the collected data to the email.

**Note**

You must set up the email Host and Port options on the Email options page of the System Information application in order for the email facility to work correctly.

**To**  Enter the recipient's email address. You can enter more than one recipient in this field.

**From**  Enter your email address (or the email address associated with the computer on which the Cary Eclipse software is installed).

**Subject**  Enter the subject of your email (or leave this field blank if you do not wish to include a subject).

**Cc**  Enter the email address of any recipient you want to copy the email to.

**Message**  Enter your message.

**Attached…**  Use the Attached button include a file attachment in your email. To do this, select Attached and browse your computer to select a file, using the Windows® Open dialog that appears.

**Attach current data**  Select this option to attach the data that has been collected during the run to the email.

**How to email your results**

**Status display**

Select this check box to display the Status Display window, where you can view the status of your ratio run during the data collection.
How to email your results—Ratio

You may wish to send an email to a person once the read is completed. This useful facility can be used to alert you to the completion of the read, particularly if you are working away from the laboratory where the Cary Eclipse instrument is.

To do this:

Step 1

Ensure that you have set up the **Host** and **Port** options on the **Email options** page of the System Information application in order for the email facility to work correctly.

Step 2

From the **Setup** menu, select **Auto Store**. The **Auto Store** page is displayed.

Step 3

Select the **Email** check box to enable the email options.

Step 4

In the **To** field, enter the recipient's email address (for example, rchandle@varianinc.com). To enter multiple recipients, simply type a comma ',' between each email address.

Step 5

In the **From** field, enter the email address of the computer on which the Cary Eclipse software is installed. This may be your email address, or a communal email address, if the Cary Eclipse software is installed on a computer that has multiple users.

Step 6

Enter information in the **Subject** and **Message** fields as required.

Step 7

Select **Attach current data**. The data that has been collected during the run will be attached to the email.

**Hot Tip**

You can attach other documents to your email. To do this, select the **Attached** button and browse your computer to select a file, using the Windows® **Open** dialog that is displayed.

Step 8

Select **OK** to clear the **Auto Store** dialog. Your email will be sent once the run has been completed.
Performing scans—Ratio

- Starting a run
- Sample Name dialog
- Cell Loading Guide
- Sync Start dialog
- Start/Stop buttons
- Stop Time dialog
- Go To dialog
- Go To Temperature dialog
- Rapid Result dialog
- Commands menu
- How to perform a single-wavelength ratio run
- How to perform a multi-wavelength, temperature controlled ratio run

Information about starting a collect—Ratio

The sequence that occurs when you select Start to commence the ratio run depends upon your storage and accessory selections.

Storage selections

<table>
<thead>
<tr>
<th>If you have selected … as the Storage option on the Auto Store page</th>
<th>The following occurs</th>
</tr>
</thead>
<tbody>
<tr>
<td>On, prompt at start</td>
<td>The Windows Save As dialog will appear as soon as you select Start. You can save the current method, collected data, report and graph template as a Batch file.</td>
</tr>
<tr>
<td>On, prompt at end</td>
<td>The Windows Save As dialog will appear at the end of the data collection. You can save the current method, collected data, report and graph template as a Batch file.</td>
</tr>
<tr>
<td>Off</td>
<td>The Windows Save As dialog will not appear.</td>
</tr>
</tbody>
</table>

Accessory selections

<table>
<thead>
<tr>
<th>If you have selected … on the Accessories page</th>
<th>The following dialog is displayed</th>
</tr>
</thead>
<tbody>
<tr>
<td>No accessories</td>
<td>The Sample Name dialog will appear when you select Start, prompting you to load the sample and enter your sample name. Select OK to display the Sync Start dialog and commence the ratio run.</td>
</tr>
<tr>
<td>Multicell Holder</td>
<td>The Cell Loading Guide dialog will appear. Enter the sample name of each activated cell then select OK to display the Sync Start dialog. When you select OK to clear the Cell Loading Guide dialog, the cell</td>
</tr>
</tbody>
</table>
### Sample Name—Ratio

If you are using a single cell holder, the Sample Name dialog appears once you select the **Start** button to commence an analysis.

The page comprises the following:

**Sample Name**  Enter the name of the sample to be used in the ratio run. Click on the arrow to the right of the field to view and select the most recently entered sample names.

Once you have entered the **Sample Name**, select **OK** to display the **Sync Start** dialog and commence the ratio run.

See also:
- **Cell Loading Guide**
- **Sync Start**
- **Start/Stop buttons**
- **Setup**
- **Accessories**
- **Auto Store**

### Cell Loading Guide—Ratio

If you are using a Multicell Holder, the **Cell Loading Guide** dialog appears once you select the **Start** button to commence an analysis.

The **Cell Loading Guide** dialog prompts you to insert blank samples in the Multicell Holder to collect the Ratio scans. You can also use the dialog to name the samples in each selected cell in the Multicell Holder.

The dialog comprises the following:

---

<table>
<thead>
<tr>
<th>Wellplate Reader</th>
<th>The <strong>Sync Start</strong> dialog will appear after you press <strong>Start</strong>. The sample labels and the positions in the wellplate that you wish to use have been set up before you press <strong>Start</strong>.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fast Filter</td>
<td></td>
</tr>
</tbody>
</table>
Loading Guide/Sample Name entry

Enter the names of the sample to be used in the ratio run in each available field. This sample name will be used as the Name of Trace in the Trace Preferences dialog.

Note
The Cell Loading Guide dialog appears only if you have selected the Multicell Holder option on the Accessories page of the Setup dialog. Only cells selected in the Multicell Holder diagram on the Accessories page will be available in the Loading Guide.

In the adjacent example, cells 1—3 will be available in the Loading Guide dialog, as they have been selected in the Multicell Holder diagram.

Once you have entered the Sample Name, select OK to display the Sync Start dialog and commence the Ratio run.

See also:
Accessories
Sample Name

Sync Start—Ratio

Use the Sync Start dialog to synchronize the start of a reaction and the start of the ratio data collection. Before displaying the Sync Start dialog, the Cary Eclipse will set up all the instrument and graphing parameters so that data collection can start as soon as the countdown finishes or you select OK.

The sequence for a Sync Start is as follows:

- Set up your data collection parameters
- Click the Start button (and load the samples if necessary)
- Once the Sync Start dialog is displayed, add your active reagent
- Commence the data collection by selecting the OK button

The dialog comprises the following:

Count Down
This field displays a two minute countdown in seconds. If you want to start your data collection before the two minute countdown ends select the OK button. Alternatively, you can wait for the countdown to end and the data collection will start automatically.

Note
If you are using a Multicell Holder and you select the OK button, the system will automatically Reset this accessory and immediately start the ratio run.

Reset Slide
Click this button to drive the Multicell Holder to its zero point (Cell position 1).

Start/Stop Instrument buttons—Ratio

The following buttons may be displayed at the top of the Ratio Application window. The Start, Pause, Continue and Connect buttons will change depending upon the status of the run. For example, if a run has not been started, the Start button will be visible to indicate that the system is ready to commence a run.

Select Start to commence a ratio run using the current Setup parameters. If any unsaved data exists in the Graphics area you will be prompted to save the
data. If you choose not to save, any unsaved data, pictures and labels will be deleted when you start the ratio run.

When a ratio run is started, the **Start** button will change to **Pause**.

The **Pause** button is displayed in place of the **Start** button once you have selected **Start** to commence a run. Click **Pause** to temporarily suspend the data collection, so that you can modify your solutions during the ratio run. While the system is in the paused state, it will continue to keep track of the elapsed time, however data collection will stop until the **Continue** button is selected.

The benefit of using **Pause** instead of **Stop** is that you are able to add a secondary substrate to your ratio solution(s) and then continue to monitor a secondary reaction, without having to Stop and Start the collect again. It also means that you will not get a spike in your data when you open the sample compartment to add the reagent.

When a ratio run is paused, the **Pause** button will change to **Continue**.

The **Continue** button is displayed in place of the **Pause** button when you have paused the run. Click **Continue** to proceed with the ratio run after it has been paused.

Once you select **Continue**, is selected the button changes back to **Pause**.

If another application is already online with the Cary Eclipse, the **Start** button will change to **Connect**, and you will need to select **Connect** to bring this application online before the **Start** button will re-appear. (You will not be able to Connect if the other application is collecting data.)

Select this to end a ratio run.

If you have selected **Storage On** in the **Auto Store** page of the **Setup** dialog, all data collected is automatically stored in the file specified in the Batch name field of the Windows **Save As** dialog before or after the run commenced.

---

**Zero—Ratio**

Use the **Zero** button to zero the current intensity value. The intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before commencing a data collection if you want your data to start at 0 intensity. The Cary Eclipse will prompt you to load a blank before performing the zero.

Before you select **Zero**, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.

When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the Ratio Application window.

Select the **Clear Zero** command from the **Commands** menu to remove zero correction and thus remove the subtraction effect.

To access the **Zero** option, select the **Zero** command from the **Commands** menu, or select the...
Stop Time—Ratio

Use the Stop Time dialog to extend the length of a ratio run while the run is in progress. Extending a run may be necessary if you decide whilst a run is in progress that the data collection period is too short. The Stop Time command is not available until a run is in progress (that is, the Start button is selected).

To extend the length of a ratio run in progress:

1. Select the Stop time option from the Ratio Application window or the Commands menu. The Stop Time dialog is displayed.

2. In the Stop Time field, the minimum allowable Stop Time will be displayed. This is the Stop Time that has been specified on the Cary page of the Setup dialog. In the Stop Time field, enter the new Stop Time and select OK. The X axis (Time) of the graph in the Graphics Area will be updated to reflect the new setting.

Go to Wavelength—Ratio

Use the Go to Wavelength dialog to temporarily change the instrument to a new excitation or emission wavelength. You would mainly use this dialog if you want to quickly go to a wavelength and check the intensity reading of the sample. The lamp will flash continuously in this mode. This option is only available if the instrument is not currently collecting data.

To access the Go to Wavelength dialog, select the Go To Wavelength command from the Commands menu. You can also select F4 to open the dialog.

Note
This is not the wavelength used in the application method. If your method uses a wavelength that is different from the 'Go to wavelength' setting you select, then when you select Start the system will change to the wavelength selected. (In most applications, this is set in the Cary page of the Setup dialog.)

The Go to Wavelength page comprises the following:

Excitation

Enter the new wavelength in this numeric entry field. When you select Go to the instrument will change to the new value.
Zero Order
Select Zero Order to drive the Cary Eclipse fluorescence spectrophotometer to 0 nm (white light). You can then use this light for aligning accessories.

Emission

Wavelength
Enter the new wavelength in this numeric entry field. When you select Go to the instrument will change to the new value.

Zero Order
Select Zero Order to drive the Cary Eclipse fluorescence spectrophotometer to 0 nm (white light). You can then use this light for aligning accessories.

Go To
Click Go to once you have entered a new wavelength, to change the instrument to the entered value.

See also:
Commands menu
Function Keys

Go to Temperature—Ratio

Use the Go to Temperature dialog to change the current temperature to a new value.

To access the Go to Temperature dialog, select the Set temp button from the Ratio Application window or the Set temp command from the Commands menu.

The Set Temp button is only accessible if you are using a thermostatted accessory. (On the Accessories page, the Temperature control option must be selected.)

You can access the Go to Temperature dialog prior to starting a run or once a run is in progress.

To change the current temperature of a run in progress:

1. Access the Set Temp button by selecting the Pause button to pause the run. You can then select the Set Temp button.

2. In the Set temp field, enter the temperature you want the instrument to change to. Select OK to return to the Ratio Application window.

3. Select Continue to proceed with the run.

See also:
Ratio Application

Rapid Result—Ratio

Use the Rapid Result dialog to take a reading of a blank and sample/s, without having to change the current application method. You can use the Multi Cell Changer dialog to specify the cell you want to read. This means that between runs, while new samples are being prepared and loaded, other samples can quickly be measured. The result is printed in the Report area.

To access the Rapid Result dialog, select the Rapid Result command from the Commands menu or press F6.

Excitation

Wavelength
Enter the wavelength at which you want to take a reading.
**Zero order**
Select this option to set the excitation monochromator to zero order (white light).

**Emission**
**Wavelength**
Enter the wavelength at which you want to take a reading.

**Zero order**
Select this option to set the excitation monochromator to zero order (white light).

Zero
Click **Zero** to change the wavelength to the entered value and measure a blank.

Result
Click **Result** to change the wavelength to the entered value and measure a sample. The X,Y results are corrected for the blank and the results are displayed in the Report window.

See also:
- Multi Cell Changer
- Commands menu

**Commands menu—Ratio**

Use the **Commands** menu to access major operational commands in the Ratio application. The menu is accessible from the menu line at the top of the Ratio Application window.

Alternatively you can use function key shortcuts for some of the commands. These appear next to the item in the **Commands** menu. Note that not all commands appear as both button and menu items.

**Hot Tip**
If you access these commands using the **Commands** menu, rather than from the application buttons, you can remove these buttons from your application window. To do this, clear the **Buttons** command in the **View** menu. This will enlarge the Graphics area, making viewing and selection of scans easier.

Tell me how to remove or display the command buttons

The **Commands** menu comprises the following commands:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Start</td>
<td>Select this to start a data collection using the currently set up parameters.</td>
</tr>
<tr>
<td></td>
<td>If the <strong>Connect</strong> command is displayed in place of the <strong>Start</strong> command, this indicates that another</td>
</tr>
<tr>
<td></td>
<td>Cary Eclipse application is in use and has control of the Cary Eclipse instrument. To make the Ratio</td>
</tr>
<tr>
<td></td>
<td>application active so that you can perform a read, you will need to select <strong>Connect</strong>. However, you</td>
</tr>
<tr>
<td></td>
<td>will not be able to select <strong>Connect</strong> if another application is collecting data.</td>
</tr>
<tr>
<td>Stop</td>
<td>Select this to stop the data collection.</td>
</tr>
<tr>
<td></td>
<td>If you have selected Storage On in the <strong>Auto Store</strong> page of the <strong>Setup</strong> dialog, all data collected</td>
</tr>
<tr>
<td></td>
<td>is automatically stored in the file specified in the <strong>Batch name</strong> field of the Windows <strong>Save As</strong></td>
</tr>
<tr>
<td></td>
<td>dialog before or after the run commenced.</td>
</tr>
<tr>
<td>Clear Zero</td>
<td>Select this option to clear the <strong>Zero</strong> command. When you select the <strong>Zero</strong> command (from the</td>
</tr>
<tr>
<td></td>
<td><strong>Commands</strong> menu or by pressing F5), the intensity at that time will be reset to zero and all</td>
</tr>
<tr>
<td></td>
<td>subsequent results will have the intensity value that was reset to zero subtracted from them.</td>
</tr>
<tr>
<td></td>
<td>Selecting <strong>Clear zero</strong> cancels this zeroing (and thus subtraction) effect.</td>
</tr>
<tr>
<td>Pre-scan</td>
<td>Select <strong>Pre-scan</strong> to quickly determine the optimum instrument parameters to be used for your sample.</td>
</tr>
<tr>
<td></td>
<td>At the end of this collection sequence the software will report the optimum Ex and Em wavelengths and</td>
</tr>
<tr>
<td></td>
<td>PMT voltage. Once you are satisfied with the parameters, the software</td>
</tr>
</tbody>
</table>
Zero order… Select the **Zero order** command to set one of the monochromators to zero order. If you use this command to zero either the excitation or emission monochromators, this will override the **Ex. Wavelength** or **Em. Wavelength** set on the Cary page. The Cary Eclipse software will use the zero setting when collecting data.

The **Zero order** command is generally used during alignment procedures.

**Excitation**
Select this command to set the excitation monochromator to zero order (white light).

**Emission**
Select this command to set the emission monochromator to zero order (white light).

**Reset**
Select this command to re-initialize the instrument. This will perform the same initializing procedure that is performed when the instrument is powered on.

**Rapid Result**
Select this to display the **Rapid Result** dialog. Use this dialog to take a reading of a blank and multiple sample readings without changing the application method. This means that between runs, while new samples are being prepared and loaded, other samples can quickly be measured.

This command can also be accessed by pressing F6.

**Zero**
Select this to zero the current intensity value. The intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before selecting **Start** if you want your data to start at 0 intensity.

This command is also accessible by pressing F5.

**Note**
Before you select **Zero**, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.

**Go to…**
Select **Go To…** to display the **Go To Wavelength** dialog where you can change the instrument to a new wavelength. Use this command if you want to quickly go to a wavelength to check the intensity reading.

This command is only available if the instrument is not currently collecting data. It can also be accessed by pressing F4.

**Note**
This is not the wavelength used in the application method. If your method uses a wavelength that is different from the 'Go To wavelength', then when you select **Start** the system will change to the wavelength selected. (In most applications, this is set in the Cary page of the Setup dialog.)

**Set temp**
Select **Set temp** to open the **Set Temperature** dialog where you can change the temperature during a ratio run.

The **Set temp** command button is only accessible if you are using a thermostatted accessory. (On the Accessories page, the Temperature control option must be selected.) Once you have started a run, select the **Pause** button to pause the run and then select the **Set temp** command.

**Stop time**
Select the **Stop Time** command to open the **Stop Time** dialog which you can use to extend the length of a ratio run while the run is in progress. Extending a run may be necessary if you decide whilst a run is in progress that the data collection period is too short. The **Stop Time** command is not available until a run is in progress (that is, the **Start** button is selected).

**ADL**
Select this to display the **ADL Command Line** window where you can execute an ADL
Viewing run progress and results—Ratio

- Reports page
- Reports toolbar
- View menu
- Edit menu
- Status display
- Print Preview dialog
- Print

View menu—Ratio

The View menu is accessible from the menu line at the top of the Ratio Application window.

**Hot Tip**
Highlight a method or data file in Explorer and simply drag and drop it onto the application to open it. Any data will be displayed in the Graphics area.

The following commands can be accessed from the View menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Toolbar</td>
<td>Select this option to toggle the Graphics and Reports toolbar on and off. You can also select to display or hide the toolbar Text Labels.</td>
</tr>
<tr>
<td>Buttons</td>
<td>Select this option to toggle the left-hand button display (button area) on and off.</td>
</tr>
<tr>
<td>Graphics</td>
<td>Select this option to toggle the Graphics area on and off.</td>
</tr>
<tr>
<td>Report</td>
<td>Select this option to toggle the Report area on and off.</td>
</tr>
<tr>
<td>Report Vertical</td>
<td>If the Graphics option is selected, select Report Vertical to display the Graphics area, Report area and/or ADL Editor side by side vertically.</td>
</tr>
<tr>
<td>ADL</td>
<td>Select this option to toggle the ADL Editor on and off.</td>
</tr>
<tr>
<td>ADL Vertical</td>
<td>If the ADL and Report options are selected, select ADL Vertical to display the ADL Editor vertically on the right hand side of the Report area.</td>
</tr>
</tbody>
</table>
This displays the **Quick Reference** window where you can easily add a command in the correct syntax to your ADL program. A detailed description of all the commands in the Quick Reference is available by selecting the Help button.

**ADL Hooks**

Select this to activate a link (or hook) to an ADL program at a specific point in the application, and then run that ADL program. For example, ADL hooks have been supplied to run specific programs:

- on start-up of the Cary Eclipse software;
- when the system goes online or offline;
- when the Start button is selected;
- when a run finishes;
- when a zero is performed etc.

For the hook to work, **ADL Hooks** must be selected, and the appropriate ADL file for the application must be present in the ADL folder in the directory.

**Note**

Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.

**Hints**

Select this option to toggle the Hints bubble text on and off. The bubble text appears when you place the arrow cursor over a field, button or text entry field for a short period. It offers a quick explanation of the item’s function.

**Status Display**

Select this option to display the **Status Display** window, where you can view the status of various parameters during the data collection.

---

**Edit menu—Ratio**

Use the **Edit** menu to copy, cut, paste and delete information. You can also use the menu to access the Report editing commands. The menu is accessible from the menu line at the top of the Ratio Application window.

The following commands can be accessed from the **Edit** menu:

**Note**

Some options are only available once you select **Edit Report**.

<table>
<thead>
<tr>
<th>Command</th>
<th>Button</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Undo</td>
<td>🔄️</td>
<td>If available, this undoes the last action. This command is also accessible by pressing CTRL+Z.</td>
</tr>
<tr>
<td>Cut</td>
<td>🔁</td>
<td>Select this command to remove the highlighted text and place it on the Windows Clipboard. This command is also accessible by pressing CTRL+X.</td>
</tr>
<tr>
<td>Copy</td>
<td></td>
<td>Select this command to copy the highlighted text onto the Windows Clipboard.</td>
</tr>
</tbody>
</table>
Use the Status Display window to view the status of your ratio run during the data collection. Depending upon settings you have for the Ratio run, the following parameters may be displayed:

- Current Excitation Wavelength (nm)
- Current Emission Wavelength (nm)
- Excitation Slit Width (nm)
- Emission Slit Width
- PMT Voltage (V)

To access the Status Display window, select Status Display from the View menu, or from any page of the Setup dialog. You can choose to view the Status Display at any time during a ratio run.

See also:
- View menu
- Setup
- Status Line
Print Preview—Ratio

Use the Print Preview dialog to view the current report before you print it. From the File menu, select Print Preview to access the dialog.

The contents of the Graphics area appear first in the preview area followed by the contents of the Report area. You can reduce or enlarge the print preview on screen. To do this, click the left mouse button in the preview area to toggle through the five available views. Click the right mouse button in the preview area to return to the full page view.

The page comprises the following commands:

- **First arrow** [↩] Use this arrow to view the first page of the report.
- **Previous arrow** [↩] Use this arrow to view the previous page of the report.
- **Next arrow** [↩] Use this arrow to view the next page of the report.
- **Last arrow** [↩] Use this arrow to view the last page of the report.

Q. Why are the arrow buttons grayed out?

A. The buttons will be grayed out if the report has only one page of data.

**Print** [_printer_] Use this to display the Windows Print dialog where you can choose the printer and pages to print as well as the number of copies you wish to print.

See also:
- File menu

Print—Ratio

Use the Print dialog to select the printer and pages to print of the currently displayed report as well as the number of copies you wish to print.

To access the Print option, select the Print command from the File menu, or select the Print button from the Ratio Application window.

All graphical information displayed on screen will be printed with the report, therefore it is advisable to edit the report on the screen prior to printing. You can also preview the report prior to printing by using the Print Preview command.

If you require a printout of the graphics in the Graphics area only, select the Clear Report button to erase the contents of the Report area before you print.

**Note**
The options on the Report page are used to generate the contents of the Report area during a run or on a recalculation. If Auto Print has been selected the Cary Eclipse will automatically print a report at the end of a data collection.

See also:
- Print Preview
- Report page
- Edit Report
Performing Calculations—Ratio

- Recalculate
- How to generate a report from previously stored data
- How to use advanced reporting options
- How to calculate autofluorescence
- How to subtract autofluorescence
- How to obtain a ratioed trace
- How to use the Gryniewicz equation to create a concentration graph
- Analyze
- Maths Calculator
- Maths Operations
- Maths How To
- User Data Form

Recalculate—Ratio

Use the pages accessible from the Recalculate option to perform re-calculations on data that has been analyzed. These re-calculations can be made using new Setup parameters. For example you may wish to change the order of a reaction. You can also use the Recalculate button to access Report options.

To access the Recalculate option, select the Recalculate button from the Ratio Application window.

When you select this option, the Setup dialog will appear, containing the following tabbed pages:

- Analyze
- Reports

See also:
Ratio Application

How to generate a report from previously stored data—Ratio

Step 1

Select Open… from the File menu to display the Windows Open dialog.

Step 2

A list of previously stored files will appear. If this is not a list of batch files, select the down arrow at the right of the Files of type list box and select 'Batch' to list all the batch files.

Step 3

Select the Batch file you require then select Open.

The system will automatically retrieve the collected data into the Graphics area. The method that was used to collect the data will also be retrieved.
Note
You can also retrieve data from Methods (*.FMRT), Data (*.FDRT), Report (*.FRRT), Spreadsheet ASCII (*.CSV), GRAMS (*.SPC) and Rich Text Format (*.RTF) file types.

Step 4
Select the **Recalculate** button to display the **Setup** dialog.

Step 5
Select the **Reports** tab and on the **Reports** page specify the information you would like to include in the report. Select **Auto Print** if you require the new report to print to the Reports area and to a printer.

Step 6
Click **OK** to display the results in the Report area (and send to the printer if **Auto Print** is selected).

Note
If you require a printout of only the current graphics in the Graphics area of a previously stored data file:

a. Select the **Clear Report** option in the Commands menu to clear any text that may be displayed in the Report area.

b. Press the **Print** button to obtain a printout of the graphics.

Alternatively tick only the **Graph** check box in the **Reports** page to obtain a printout of graphics only.

See also:

**How to use advanced reporting options**

**The Grynkiewitz Equation—Ratio**

The Grynkiewitz equation is used to create a concentration graph from your ratioed trace.

\[
[\text{Ion}]_t = \frac{\text{Kd} \times (\text{Rt} - \text{Rmin}) \times \text{Sf}_2}{(\text{Rmax} - \text{Rt}) \times \text{Sb}_2}
\]

The equation consists of a number of variables:

- \([\text{Ion}]_t\): The concentration of the target ion at time \(t\).
- \(\text{Kd}\): The dissociation constant (Kd) measures the ability of a chelation agent or dye to associate itself to a target ion.
- \(\text{Rt}\): The fluorescence intensity ratio for the dye at time \(t\).
- \(\text{Rmin}\): The fluorescence intensity ratio for the probe dye (unbound).
- \(\text{Rmax}\): The fluorescence intensity ratio for the ion-bound probe in the presence of saturating ion concentrations.
- \(\text{Sf}_2/\text{Sb}_2\): The ratio of the fluorescent intensities of the free and bound ion probe. This factor eliminates any instrument dependent artifacts.

Use the **Analyze tab** to select from various mathematical equations and set the parameters used to generate the calculation of reaction rates.
See also:
- What is autofluorescence
- How to get a ratioed trace
- Using the Grynkiewitz equation and entering the values manually
- Using the Grynkiewitz equation and determining the values from the graph

**Maths window**

Use the **Maths** window to perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator.

To access the **Maths** window, select the **Calculator** command from the **Graph** menu, or select the **Calculator** button on the **Graphics** toolbar.

The **Maths** window is only available in the Kinetics, Lifetimes, Scan and Validate applications.

The **Maths** window comprises the following:

- **Calculation display field**: View the current calculation, including the trace/s being used, the trace operation being carried out and any functions that have been applied to the trace.

  - **Hot Tip**: Click on the down arrow adjacent to this field to view the last ten unique calculations that have taken place. Use this list to keep track of previous manipulations and repeat calculations quickly by simply selecting and then applying the desired calculation.

- **Selected Trace/Focused Trace**: Choosing either of these options selects the highlighted (red) trace in the current Graph box for manipulation. When choosing between these options, use this simple rule:
  - Use the **Selected Trace** option when the selected calculation is only to be applied to one trace.
  - Use the **Focused Trace** option when the same calculation is to be applied to a number of traces.

  The following example illustrates the difference between these options.

  To perform the same manipulation on several traces:
  
  a. Highlight a trace by clicking on it with the mouse. The trace will become red.
  b. Select **Selected Trace, Apply** and then = to plot the result of the calculation
  c. Highlight the next trace and repeat step b.

  OR

  a. Highlight the first trace.
  b. Select **Focused Trace, Apply** and then = to plot the result of the calculation;
  c. Highlight the next trace and select the = button to perform the same calculation on the newly selected trace.

- **Trace**: Use the Trace group options to select the operation to be performed on the traces, the filter size and the interval (where applicable).

  Click **Apply** to insert the selected operation into the equation
  Click **=** (equals) to display the result of the calculations.
Operation
Click on an operation to find out more about it

- Anisotropy
- Polarization
- Sum Function
- Mean
- Boxcar Averaging
- Moving Average
- Savitzky-Golay
- Spline
- Deriv 1
- Deriv 2
- Deriv 3
- Deriv 4

Display Options
Select whether the results of the operation performed on the trace will be viewed in a New Graph box or in the Selected Graph box. If you choose the Selected Graph option, you will be able to directly compare the results with existing traces.

Functions
This group contains number and function buttons that can be used as a standard calculator in addition to being applied to traces.

To use the functions:

1. select a trace or value
2. select either a second trace or number
3. select the equals (=) button to display the result

All the basic mathematical functions are available (addition, subtraction, multiplication and division), as well as logarithm (Log) and square root (Sqrt) functions. When used as a calculator, the results of equations appear in the Report area.

Additional buttons
C
Click C to clear the entire operation in the calculation display field.

CE
Click CE to clear the last entry entered in the calculation display field.

= 
Click = (equals) to start the operation shown in the calculation display field.

See also:
Maths Home Page
Maths How To

Maths Operations

The following operations can be selected on the Maths window. To access the Maths window, select the Calculator command from the Graph menu, or select the Calculator button on the Graphics toolbar.

Anisotropy, Polarization and Sum Function
The term Anisotropy refers to an unequal angular distribution about a point in space. Anisotropy also refers specifically to a simple expression for measuring the degree of polarization of the fluorescence. It is almost identical to the expression for polarization apart from having a factor of two in the denominator. Anisotropy is usually represented by the lowercase letter "r", while polarization by an uppercase "P".
The two expressions are listed below:

**Anisotropy**
\[ r = \frac{I_{vv} - G_I_{vh}}{I_{vv} + 2G_I_{vh}} \]

**Polarisation**
\[ P = \frac{I_{vv} - G_I_{vh}}{I_{vv} + G_I_{vh}} \]

where

- \( I_{vv} \) equals the intensity of fluorescence with excitation polarizer = vertical, emission polarizer = vertical
- \( I_{vh} \) equals the intensity of fluorescence with excitation polarizer = vertical, emission polarizer = horizontal
- G is a factor that accounts for the polarization bias of the instrument. In particular it accounts for the wavelength-dependent polarization bias of the emission side of the instrument (dominated by the polarization bias of the emission grating).

The G factor is given by:
\[ G = \frac{I_{hv}}{I_{hh}} \]

where

- \( I_{hv} \) equals the intensity of fluorescence with excitation polarizer = horizontal, emission polarizer = vertical
- \( I_{hh} \) equals the intensity of fluorescence with excitation polarizer = horizontal, emission polarizer = horizontal

The expression for the anisotropy is a quotient. The denominator of the expression, as given by \( I_{vv} + 2G_I_{vh} \) is the ‘sum’ function, and actually represents the total emission unbiased by any sample polarization effects.

**Mean**
The Mean operation allows the average of two or more traces to be taken and the resulting trace to be plotted. The mean of the traces can also be used in conjunction with other functions.

The equation used in mean calculations is:
\[ \frac{\text{scan 1} + \text{scan 2} + \ldots + \text{scan } x}{x} \]

The calculation is performed over the common X range of the traces and the trace with the most dense data is used to determine the data points. The other traces are interpolated at the relevant data points.

**Boxcar Averaging**
This algorithm will take the number of points as defined by the Filter size, and average these, displaying the average at the middle X-point of the filter.

For example, for the following:

Data: \((x_1, y_1), (x_2, y_2), (x_3, y_3) \ldots \) (xn,yn)
Filter size: 5
The first smoothed data point will be \((x_3, \text{sum}(y_1:y_5)/5)\), the next \((x_8, \text{sum}(y_6:y_{10})/5)\) ..... \((x_{n-2}, \text{sum}(y_{n-4}:y_n)/5)\).

If there are not enough points at the end of the data set to complete the filter size, then these points will be ignored, hence, the gap at the end may be larger than the gap in the beginning. There will be a gap before the first point and after the last point of the smoothed continuum, with respect to the original unsmoothed data. This algorithm is only valid for evenly x-spaced data points.

Within a time scan, (e.g., Advanced collect in kinetics) the data points may be unevenly spaced between different stages. Each stage will be treated as a new collect, hence, there may be large gaps between stages. If there are not enough points at the end of a stage to complete the filter size, then these points are ignored, hence, there may be large gaps at the end of a stage. The filter size will be limited to odd numbers, with the minimum of 3 to a maximum of 101.

**Moving Average**

This algorithm will take the number of points as defined by the Filter size, and average these, displaying the average at the middle X-point of the filter.

For example, for the following:

Data: \((x_1, y_1), (x_2, y_2), (x_3, y_3) ..... (x_n, y_n)\)

Filter size: 5

The first smoothed data point will be \((x_3, \text{sum}(y_1:y_5)/5)\), the next \((x_4, \text{sum}(y_2:y_6)/5)\) ..... \((x_{n-2}, \text{sum}(y_{n-4}:y_n)/5)\). There will be a gap before the first point and after the last point of the smoothed continuum, with respect to the original unsmoothed data. This algorithm is only valid for evenly x-spaced data points. Within a time scan, (e.g.g, Advanced collect in kinetics), the data points may be unevenly spaced between different stages. This will be treated by interpolation of data from the previous stage so as to give evenly spaced data points into the next stage. If there is not enough data for interpolation, then the next stage will be treated as a new collect, hence, there may be gaps and further delays in displaying the point. The filter size will be limited to odd numbers, with the minimum of 3 to a maximum of 101.

**Savitzky-Golay**

The Savitzky-Golay technique operation enables you to reduce the noise in the currently selected Maths scan by using smoothing.

When you select this operation you will be prompted to enter a Filter Size from 5 to 101. The larger the Filter Size, the more data points that will be smoothed.

**Q.** I cannot enter a particular value; it keeps changing to another number — why?

**A.** The Savitzky-Golay technique requires an odd number to be entered, not an even number.

You will also need to enter an Interval. The Interval refers to how many of the averaged points will be plotted on the smoothed trace. The smaller the interval, the more detail there will be in the trace.

Consider selecting an Interval of 1. This would mean that when the smoothed continuum is plotted, every smoothed point will be displayed.

**Q.** Why can't I see the Filter Size and Interval fields?
A. These will be displayed when you select Smooth in the Operation field.

**Spline**

**Deriv 1–4**

The Deriv operations allow you to obtain derivatives of a selected scan. The derivative can be of the orders 1 to 4. The resulting trace is then plotted. The Savitzky-Golay technique is used for derivative calculations.

On selection of the Deriv operation you will be prompted to enter a Filter Size from 5 to 101. The Filter Size refers to the number of points around a particular point that will be averaged to produce a new derived Y value. The lower the Filter Size, the more data points that will be used in the derivative trace.

**Q.** I cannot enter a particular value; it keeps changing to another number — why?
**A.** The Savitzky-Golay technique requires an odd number to be entered, not an even number.

You will also need to enter an Interval. The Interval refers to how many of the derived points will be plotted on the resulting trace. The smaller the Interval, the more detail there will be in the trace.

**Q.** Why can't I see the Filter Size and Interval fields?
**A.** These will be displayed when you select one of the ‘Derivs’ in the Operation field.

See also:
- About smoothing

**Maths How To…**

- How to perform a spectral subtraction
- How to perform a mean calculation
- How to smooth a trace
- How to derive a trace

See also:
- Maths Home Page

**User Data Form—Ratio**

Use the User Data Form to enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data.

To access the User Data Form, select the User Data Form command from the Graph menu, or select the User Data Form button on the Graphics toolbar. The User Data Form command is only available if you have selected a graph box.

The dialog comprises the following:

**Data Name**

This column lists the names of each visible trace in the highlighted graph box.

Use the extra columns for recording additional information about the experimental conditions you used to collect the data, eg. pH.

**Other Columns**

Use this to record the name of the parameter in which you are interested as a column heading, (for example, pH, ionic concentration, substrate concentration etc.).
Note
You must enter a column heading, or else the data in that column will not be saved.

See also:
Graph menu
Graphics toolbar

Displaying data graphically—Ratio

- Graphics area
- Graphics How To
- **Graph menu**
- Graphics toolbar
- **Graph Labels**
  - Add Label dialog
  - Edit Label dialog
  - How to add a label to a graph
  - How to edit a label
  - How to move a label
  - How to resize a label
  - How to delete a label
- **Graph Pictures**
  - Add Picture dialog
  - Edit Picture dialog
  - How to add a picture
  - How to replace a picture
  - How to move a picture
  - How to resize a picture
  - How to delete a picture
- **Trace Preferences dialog**
  - Trace Preferences menu
  - How to select the focused trace
  - How to display a trace
  - How to remove a trace from a graph
  - How to remove a trace from the trace list
- **Graph Preferences dialog**
  - How to alter the look of your graphs
- **Cursor Modes dialog**
  - How to track the focused trace
- **Axes Scales dialog**
  - How to scale your graph manually
  - How to scale your graph automatically
- Single/Multi Graph
- User Data Form
- **Graphics Templates**
  - How to create a graphics template
- **Ratio Ruler**
  - How to use the ratio ruler

**Graphics area—Ratio**

The Graphics area of the Ratio application appears in the top half of the Ratio Application window and is used to display traces of collected data. In the Cary Eclipse system, the term 'graph' refers to the plotting area and is
represented by a set of axes. The rectangular area surrounding the graph is referred to as the graph box. A graph will appear in the Graphics area when you select Start to commence a Ratio run.

To display or hide the Graphics area, select the Graphics command on the View menu. Double-clicking on the Graphics area enables you to toggle between half or full size display sizes.

You can access the functions associated with the Graphics area by doing the following:

- selecting one of the Graphics toolbar buttons;
- selecting a command from the Graph menu; or
- clicking the right mouse button in the Graphics area and select a command from the menu

**Note**
Not all functions are available on every menu.

You can customize the graphics area to suit your requirements in the following ways:

**Note**
All of the commands and dialogs listed are accessible from the *Graph menu*.

<table>
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<tr>
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<th>Use the…</th>
<th>Tell me how…</th>
</tr>
</thead>
<tbody>
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<td>Autoscale graph command</td>
<td><a href="#">How to scale your graph automatically</a></td>
</tr>
<tr>
<td>automatically</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Change the axes scaling</td>
<td>Axes Scales dialog</td>
<td><a href="#">How to scale your graph manually</a></td>
</tr>
<tr>
<td>manually</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Modify the graph appearance</td>
<td>Graph Preferences dialog</td>
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<tr>
<td>(axes, trace)</td>
<td></td>
<td><a href="#">How to specify the number of tick marks and decimal places for your X and Y axes</a></td>
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<tr>
<td>Add a new graph to the display</td>
<td>Add graph command</td>
<td></td>
</tr>
<tr>
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<td>Remove graph command</td>
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<td>View single or multiple graphs</td>
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<tr>
<td>Resize a graph box</td>
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<td>Track the focused trace</td>
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<td>Select the focused trace</td>
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<tr>
<td>Display a trace in the graph</td>
<td>Trace Preferences dialog</td>
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</tr>
<tr>
<td>Remove a trace from a graph</td>
<td>Trace Preferences dialog</td>
<td><a href="#">How to remove a trace from a graph</a></td>
</tr>
<tr>
<td>Remove a trace from the trace list</td>
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</tr>
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<td>Zoom in and out on the graph</td>
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<td>Add a label to the graph box</td>
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<td>Edit labels in the graph box</td>
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<tr>
<td>Moving labels in the graph box</td>
<td>Cursor</td>
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</tr>
<tr>
<td>Resizing labels in the graph box</td>
<td>Cursor</td>
<td>How to resize a label</td>
</tr>
<tr>
<td>Delete labels from the graph box</td>
<td>Delete Annotation command</td>
<td>How to delete a label</td>
</tr>
<tr>
<td>Add a picture to the graph box</td>
<td>Add Picture dialog</td>
<td>How to add a picture to a graph</td>
</tr>
<tr>
<td>Move pictures in the graph box</td>
<td>Cursor</td>
<td>How to move a picture</td>
</tr>
<tr>
<td>Resizing pictures in the graph box</td>
<td>Cursor</td>
<td>How to resize a picture</td>
</tr>
<tr>
<td>Replace pictures in the graph box</td>
<td>Edit Picture dialog</td>
<td>How to replace a picture</td>
</tr>
<tr>
<td>Delete pictures from the graph box</td>
<td>Delete Annotation command</td>
<td>How to delete a picture</td>
</tr>
<tr>
<td>Save the Graphics area you have set up as a graphics template</td>
<td>Save As dialog (from the File menu)</td>
<td>How to create a graphics template</td>
</tr>
</tbody>
</table>

**Note**

The red trace in any graph box is the one on which all information appears and the one used for the cursor tracking. It is called the focused trace.

**See also:**

- Toolbar
- Graphics How To

---

**Graphics How to—Ratio**

**Traces**

- How to track the focused trace
- How to select the focused trace
- How to display a trace that is not appearing in a graph box
- How to remove a trace from a graph
- How to remove a trace from the trace List

**Sizing and scale**

- How to resize graph boxes
- How to scale your graph manually
- How to scale your graph automatically
- How to alter the look of your graphs
- How to zoom a graph

**Labels**

- How to add a label to a graph
- How to edit a label
Graph menu—Ratio

Introduction

Use the Graph menu to view data in various graphical formats or in multiple graphs in the Graphics area. You can double-click on the Graphics area to toggle the display through graph and report to graph only.

In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area surrounding the graph is referred to as the graph box. A graph will appear in the Graphics area when you select Start to commence a Ratio run.

The Graph menu can be accessed in the following ways:

- from the main menu at the top of the Ratio Application window
- by clicking the right mouse button in a graph box or the Graphics area

**Note**
The red trace in any graph box is the one on which all information appears and the one used for the cursor tracking. It is called the focused trace.

**Menu commands**

The following commands can be accessed from the Graph menu:

<table>
<thead>
<tr>
<th>Menu command</th>
<th>Button</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Print XY point</td>
<td></td>
<td>Select this to print the cursor's current X and Y coordinates in the report area.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>This option is only available from the Graph menu that appears when you right-click in a graph box.</td>
</tr>
<tr>
<td>Copy Graph</td>
<td></td>
<td>Select <strong>Copy Graph</strong> to copy the entire Graphics area to the Windows Clipboard. All of the graphs in the Graphics area will be copied to the Clipboard.</td>
</tr>
<tr>
<td>Paste to Graph</td>
<td></td>
<td>Select <strong>Paste to Graph</strong> to paste the contents of the Windows Clipboard into the Graphics area. Text is pasted as a label, graphics are pasted as pictures.</td>
</tr>
<tr>
<td>Add Graph</td>
<td>![Add Graph Icon]</td>
<td>Select <strong>Add Graph</strong> to add a new graph box to the Graphics area. You may want to do this if you have, for example, a graph containing existing data and you want to collect some new data in a new graph.</td>
</tr>
<tr>
<td>Remove Graph</td>
<td>![Remove Graph Icon]</td>
<td>Select <strong>Remove Graph</strong> to delete the selected graph box from the Graphics area. The graph box is removed, but the data is not deleted and is still available from the <strong>Trace</strong> menu.</td>
</tr>
</tbody>
</table>
## Preferences window

You may want to use remove graph if you have a large number of graph boxes open but only want to focus on some of these.

**Note:**

You can remove multiple graph boxes by first holding down the CTRL key and selecting the graphs to be removed and then selecting the **Remove Graph** button or menu item. Use the SHIFT key to highlight a range of graphs for removal.

### Clear All Traces

Select **Clear All Traces** to delete all traces from the graph/s. Note that selecting this option simply turns off the **Visible** property for the traces in the Trace Preferences window. You can redisplay the traces by making the traces **Visible** again in the Trace Preferences window.

### Add Label

Select **Add Label** to display the **Add Label** dialog where you can enter a text label to be placed on a graph or in the background of the Graphics area.

### Add Picture

Select Add Picture to display the Add Picture dialog where you can select a picture, bitmap or chemical structure to be placed on a graph or in the background of the Graphics area.

### Edit Annotation

Select **Edit Annotation** to alter the content or look of the selected annotation, (either a label or a picture) in the Graphics area.

**Note:**

This option can only be activated if an annotation is selected.

If the selected annotation is a label, selecting **Edit Annotation** will open the **Edit Label** dialog where you can change the text of the selected label.

If the selected annotation is a picture, selecting **Edit Annotation** will open the **Edit Picture** dialog where you can replace the selected picture with a new picture.

### Delete Annotation

Select **Delete Annotation** to remove an annotation, (such as a label or a picture) from the Graphics area.

You can use this menu option to remove highlighted annotations in the Graphics area such as labels, pictures and ruler/fit information.

**Note:**

This option can only be activated if an annotation is selected.

### Trace Preferences

Select this command to display the Trace Preferences dialog where you can select the trace(s) to display in the selected graph.

### Graph Preferences

Select this command to display the Graph Preferences dialog where you can change the appearance of your graph. Any changes you make to the appearance of the graph will be applied to all graphs.

### Cursor Mode…

Select this command to display the Cursor Modes dialog where you can choose the type of cursor to be used on the screen. For example, you can select to use a free cursor that can move anywhere in the graph or exactly trace the focused scan (red scan). Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the Least Squares method or the Point to Point method.

### Axes Scales…

Select this command to display the Axes Scales dialog where you can set the scale range for the X and Y axes.

### Auto scale graph

To automatically scale the graph, point to one of the following options:

* Autoscale[XY]*
Use the Graphics toolbar buttons to gain quick access to many of the Graph menu options. You can also use:

<table>
<thead>
<tr>
<th>Button</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Autoscale[XY]</td>
<td>Select Autoscale[XY] to automatically scale collected data by scaling the Y height and X width to full screen.</td>
</tr>
<tr>
<td>Autoscale[X]</td>
<td>Select Autoscale[X] to automatically scale collected data by scaling the X width to full screen.</td>
</tr>
<tr>
<td>Autoscale[Y]</td>
<td>Select Autoscale[Y] to automatically scale collected data by scaling the Y height to full screen.</td>
</tr>
<tr>
<td>Zoom Out</td>
<td>Select Zoom Out to zoom back to the original X and Y ranges which were used to collect the data (i.e. the method setup conditions of the focused scan). For example, the system will revert back to the X and Y ranges set in the method if you zoom in and out during a data collection.</td>
</tr>
<tr>
<td>Single/Multi Graph</td>
<td>Select Single/Multi Graph to toggle between displaying one or all graphs. Note: The graph highlighted in the Multi Graph view will be displayed when you toggle the Single Graph option.</td>
</tr>
<tr>
<td>Auto Arrange Graphs</td>
<td>Select Auto Arrange Graphs to view and arrange all graphs simultaneously. Note: You can move graph boxes manually by clicking the left mouse button outside the graph axes and dragging the graph box to a new position. You can also resize graphs.</td>
</tr>
<tr>
<td>User Data Form</td>
<td>Select User Data Form to display the User Data Form where you can enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data, such as the pH at which data was collected.</td>
</tr>
<tr>
<td>Calculator</td>
<td>Select Calculator to display the Maths window where you can perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator.</td>
</tr>
<tr>
<td>History List</td>
<td>This displays a list of the current graphs.</td>
</tr>
</tbody>
</table>

See also:
- Add Label
- Add Picture
- Edit Label
- Edit Picture
- Trace Preferences
- Graph Preferences
- Cursor Modes
- Axes Scales
- Single/Multi Graph
- User Data Form
- Maths
- Graphics toolbar
- Graphics area

**Graphics Toolbar—Ratio**

Select Autoscale[XY] to automatically scale collected data by scaling the Y height and X width to full screen.

**Autoscale[X]**
Select Autoscale[X] to automatically scale collected data by scaling the X width to full screen.

**Autoscale[Y]**
Select Autoscale[Y] to automatically scale collected data by scaling the Y height to full screen.

Select Zoom Out to zoom back to the original X and Y ranges which were used to collect the data (i.e. the method setup conditions of the focused scan). For example, the system will revert back to the X and Y ranges set in the method if you zoom in and out during a data collection.

Select Single/Multi Graph to toggle between displaying one or all graphs.

Note: The graph highlighted in the Multi Graph view will be displayed when you toggle the Single Graph option.

Select Auto Arrange Graphs to view and arrange all graphs simultaneously.

Note: You can move graph boxes manually by clicking the left mouse button outside the graph axes and dragging the graph box to a new position. You can also resize graphs.

Select User Data Form to display the User Data Form where you can enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data, such as the pH at which data was collected.

Select Calculator to display the Maths window where you can perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator.

This displays a list of the current graphs.
the toolbar to select the graph you want to view and to change the look of your graphs.

To display or hide the toolbar, point to the **Toolbars** option in the **View** menu and select **Graphics**. To display or hide the toolbar's text labels, point to the **Toolbars** option in the **View** menu and select **Text Labels**.

**Hot Tip**
To see the name of a button, point to it with the mouse.

The following commands can be accessed from the Graphics toolbar:

<table>
<thead>
<tr>
<th>Button</th>
<th>Menu command</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image1" alt="Image" /></td>
<td>Trace Preferences</td>
<td>Click this button to display the <strong>Trace Preferences</strong> window where you can select the trace(s) to display in the selected graph.</td>
</tr>
<tr>
<td><img src="image2" alt="Image" /></td>
<td>Graph Preferences</td>
<td>Click this button to display the <strong>Graph Preferences</strong> dialog where you can change the appearance of your graph. Any changes you make to the appearance of the graph will be applied to all graphs.</td>
</tr>
<tr>
<td><img src="image3" alt="Image" /></td>
<td>Cursor Mode</td>
<td>Click this button to display the <strong>Cursor Modes</strong> dialog where you can choose the type of cursor to be used on the screen. For example, you can select to use a free cursor that can move anywhere in the graph or exactly trace the focused scan (red scan). Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the Least Squares method or the Point to Point method.</td>
</tr>
<tr>
<td><img src="image4" alt="Image" /></td>
<td>Axes Scales…</td>
<td>Click this button to display the <strong>Axes Scales</strong> dialog where you can set the scale range for the X and Y axes.</td>
</tr>
<tr>
<td><img src="image5" alt="Image" /></td>
<td>Autoscale [XY]</td>
<td>Click the <strong>Autoscale[XY]</strong> button to automatically scale collected data by scaling the Y height and X width to full screen.</td>
</tr>
<tr>
<td><img src="image6" alt="Image" /></td>
<td>Autoscale [X]</td>
<td>Click the <strong>Autoscale[X]</strong> button to automatically scale collected data by scaling the X width to full screen.</td>
</tr>
<tr>
<td><img src="image7" alt="Image" /></td>
<td>Autoscale [Y]</td>
<td>Click the <strong>Autoscale[Y]</strong> button to automatically scale collected data by scaling the Y height to full screen.</td>
</tr>
<tr>
<td><img src="image8" alt="Image" /></td>
<td>Zoom Out</td>
<td>Click the <strong>Zoom Out</strong> button to Zoom back to the original X and Y ranges which were used to collect the data (i.e. the method setup conditions of the focused scan). For example, the system will revert back to the X and Y ranges set in the method if you zoom in and out during a data collection.</td>
</tr>
<tr>
<td><img src="image9" alt="Image" /></td>
<td>Add Label</td>
<td>Click this button to display the <strong>Add Label</strong> dialog where you can enter a text label to be placed on a graph or in the background of the Graphics area.</td>
</tr>
<tr>
<td><img src="image10" alt="Image" /></td>
<td>Add Picture</td>
<td>Click this button to display the <strong>Add Picture</strong> dialog where you can select a picture, bitmap or chemical structure to be placed on a graph or in the background of the Graphics area.</td>
</tr>
</tbody>
</table>
| ![Image](image11) | Single/Multi Graph | Select **Single/Multi Graph** to toggle between displaying one or all graphs.  
**Note:** The graph highlighted in the Multi Graph view will be displayed when you toggle the Single Graph option. |
| ![Image](image12) | Auto Arrange | Click the **Auto Arrange Graphs** button to view and arrange all graphs simultaneously. |
### Graph Labels—Ratio

<table>
<thead>
<tr>
<th>Graphs</th>
<th>Note: You can move graph boxes manually by clicking the left mouse button outside the graph axes and dragging the graph box to a new position. You can also resize graphs.</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Add Graph</strong></td>
<td>Click this button to add a new graph box to the Graphics area. You may want to do this if you have, for example, a graph containing existing data and you want to collect some new data in a new graph.</td>
</tr>
<tr>
<td><strong>Remove Graph</strong></td>
<td>Click this button to delete the selected graph box from the Graphics area. The graph box is removed, but the data is not deleted and is still available from the Trace Preferences window. You may want to use remove graph if you have a large number of graph boxes open but only want to focus on some of these.</td>
</tr>
<tr>
<td><strong>Clear All Graphs</strong></td>
<td>Click this button to delete all traces from the graph/s. Note that selecting this option simply turns off the Visible property for the traces in the Trace Preferences window. You can redisplay the traces by making the traces Visible again in the Trace Preferences window.</td>
</tr>
<tr>
<td><strong>User Data Form</strong></td>
<td>Click this button to display the User Data Form where you can enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data, such as the pH at which data was collected.</td>
</tr>
<tr>
<td><strong>Calculator</strong></td>
<td>Click this button to display the Maths window where you can perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator.</td>
</tr>
</tbody>
</table>

See also:
- Add Label
- Add Picture
- Edit Label
- Edit Picture
- Trace Preferences
- Graph Preferences
- Cursor Modes
- Axes Scales
- Single/Multi Graph
- User Data Form
- Maths
- Graphics area
- Graph menu
- Graphics How to

**Graph Labels—Ratio**

- Add Label dialog
- Edit Label dialog
- How to add a label to a graph
Add Label—Ratio

Use the Add Label dialog to place a label on your graph or into the background of the Graphics area.

To add a label to a graph box, you must have at least one trace Visible in the graph box. (You can select which traces you want to make visible in the Trace Preferences dialog.) If you are viewing multiple graphs, the label will be placed in the graph box that is currently selected. However, you can add a label outside of a graph box in the background of the Graphics area at any time.

Q. Can I add a label that will print with all my graphs.
A. Yes, you can add your label to the background of the Graphics area. To do this, right-click on a section of the background (gray area) and select Add Label from the menu. (You will notice that this menu has less options than the menu that appears when you right-click on a graph.) In the Add Label dialog, enter the label to be added to all graphs and select OK. The label will appear in the background of the Graphics area.

To access the Add Label dialog, select the Add Label command from the Graph menu, or select the Add Label button on the Graphics toolbar.

The Add Label dialog comprises the following:

- **To selected trace**
  - Select this option to associate a label with the selected trace. When you associate a label with a trace and make the trace visible in the Graph box, the label will also be displayed with the trace.

- **To selected graph**
  - Select this option to associate the label with the selected graph. When you associate a label with a graph, the label will always be displayed in the Graph box, regardless of what traces are visible.

- **Selected trace**
  - This will identify the trace that is currently selected and display the name of the trace.

- **Font…**
  - Click Font to open the Windows Font dialog. In this dialog you can specify the font type, style and size to be used for the label.

- **Label**
  - Use the Label field to enter the text for the label. You can enter as many lines of text as you require.

**Hot Tip**
If you wish to include certain XY co-ordinates on the graph as a label, position your cursor at the appropriate place, right-click and select Add Label from the menu. When the Add Label dialog appears, the co-ordinate positions will be added to the Label field.

**Note**
When you save the data (as a Batch or Data file) the label is saved with its associated trace or graph. Labels outside graph boxes can be saved in a graphics template. To do this, choose Graph Template as the Save As type in the Save As dialog. See Example of a Graphics Template.

See also:
- Trace Preferences dialog
- Graph menu
Use the Edit Label dialog to alter the content or look of your label.

To access the Edit Label dialog, right-click on the label you wish to change and select Edit Annotation from the menu.

The dialog comprises the following:

Font
Select Font to open the Font dialog, where you can specify the font type, style and size to be used for the label.

Label
Use the Label field to alter the text for the label.

See also:
How to edit a label
How to add a label to a graph
How to resize a label
How to move a label

Edit Label—Ratio

How to add a label to a graph—Ratio

You can add a label to your graph or Graphics area background. To do this:

Step 1

Click on the graph box you require or click outside the graph box(es) to choose the Graphics area background.

Note
There must be at least one trace Visible in the graph box before you can add a label to it.

Step 2

Click the Add Label button on the toolbar or right-click on the graph and select Add Label from the menu.
The Add Label dialog will appear.
The Add Label command is also accessible from the Graph menu.

Step 3

Click the Font button. The Windows Font dialog appears. Select the font type, size and style you require for your label. A sample of the font style appears in the Sample field. When you are satisfied with your selections, select OK to return to the Add Label dialog.

Step 4

Enter the label in the Label field and select OK. The label will be displayed on the selected graph. If
necessary, you can move a label so that it is more appropriately placed on your graph.

See also:
- Graphics Area
- How to move a label
- How to edit a label
- How to resize a label

**How to edit a label—Ratio**

**Step 1**
Place the cursor on the label and click the left mouse button to select the label.

**Hot Tip**
If you are having trouble selecting the label it may be that a trace is interfering with the label selection. Try zooming in and clicking in a different part of the label.

**Step 2**
Click the right mouse button and select **Edit Annotation** from the drop-down menu. Alternatively you can select the **Edit Annotation** command from the **Graph** menu. The **Edit Label** dialog is displayed.

**Step 3**
Use the **Edit Label** dialog to change the label.

For example, if you want to change the font size of the label, select the **Font** button. In the **Size** field, scroll through the available sizes for the font and select the size you require. Alternatively, highlight the currently selected size in the **Size** entry field and enter the size you require. Click the **OK** button in this dialog to return to the **Edit Label** dialog.

**Step 4**
Click **OK** to clear the **Edit Label** dialog. Your label will appear with the new font size.

See also:
- How to add a label to a graph
- How to move a label
- How to resize a label
- How to delete a label

**How to move a label—Ratio**

To move a label so that it is more appropriately placed on your graph or Graphics area background, do the following:

**Step 1**
Place the cursor on the label.

**Step 2**
Click and hold down the left mouse button.

**Hot Tip**
If you are having trouble selecting the label, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 3

Drag the label to the correct position and release the mouse button. The label will be displayed at the new position.

**Note**
Labels are transparent so as not to interfere with any traces on graphs.

See also:
- How to add a label to a graph
- How to edit a label
- How to delete a label
- How to resize a label

**How to resize a label—Ratio**

To resize a label so that it is more appropriately sized on your graph or Graphics area background, do the following:

Step 1

Select the label by clicking on it with the mouse. A border will be displayed around the label to indicate it is selected.

**Hot Tip**
If you are having trouble selecting the label, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Position the cursor over one of the corner edges of the label until the cursor becomes a diagonal double-headed arrow. Click and hold down the left mouse button.

Example of label showing the border that indicates it is selected and the arrow to be used for re-sizing

Step 3

Increase or decrease the size of the label by dragging the mouse. The label text will also be resized. Release the mouse button when the label and text are the correct size.

**Note**
Labels are transparent so as not to interfere with any traces on graphs.
How to delete a label—Ratio

To delete a label from a graph or Graphics area background, do the following:

Step 1

Position the cursor on the label and click the left mouse button to select it.

Hot Tip
If you are having trouble selecting the label it may be that a trace is interfering with the label selection. Try zooming in and clicking in a different part of the label.

Step 2

Click with the right mouse button and select Delete Annotation from the drop-down menu displayed. Alternatively, you can select the Delete Annotation command from the Graph menu. The label will be deleted from the graph or Graphics area.

Graph Pictures—Ratio

- Add Picture dialog
- Edit Picture dialog
- How to add a picture
- How to replace a picture
- How to move a picture
- How to resize a picture
- How to delete a picture

Add Picture—Ratio

Use the Add Picture dialog to insert a picture or bitmap into the Graphics area. The picture can be added as a background picture or to a particular graph box.

To add a picture to a graph box you must have at least one trace Visible in the graph box. (You can select which traces you want to make visible in the Trace Preferences dialog.) If you are viewing multiple graphs, the picture will be placed in the graph box that is currently selected. However, you can add a picture outside of a graph box in the Graphics area at any time.

Q. Can I add a picture that will print with all my graphs.

A. Yes, you can add your picture to the background of the Graphics area. To do this, right-click on a
section of the background and select **Add Picture** from the menu. (You will notice that this menu has less options than the menu that appears when you right-click on a graph.) In the **Add Picture** dialog, load the picture to be added to all graphs and select **OK**. The picture will appear in the background of the Graphics area.

To access the **Add Picture** dialog, select the **Add Picture** command from the **Graph** menu, or select the **Add Picture** button on the Graphics toolbar.

The **Add Picture** dialog comprises the following commands:

<table>
<thead>
<tr>
<th>Command</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>To selected trace</strong></td>
<td>Select this option to associate a picture with the selected trace. When you associate a picture with a trace and make the trace visible in the Graph box, the label will also be displayed with the trace.</td>
</tr>
<tr>
<td><strong>To selected graph</strong></td>
<td>Select this option to associate the picture with the selected graph. When you associate a picture with a graph, the label will always be displayed in the Graph box, regardless of what traces are visible.</td>
</tr>
<tr>
<td><strong>Load</strong></td>
<td>Click <strong>Load</strong> to open the Windows <strong>Open</strong> dialog. Use this dialog to browse your computer for the picture you want to insert. The picture must be one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.</td>
</tr>
</tbody>
</table>

If you are viewing multiple graphs the picture will be placed on the graph that is currently selected. If no graph is selected, the picture will be placed on the Graphics area background.

**Note**

When you save the data (as a Batch or Data file) the label is saved with its associated trace or graph. Labels outside graph boxes can be saved in a graphics template. To do this, choose **Graph Template** as the **Save As** type in the **Save As** dialog.

See 'Example of a Graphics Template'.

See also:
- Trace Preferences
- Graph menu
- Graphics toolbar
- How to add a picture to a graph
- How to move a picture
- How to delete a picture
- How to replace a picture

**Edit Picture—Ratio**

Use the **Edit Picture** dialog to change a picture you have inserted into the Graphics area.

To access the **Edit Picture** dialog, right-click on the picture you wish to change and select **Edit Annotation** from the menu.

The **Edit Picture** dialog comprises the following commands:

<table>
<thead>
<tr>
<th>Command</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>To selected trace/graph</strong></td>
<td>Displays whether you originally selected to add the picture to a trace or a graph. This option is not editable.</td>
</tr>
<tr>
<td><strong>Load</strong></td>
<td>Click <strong>Load</strong> to open the Windows <strong>Open</strong> dialog. Use this dialog to browse your computer for the picture you want to insert. The selected picture will replace the current picture. The picture must be one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.</td>
</tr>
</tbody>
</table>
How to add a picture to a graph—Ratio

You can add a picture to your graph or the Graphics area background. To do this:

Step 1

Click on the graph box you require or click outside the graph box(es) to choose the Graphics area background.

**Note**

There must be at least one trace Visible in the graph box before you can add a picture to it.

Step 2

Click the **Add Picture** button on the toolbar or right-click on the graph and select **Add Picture** from the menu. The **Add Picture** dialog will appear. The **Add Picture** command is also accessible from the **Graph** menu.

Step 3

Click the **Load** button. The Windows **Open** dialog appears.

Step 4

Browse through your directories to find the picture you want to insert. All pictures must be in the bitmap format and will have the extension .BMP.

Step 5

Highlight the .BMP that you want to insert and select **Open**.

Step 6

The **Add Picture** dialog will appear showing a preview of the bitmap. If the bitmap is the correct one select the **OK** button. The bitmap will be placed onto the graph.

**Note**

You should take care not to add a bitmap that is too big as it will obscure part or all of your graph.

You can move a picture so that it is more appropriately placed on your graph.

See also:

- How to replace a picture
- How to move a picture
- How to resize a picture
- How to delete a picture
How to replace a picture—Ratio

To change a picture that you have added to a graph or Graphics area background do the following:

Step 1

Move the cursor to the picture and select the left mouse button to select it.

**Hot Tip**
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Click the right mouse button and select **Edit Annotation** from the drop-down menu or select **Edit Annotation** from the **Graph** menu. The **Edit Picture** dialog is displayed.

Step 3

From the **Edit Picture** dialog, select the **Load** button. The Windows **Open** dialog is displayed.

Step 4

Browse through your directories to find the picture to replace the current bitmap. Picture files must be in one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.

Step 5

Highlight the file that you want to insert and select **Open** to return to the **Edit Picture** dialog.

Step 6

The **Edit Picture** dialog will display a preview of the bitmap. If the bitmap is the correct one select the **OK** button. The bitmap will be placed onto the graph. If you need to, move the picture so that it is more appropriately placed on your graph.

**Note**
Do not to add a bitmap that is too big as it will obscure part or all of your graph.

See also:
- How to add a picture to a graph
- How to move a picture
- How to delete a picture
- How to resize a picture

How to move a picture—Ratio

To move a picture so that it is more appropriately placed on your graph or Graphics area background, do the following:

Step 1
Place the cursor on the picture.

Step 2

Click and hold down the left mouse button.

**Hot Tip**
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 3

Drag the picture to the correct position and release the mouse button. The picture will be displayed at the new position.

**See also:**
- How to add a picture to a graph
- How to delete a picture
- How to replace a picture
- How to resize a picture

**How to resize a picture—Ratio**

To resize a picture so that it is more appropriately sized on your graph or Graphics area background, do the following:

Step 1

Select the picture by clicking on it with the mouse. A border will be displayed around the picture to indicate it is selected.

**Hot Tip**
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Position the cursor over one of the corner edges of the picture until the cursor becomes a diagonal double-headed arrow. Click and hold down the left mouse button.

*Example of picture showing the border that indicates it is selected and the arrow to be used for re-sizing*

Step 3

Increase or decrease the size of the picture by dragging the mouse. The picture will also be resized. Release the mouse button when the picture is the correct size.
How to delete a picture—Ratio

To delete a picture from a graph or Graphics area background, do the following:

Step 1

Position the cursor on the picture and click the left mouse button to select it.

Hot Tip
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Click with the right mouse button and select Delete Annotation from the drop-down menu displayed. Alternatively, you can select the Delete Annotation command from the Graph menu. The picture will be deleted from the graph or Graphics area.

Trace Preferences—Ratio

A single graph box can display many traces. This window enables you to select the trace(s) to display (make Visible) in the selected graph box.

To access the Trace Preferences dialog, select the Trace Preferences command from the Graph menu, or select the Trace Preferences button on the Graphics toolbar.

The dialog comprises the following:

Hot Tip
The Trace Preferences menu is also available on clicking the right mouse button anywhere in the trace listing.

Trace Preferences table

<table>
<thead>
<tr>
<th>Visible</th>
<th>Use the Visible field to select which traces will appear on the selected graph. To select or clear the Visible field, simply click the left mouse button in this column. A tick will appear next to traces that are to be displayed in the selected graph.</th>
</tr>
</thead>
</table>

See also:
How to add a picture to a graph
How to move a picture to a graph
How to delete a picture
How to replace a picture
How to resize a picture
If you are in single graph view, all selected traces will appear on the one graph. If you are in Multi Graph view the traces will appear on the graph that is currently selected.

**Hot Tip**
To make all traces Visible, right-click on the table and select the Select All command from the menu. Then right-click on the table again and select the Show Traces command from the menu.

**Color**
The Color field enables you to view the color of each trace that appears on the selected graph. To change the color of a trace, highlight it, click the right mouse button in the Trace Preferences table and select Change Trace Color. You can then select a color from the Color dialog that is displayed.

The red trace in any graph box is the one on which all information appears. This trace, which is used for the cursor tracking, is called the focused trace and it is always red. When you select a trace in the Color column it will become the focused trace in the selected graph box.

**Note**
When you change the trace line width in the Graph Preferences dialog it also will change the width of the color trace line in this dialog.

**Name of trace**
The Name of Trace field contains a description of the traces currently available for display in the selected graph box.

**Filename**
This field displays the filename and path for the trace. If the name is too long to appear in the field you can use the keyboard arrow keys to scroll to the end of the filename or you can resize the Trace Preferences window. If the file path for a trace shows 'not saved' then the trace is unsaved data and is only stored in memory.

**Trace Audit Log**
The Trace Audit Log displays information about the highlighted trace. This includes instrument parameters and data form information relating to the trace that has been selected. It also logs all information about data manipulation. When more than one trace is highlighted, the Trace Audit Log will display information about the trace that was first selected.

**See also:**
Graph menu
Graphics toolbar
Graph Preferences
Trace Preferences menu
How to make multiple traces visible in a graph
How to remove a trace from the trace list
How to remove a trace from a graph

**Trace Preferences menu—Ratio**
The Trace Preferences menu is displayed when you click the right mouse anywhere on the Trace Preferences table. The menu contains the following commands:

**Select All**
This selects or highlights all the traces in the listed on the Trace Preferences page.

**Select None**
This de-selects all the traces in the listing.

**Show Traces**
Select Show Traces to make all of the traces currently highlighted Visible, meaning that
they will be displayed in the current graph box.

**Hide Traces**  Select **Hide Traces** to remove the **Visible** property from all the traces currently highlighted, which will remove them from the current graph box.

**Delete Traces**  Select **Delete Traces** to delete all the highlighted traces in all graphs. Note that if you save the data file before you select **Delete Traces**, the traces are not deleted from the saved file, and they can be retrieved at a later date.

Selecting **Delete Traces** will delete the traces from the application memory, and it is recommended that you select this option if you need to free up memory on your computer. This is especially relevant for the Ratio application where the data files can be quite large.

**Note**  If the application is controlled by the GLP Administration application, you will be prompted to save an unsaved data file before it is removed from the application.

**Change Trace Color**  Select **Change Trace Color** to display the Windows **Color** dialog where you can change the color of the currently highlighted trace(s). The focused trace will remain red while it retains the focus, and will revert to the selected color when the focus is moved.

**See also:**
- [Trace Preferences page](#)
- [Graph Preferences page](#)
- [How to remove a trace from the trace list](#)
- [How to remove a trace from a graph](#)

### How to select the focused trace—Ratio

To select the scan that you want to be the focused (red) trace, do one the following:

- From the Graph, click the left mouse button on the trace that you want to make the focused trace.

- From the Graph Legend (if displayed), click the left mouse button on the trace that you want to make the focused trace.

- From the **Trace Preferences** dialog, click in the **Color** column of the trace that you want to make the focused trace.

**Note**  Select the **Show Legend** check box in the **Graph Preferences** dialog to display the **Graph Legend**.

**See also:**
- [How to track the focused trace](#)
- [How to view a particular trace](#)

### How to display a trace that is not appearing in the graph box—Ratio

**Step 1**

Click on the graph in the Graphics area.

**Step 2**

Select the **Trace Preferences** button from the Graphics toolbar, or select **Trace Preferences** from the **Graph** menu. The **Trace Preferences** dialog is displayed.
Step 3

Find that trace that you wish to see and select the **Visible** option. A tick will appear in the **Visible** column to indicate that it will now be displayed.

If you wish to make multiple traces visible, do the following:

a. Hold down the CTRL key select all the traces that you want to display.

b. Click the right mouse button and select the **Show Traces** command from the drop-down menu. Ticks will appear next to the selected traces to indicate that they will now be displayed.

Step 4

Press **OK** to clear the **Trace Preferences** dialog. The selected trace(s) will be displayed.

See also:
- [How to remove a trace from a graph](#)
- [How to remove a trace from the trace list](#)

**How to remove a trace from a graph—Ratio**

Step 1

Select a graph in the Graphics area.

Step 2

Click the **Trace Preferences** button on the Graphics toolbar or select **Trace Preferences** from the **Graph** menu. The **Trace Preferences** dialog is displayed.

Step 3

Find that trace that you no longer wish to see and select the **Visible** option. The tick will be removed from the **Visible** column to indicate that it will not be displayed.

If you wish to make multiple traces visible, do the following:

a. Hold down the CTRL key select all the traces that you no longer want to display.

b. Click the right mouse button and select the **Hide Traces** command from the drop-down menu. The ticks will be removed from the selected traces to indicate that they will not be displayed.

Step 4

Select **OK** to clear the **Trace Preferences** dialog. The trace will be removed from the graph.

See also:
- [How to display a trace that is not appearing in the graph box](#)
- [How to remove a trace from the trace list](#)

**How to remove a trace from the trace list—Ratio**
If your graphs contain unwanted traces, you may wish to delete them (rather than simply making them invisible) in order to free up application memory and aid system performance.

Step 1

Select a graph in the Graphics area.

Step 2

Click the Trace Preferences button on the Graphics toolbar or select Trace Preferences from the Graph menu. The Trace Preferences dialog is displayed.

Step 3

Select the trace to be removed by clicking on it.

Step 4

Click the right mouse button to display the Trace Preferences menu.

Step 5

Select Delete Traces to delete the sample from the trace list. You will be prompted to confirm this action.

Step 6

Select OK to clear the Trace Preferences dialog. The trace will be removed from all graphs.

See also:
How to remove a trace from a graph
How to display a trace that is not appearing in the graph box

Graph Preferences—Ratio

Axes Preferences | Advanced Options | Trace Style | Show Legend

Introduction

Use the Graph Preferences dialog to change the appearance of your graph.

All graphs will appear labeled with the X axis label being horizontal and the Y axis vertical. You cannot change the orientation of these labels. However you can alter the font and size of the axes and their labels.

Any changes you make to the appearance of the graph will apply to all graphs until the settings are changed again.

To access the Graph Preferences dialog, select the Graph Preferences command from the Graph menu, or select the Graph Preferences button on the Graphics toolbar.

Dialog controls

The Graph Preferences dialog comprises the following:
Axes Preferences

**Axes Color**  
Select the color that you want to assign to the axes of the graph/s from the drop down list.

**Axes Width**  
Select the width of the axes from the drop down list. The width, which is in pixels, ranges between one and five pixels.

**Grid Style**  
Select the style of grid for the graph from the drop down list. If you do not wish to include a grid on the graph, select **None**.

**Plot Area Color**  
Select the color to be used as the background of the graph from the drop down list. If you do not want a background color on the graph, select **White**.

**Trace Width**  
Select the width of the trace from the drop down list. The width is given in pixels and ranges between one and five pixels.

**Font**  
Click the **Font** button to display the Windows **Font** dialog where you can define the font style for the axes of the graph/s. The style and size of the font will be applied to all graph. The style, name and point size of the font appears on the font button.

**Preview Box**  
Preview any changes you make to the axes preferences in the preview box. The changes shown in this box will not be made until you select **OK**.

Advanced Options

**Auto tick marks**  
The **Auto tick marks** option enables you to approximately set how many tick marks (or points) will be included on the graph axes. The tick marks generated will be whole, rounded, even numbers. The **Auto tick marks** option is not available if you have selected **Major ticks**.

**Y-axis**  
Use the **Y-axis** slider bar to increase or decrease the number of tick marks (or points) to be included on the Y axis.

**X-axis**  
Use the **X-axis** slider bar to increase or decrease the number of tick marks (or points) to be included on the X axis.

**Major ticks**  
Use the **Major Ticks** option to enable you to set exactly how many ticks (or points) will be added to the X and Y axes. The number of marks you specify will be divided by the axis range, which can result in odd numbers on the axis. Note that any changes are not reflected in the graph bitmap at the top of the screen.

**Decimals**  
Use the **Decimals** option to set the number of X and Y decimal places to be displayed on the axes. To set the number of decimal places, select the **Decimal** option and enter the number in the Y-axis and X-axis combo boxes. Note that any changes are not reflected in the graph bitmap at the top of the screen.

Trace Style

**Points**  
Select **Points** to display each individual data point collected by the Cary Eclipse on the graph.

**Points and**  
Select **Points and solid lines** to display each individual data point collected by the Cary
**solid lines**  Eclipse on the graph. Additionally, an interpolated line that connects the points will be drawn.

**Solid lines**  If you select **Solid lines**, the Cary Eclipse system will interpolate the data between the actual, collected data points and show the data as a continuous line.

**Style cycles automatically**  If you select **Style cycles automatically**, the Cary Eclipse system will display each of the above three trace styles. This makes it easier to differentiate between traces, especially on black-and-white printers.

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**Show Legend**

**Show Legend**  Select **Show Legend** to display a legend in the right side of each graph box. A single left click on a sample name or example line within the legend will make it the focused or red scan. Information about the trace will be displayed below the X axis.

**Hot Tip**  Double-clicking on the legend will automatically display the **Trace Preferences** window, allowing you to view more detailed information about your trace (e.g. audit log, File name, etc.). You can also use the legend to select which trace will be the focused trace.

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**How to alter the look of your graphs—Ratio**

**Step 1**

Click the **Graph Preferences** button on the Graphics toolbar, or select **Graph Preferences** from the **Graph** menu. The **Graph Preferences** dialog is displayed.

**Step 2**

Use the available options available to choose your preferred appearance for the graphs. A preview box in the top right-hand corner will demonstrate how the graphs will appear.

**Step 3**

Select OK to clear the **Graph Preferences** dialog and update all graphs to reflect your selections.

**Note**

You can also add labels and pictures to your graphs to alter their appearance.

**Step 4**

To re-use your graphics setup at a later date, save the setup as a graphics template.

---

See also:

- [Graph menu](#)
- [Graphics toolbar](#)
- [Graphics area](#)
- [Graphics How To](#)
Cursor Modes—Ratio

Use the Cursor Modes dialog to choose the type of cursor to be used on screen. For example, you can select to use a free cursor that can move anywhere in the graph. Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the Least Squares method or the Point to Point method.

To access the Cursor Modes dialog, select the Cursor Modes command from the Graph menu, or select the Cursor Modes button on the Graphics toolbar.

The dialog comprises the following:

**Cursor Modes**

- **None**
  - Select this for no cursor tracking.

- **Free**
  - The Free cursor is not confined to a particular trace on the graph. It can move anywhere in a graph box, and its ‘X,Y’ position is shown in the bottom right-hand side of the Graphics area. You can use the Free cursor to zoom in on a particular part of a graph.

  **Note**
  - The Free cursor moves from pixel point to pixel point in the selected graph box. Hence you need to be careful when positioning the cursor over a scan to obtain an accurate (X,Y) reading. It is recommended that you use the Track cursor mode for this.

- **Track**
  - Use the Track cursor to exactly trace the focused (red) scan in a graph box. The cursor position is displayed in the X and Y at the bottom right-hand side of the Graphics area.

    **Hot Tip**
    - If you wish to fix the cursor on a particular position on the curve, move the cursor to the required position. Click the left mouse button, press ‘Alt’ on the keyboard, then release the mouse button.

- **Ruler-Least Squares**
  - This uses a least-squares technique to determine the slope and thus the reaction rate between the start and end points.

  This ratio ruler function can be used for measuring zero order reaction rates in ratio traces. You apply the ruler after ratio data is collected to manually select the range over which to calculate the reaction rate. The slope is then determined and thus the rate of reaction.

- **Ruler-Point to Point**
  - This uses the slope of the line drawn between two selected points to determine the point to point slope and thus the reaction rate.

  This ratio ruler function can be used for measuring zero order reaction rates in ratio traces. You apply the ruler after ratio data is collected to manually select the range over which to calculate the reaction rate. The slope is then determined and thus the rate of reaction.

See also:
- About the ratio ruler
- How to use the ratio ruler.
How to track the focused trace—Ratio

To track the focused (red) trace in the graph:

Step 1

Click the Cursor Modes button from the Graphics toolbar, or select Cursor Mode from the Graph menu. The Cursor Modes dialog will appear.

Step 2

Select Track and select the OK button.

Step 3

Place the blue tracking cursor in the graph box you require.

Step 4

Move the cursor left and right across the graph box to trace the focused (red) scan. The cursor coordinates change (in the X, Y field at the bottom left hand corner of the Graphics area) as you move along the curve.

Hot Tip
If you wish to fix the cursor on a particular position on the curve, move the cursor to the required position. Press 'Alt' on the keyboard, then release the mouse button.

See also:
How to select the focused trace
How to view a particular trace

Axes Scales—Ratio

Use the Axes Scales dialog to manually change the axes range. This can assist in zooming in on a particular part of the graph.

To access the Axes Scales dialog, select the Axes Scales command from the Graph menu, or select the Axes Scales button on the Graphics toolbar.

The dialog comprises the following:

X Axis

Mode
View the current units for the X axis.

Scale
Use the Scale field to enter a new range for the X axis, in the format n1 - n2 (where n1 is the start of the scale and n2 is the end of the scale).

Hot Tip
The Cary Eclipse will store the ten most recently entered ranges, which you can access by selecting the down arrow adjacent to the Scale field.
Y Axis

**Mode**
- View the current units for the X axis.

**Scale**
- Use the **Scale** field to enter a new range for the Y axis, in the format n1 - n2 (where n1 is the start of the scale and n2 is the end of the scale).

**Hot Tip**
The Cary Eclipse will store the ten most recently entered ranges, which you can access by selecting the down arrow adjacent to the **Scale** field.

See also:
- Graph menu
- Graphics toolbar
- How to scale your graph manually
- How to scale your graph automatically
- How to zoom a graph

### How to scale your graph manually—Ratio

**Step 1**
Click on the graph whose axes you want to change to make it the focused trace.

**Step 2**
Click the **Axes Scales** button on the toolbar, or select **Axes Scales** from the **Graph** menu. The **Axes Scales** dialog is displayed.

**Step 3**
Type in the new ranges for the X and Y axes in the format ‘n-n’, or choose an option for each from the drop-down list.

**Step 4**
Select **OK**. The axes on the selected graph will change to reflect your chosen settings.

**Hot Tip**
Select the **Zoom Out** command to resize the graph to its original settings.

See also:
- How to zoom a graph
- How to scale a graph automatically

### How to scale your graph automatically—Ratio

**Step 1**
Click on the graph whose axes you want to change to make it the focused trace.

**Step 2**
To autoscale the graph, select one of the following:
- Click the ** Autoscale XY ** button on the toolbar to autoscale both X and Y axes.
- Click the ** Autoscale X ** button on the toolbar to autoscale the X axis only.
- Click the ** Autoscale Y ** button on the toolbar to autoscale the Y axis only.

The focused trace will be scaled according to the option selected.

**Hot Tip**
Select the ** Zoom Out ** command to resize the graph to its original settings.

**See also:**
- How to zoom a graph
- How to scale your graph manually

**Single/Multi Graph—Ratio**

You can use the **Single/Multi Graph** command to toggle between a display of either one or multiple graph boxes in the Graphics area.

The command is accessible from the **Graph** menu or by selecting the **Single/Multi Graph** button from the Graphics toolbar.

Example of a single graph box display

Example of a multi graph box display

**Note**
Up to 100 graphs can be displayed in the Graphics area at the one time.
How to make a graphics template—Ratio

A graphics template lets you set up and store the style of a graphics report for future use. You can do this by adding labels and pictures to the background of the Graphics area and altering any axes styles etc. One advantage of this over the application's built-in reporting features is that you have more flexibility as to where you can position graph boxes and any text that you may want to appear (e.g. operator name, batch number etc.) and also where you add pictures, e.g. company logos etc.

Step 1

Start the application and open the data file from which you want to make a graphics template. Alternatively, collect some new data.

Step 2

Make as many graph boxes as you require in your template by using the Add Graph function.

Step 3

Adjust the graph boxes to the correct size and position them as you want them to appear in the template.

Step 4

Do any of the following, if required:

- Change the appearance of the graph boxes in the Graph Preferences dialog
- Use the Add Label function to append text to the Graphics area
- Use the Add Picture function to add pictures, such as your company's logo to the Graphics area

Step 5

From the File menu, select Print Preview. Use the Print Preview dialog displayed to check the template that you have created.

Step 6

When you are satisfied with the appearance of the graphics, select the Save As... command from the File menu. The Save As dialog is displayed.

Step 7

A list of stored data files will appear. Select the down arrow at the right of the Files of type field and select 'Graph Template' to list all the template files. Ensure that the Save only focused trace check box is cleared.

Step 8

Enter a name for the template in the File name field.
Step 9

Select **OK** to save the graphic template. The template will be used each time you create a new file.

**Note**
You can also make a graphic template in a similar manner from existing data. No data will be saved in the template.

**See also:**
- Example of a Graphics Template
- How to add a label to a graph
- How to add a picture to a graph
- How to move a label
- How to move a picture

**Ruler Rate—Ratio**

The ‘ruler’ function in the Ratio Application enables you to measure zero order reaction rates in ratio traces. Use the ruler after ratio data is collected to manually select the range over which to calculate the reaction rate. From this data, the slope the rate of reaction is determined.

To access the Ruler Rate dialog, do the following:

1. From the Graph menu select Cursor Modes. In the Cursor Modes dialog, select Ruler-Least Squares or Ruler-Point to Point option as the cursor mode. See More Information on Least Squares and Point to Point calculations.
2. Select a region of the trace that is relatively straight and place the cursor on the point of the trace where you wish to start drawing the ruler. Click and hold down the left mouse button and drag the cursor to the second point on the trace. A ruler line will draw as you move the cursor.
3. Release the mouse button. The Ruler Rate dialog is displayed.

The Ruler Rate dialog comprises the following:

- **Slope** Displays the slope of the line of best fit in the region you selected.
- **X Start/ X Stop** Displays the beginning and end X co-ordinates of the region you selected.
- **Y Start/ Y Stop** Displays the beginning and end Y co-ordinates of the region you selected.
- **Retry** Select Retry to re-select a region of the trace.
- **Add Label** Select Add Label to add a label to the graph that displays the slope of the line you have selected.

**More information on Least Squares and Point to Point calculations**

**Least squares slope calculation**

You can calculate the rate of a zero order reaction by manually selecting the range over which to calculate the reaction rate of a ratio plot using the ruler on a particular graph.

A linear equation is fitted to the data using a Ruler-Least Squares technique. The slope of the line of best fit is reported as the ratio rate in the Ruler Rate dialog using the previous zero order equation.
**Point to point slope calculation**

You can choose to calculate the point to point slope instead of the least squares slope value.

The point to point slope is calculated from the slope of a line drawn between two ordinate values at two, specified abscissa points.

![Point to point slope diagram](image)

\[
\text{Slope} = \frac{\Delta y}{\Delta x}
\]

If you choose OK or Add Label in the Ruler Rate dialog then a corresponding Results table will appear in the Report area that includes the manually selected abscissa, ordinate range, calculated reaction rate (Slope) and Standard Deviation (SD).

The SD results means that the system looks at the difference between the actual Y value and the fitted Y value after the linear least squares fit or point to point fit has been applied. The system takes the difference of each point over the selected region and calculates the SD using the following equation:

\[
S = \sqrt{\frac{1}{n-2} \sum_{i=1}^{n} (f_{m,i} - f_{c,i})^2}
\]

where,

- \( f_{m,i} \) are measured values
- \( f_{c,i} \) are calculated values
- \( n \) is the number of points

The Factor* Slope result is also displayed. This is the product of the Factor multiplied by the calculated ratio ruler slope. The Factor value (entered in the Analyze tab) also appears above the result table in the report.

**Note**

You can only perform a manual zero order rate calculation after the data is collected. Once the data is collected, select Cursor modes in the Graph menu and choose the ratio ruler type you require. You can then select the range over which to calculate the reaction rate.

**See also:**

[How to use the ratio ruler](#)
How to use the ratio ruler—Ratio

The ‘ruler’ function can be used for measuring zero order reaction rates in ratio traces. You can use the ruler after ratio data is collected to manually select the range over which to calculate the reaction rate. The slope is then determined and thus the rate of reaction.

To use the ruler:

Step 1

From the Graph menu, select Cursor Modes. Alternatively, select the button on the toolbar. The Cursor Modes dialog is displayed.

Step 2

Select Ruler—Least Squares to use a least-squares technique to determine the slope and thus the rate of the reaction over the selected range. Alternatively, select Ruler—Point to Point to use a line drawn between two selected points to determine the point to point slope and thus the reaction rate.

Step 3

Select OK to clear the Cursor Modes dialog and return to the Ratio Application window.

Step 4

Select a region of the trace that is relatively straight and then place the cursor on the first point on the trace where you wish to start drawing the ruler. Press and hold down the left mouse button.

Step 5

Move the cursor to the second point on the trace (you will see the ruler line drawing as you move the cursor) and release the mouse button.

The Ruler Rate dialog will be displayed to enable you to view the start and end positions of the calculation range that you selected. The value of the calculated slope is also displayed.

If you are not satisfied with the rate result, select the Retry button to repeat Steps 3 to 5, this time selecting another range.

Note

If you wish to exit the ratio ruler mode, select Cancel. The ratio ruler trace will be removed from the graph.

Step 7

Select Add Label to add the determined slope value of the ruler trace to the graph. The Ruler Rate dialog will be cleared.

If you do not wish to add a label, simply select OK to clear the Ruler Rate dialog.

The results of the manual determination of the reaction rate will appear in a table in the Report area. The ordinate and abscissa start and end values are reported as well as the calculated slope. The result table will also
include the calculated standard deviation as well as the Factor*slope result.

See also:
- Cursor modes

**Examples—Ratio**

- Multi-stage ratio run
- Multi-stage ratio calculations
- Example of a graphics template
- Using the stop time

**Example of a multi-stage ratio run—Ratio**

Number of Stages = 3, Single Cell ratio

<table>
<thead>
<tr>
<th>Stage</th>
<th>Cycle (min)</th>
<th>Stop (min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2</td>
<td>10</td>
</tr>
<tr>
<td>2</td>
<td>4</td>
<td>26</td>
</tr>
<tr>
<td>3</td>
<td>5</td>
<td>45</td>
</tr>
</tbody>
</table>

The Ave Time is set to 0.500 s

**Graphical representation of data points for the above example**

**Stage 1**: 0-10 minutes. The run will commence taking data readings at the set Ave Time of 0.5 s and cycle every 2 minutes. Five cycles will be completed in the first 10 minutes (Stop Time 10 mins) of the run.

**Stage 2**: 10-26 minutes. The run will continue taking data readings at the set Ave Time of 0.5 s but cycle every 4 minutes. Four cycles will be completed in the next 16 minutes (Stop Time 26 (10+16) mins) of the run.

**Stage 3**: 26-45 minutes. The run will continue taking data readings at the set Ave Time of 0.5 s but cycle every 5 minutes. Four cycles will be completed in the next 20 minutes (Stop Time 45 (10+16+20) mins) of the run.

See also:
- Example using the Stop time

**Example of multi-stage ratio calculations—Ratio**
The table to follow indicates the regions that would be selected to perform an Advanced analysis, on the above example.

Number of stages (steps) = 2

<table>
<thead>
<tr>
<th>Stage</th>
<th>Start (min)</th>
<th>Stop (min)</th>
<th>Order</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1</td>
<td>2</td>
<td>Zero</td>
</tr>
<tr>
<td>2</td>
<td>4</td>
<td>5</td>
<td>Zero</td>
</tr>
</tbody>
</table>

**Example of a Graphics Template—Ratio**

By using the *Add Label* command in the Cary Eclipse graphics system, you can customize a graphics template such as the one shown below:

The above is created as follows:

- The graph on the left is the collected data.
Example using the Stop time—Ratio

Example

Cycle time = 5 minutes
Stop time = 30 minutes

In the above example the run will complete 6 cycles of the chosen cells, each with a Cycle time of 5 minutes. The run will end at the selected Stop time of 30 minutes.

Note
If the Stop time was set to 28 mins, the run would continue for 30 mins, as the Cycle time overrides the Stop time.

See also:
Example of a multi-stage Ratio run

File Management—Ratio

- File menu
- Open dialog
- Save As dialog
- Filename extensions
- ASCII spreadsheet file
- Auto Store page
- How to combine data files into a batch file
- How to export collected data
- How to combine data files into a batch file

File menu—Ratio

Use the File menu to open or save a method or data file, print or preview a report, or exit the Cary Eclipse application.

Hot Tip
Highlight a method or data file in Explorer and simply drag and drop it onto the application to open it. Any data will be displayed in the Graphics area.

The following commands can be accessed from the File menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>New</td>
<td>Select New to clear all method, report and graphics information and create a new file containing the default settings.</td>
</tr>
<tr>
<td>Open Method…</td>
<td>Select Open Method to display the Open dialog where you can search for and load a stored method.</td>
</tr>
<tr>
<td>--------------</td>
<td>------------------------------------------------------------------------------------------------</td>
</tr>
<tr>
<td>Open…</td>
<td>Select Open to display the Open dialog where you can search for and load method, data, report, graph template and batch files, as well as other file types compatible with the Cary Eclipse software. When you load a batch file, the method which collected the data is loaded as well as the associated report and graphics layout.</td>
</tr>
<tr>
<td>Save Method As…</td>
<td>Select this option to display the Save As dialog and store the current method. You can use the Save As dialog to store the method, data, report, or graphics template separately or together as a Batch file. The data can also be saved as a comma delimited ASCII Spreadsheet file.</td>
</tr>
<tr>
<td>Save As…</td>
<td>Select this option to display the Save As dialog and store the collected data. You can use the Save As dialog to store the method, data, report, or graphics template separately or together as a Batch file. The data can also be saved as a comma delimited ASCII file.</td>
</tr>
<tr>
<td>Print Setup…</td>
<td>Select this option to display the Windows Print Setup dialog where you can select the printer, printer page size and orientation.</td>
</tr>
<tr>
<td>Print Preview…</td>
<td>Select this option to display the Print Preview dialog where you can view the current report as it will be printed.</td>
</tr>
<tr>
<td>Print…</td>
<td>Select this option to display the Windows Print dialog where you can choose the printer and pages to print as well as the number of copies you wish to print.</td>
</tr>
<tr>
<td>Explorer…</td>
<td>Select this option to run Windows Explorer. This may help you locate method and data files.</td>
</tr>
<tr>
<td>History List</td>
<td>This displays a list of the four most recently opened files.</td>
</tr>
<tr>
<td>Exit</td>
<td>Select this option to exit the Ratio application.</td>
</tr>
</tbody>
</table>

See also:
- Open
- Save As
- Print Preview
- Filename Extensions
- ASCII Spreadsheet File

## Open—Ratio

Use the dialog to open a file previously saved by the application. To access the Open dialog, select Open or Open Method from the File menu.

### Look in

The Look In field lists the available folders and files. Click the down arrow on the right of this field to view the location of the current folder in your computer's hierarchy. You can also select a new location simply by selecting a different folder.

The folders and files in the selected folder are displayed the list box below the Look In field.

#### Note

To open the folder in the level that is one higher than the current folder, select on the toolbar.

### Look in list box

The Look In List Box, which is directly below the Look In field, displays the folders and files in the selected location. To view the contents of a folder, double-click it.
Click to create a new folder. Type the name of the new folder in the highlighted text entry field that appears and press Enter. Then double-click on the new folder to open it.

Click to display the directory structure of the drive.

Click to display the files as a list.

Click to display files as a detailed list including the file size, file type and the date the file was last modified.

Click to preview the selected trace before you load it into the application.

**File name** Enter the name of the file you want to open in this field. The easiest way to do this is to find the file in the Look in list box and then select the file. However, you can type the full path of a file and then select Open to open the file.

**Files of type** Click on the arrow on the right of this field to view a list of the different types of application files that can be opened in the list box.

**Methods (*.FMRT)** Choose this file type to open a Method file. Only the method is loaded. No data is loaded.

**Data (*.FDRT)** Choose this file type to open a Data file. Only the data is loaded. The method that was used to collect the data is not loaded. The last used method or default method will be displayed.

**Report (*.FRRT)** Choose this file type to open a Report file. You can view the report in the Report area.

**Graph Template (*.FGRT)** Choose this file to open a Graph Template file. The file will set up the layout of the Graphics area including any bitmaps as well as the range and style of the graph axes.

**Batch (*.FBRT)** Choose this file type to open a Batch file. A Batch file includes data, the method used to collect the data, report and graphics setup.

**Spreadsheet Ascii (*.CSV)** This will open ASCII files that contain single comma delimited files with two or more columns.

**GRAMS (*.SPC)** Choose this file type to open a Cary Eclipse GRAMS data file. This allows you to use data files collected using a Cary Eclipse GRAMS system. The method that was used to collect the data is not loaded.

**ADL (*.ADL)** This will open and run program files generated in the Applications Development Language (ADL).

**All files (*)** This enables you to view all files in the directory so that you can open any compatible file.

[Tell me more about Filename Extensions](#)

**Overlay Data** Select this check box to overlay individual data files in one graph box. If this is not checked the data from each file will appear in individual graph boxes. This option is only available if the Open dialog has been accessed by selecting 'Open…' from the File menu.

**Hot Tip** Use the SHIFT or Control key to select multiple data files.

**Open** This button opens the highlighted file/s or the files listed in the File name field.
Save As—Ratio

Use the Save As dialog to save the data, method, report or graphics template individually or together as a Batch file.

**Note**
This dialog appears when you select either the Save Method As or Save As menu option in the File menu. When you select Save Method As this dialog appears with 'Methods' displayed in the Files of type field. When you select Save As this dialog appears with 'Batch' displayed in the Files of type field.

Traces (or data) that are currently ‘Visible’ in the Graphics area are saved, regardless of how they are displayed (full screen graph box or tile).

For example, if six graph boxes are displayed in the Graphics area and each graph box has one Visible trace then all six traces will be saved.

If you do not wish to save particular traces, make them not Visible in the Trace Preferences dialog. To save only the focused trace, select the Save only focused trace check box.

The Save As dialog comprises the following:

**Look In**
The Look In field lists the available folders and files. Click the down arrow on the right of this field to view the location of the current folder in your computer's hierarchy. You can also select a new location simply by selecting a different folder. The folders and files in the selected folder are displayed in the list box below the Look In field.

**Note**
To open the folder in the level that is one higher than the current folder, select on the toolbar.

**Look in list box**
The Look In list box, which is directly below the Look In field, displays the folders and files in the selected location. To view the contents of a folder, double-click it.

- Click to create a new folder. Type the name of the new folder in the highlighted text entry field that appears and press ENTER. Then double-click on the new folder to open it.
- Click to display the directory structure of the drive.
- Click to display the files as a list.
- Click to display files as a detailed list including the file size, file type and the date the file was last modified.
- Click to preview the selected trace before you load it into the application.

**File Name**
Enter the name of the file you want to save in this field. You can also type the full path of a file then select the Save button to save the file.
FileName Extensions

The Cary Eclipse system uses the filename extension to distinguish between data types (e.g. method or data) and also to differentiate applications. (e.g. Concentration or Ratio).

**Files of Type**
- Click on the arrow on the right of this field to view a list of the different types of application files that can be opened in the list box.

  **Methods (*.FMRT)** Choose this file type to only save the current method. Any collected data is not stored.

  **Data (*.FDRT)** Choose this file type to only save the data currently Visible in the **Trace Preferences** dialog (for any graph box). Note that the method used to collect the data is not stored with the data.

  **Report (*.FRRT)** Choose this file type to only save the contents currently shown in the Report area.

  **Graph Template (*.FGRT)** Choose this file type to save the layout of the Graphics area as a template. For example, any bitmaps you have added to your Graphics area will be saved as well as the range and style of the graph axes.

  **Batch (*.FBRT)** Choose this file type to save the data that is currently Visible in the **Trace Preferences** dialog (for any graph box), the method used to collect the data and the currently displayed report all together as a batch file. The graphics setup is also stored.

  **Spreadsheet Ascii (*.CSV)** Choose this file type to save the raw intensity data currently Visible in the **Trace Preferences** dialog (for any graph box) as a comma delimited ASCII Spreadsheet file (*.CSV) file. The data is saved as XY pairs delimited by a comma.

  **Note**
  This file type can be easily imported into a spreadsheet program. For example, Microsoft Excel. You can also double-click on the stored *.CSV file from Microsoft Explorer to load Excel automatically with the ASCII XY data displayed.

  **GRAMS (*.SPC)** Choose this file type to only save the data currently Visible in the **Trace Preferences** dialog (for any graph box) as a Cary Eclipse GRAMS data file.

**Tell me more about Filename Extensions**

**Save only focused trace** Select this option to save only the focused trace in the graph box. If this option is not selected, all visible traces in the graph box will be saved.

**Save** Click this button to save the file currently present in the **File name** field in the directory specified in the Look in field.

See also:
- File menu
- ASCII Spreadsheet file (*.CSV)

### Filename extensions—Ratio

The Cary Eclipse system uses the filename extension to distinguish between data types (e.g. method or data) and also to differentiate applications. (e.g. Concentration or Ratio).

The Syntax for file extensions is:

<table>
<thead>
<tr>
<th>Letter</th>
<th>Represents</th>
<th>Available Options</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
The following files may be opened and saved in the Ratio Application:

<table>
<thead>
<tr>
<th>Extension</th>
<th>Available action</th>
<th>File type</th>
</tr>
</thead>
<tbody>
<tr>
<td>*.FMRT</td>
<td>Open/Save</td>
<td>A <em>method</em> file (*.FMRT) consists of the software setup by which data is collected.</td>
</tr>
<tr>
<td>*.FDRT</td>
<td>Open/Save</td>
<td>A <em>data</em> file (*.FDRT) contains data collected that is currently visible in the Trace Preferences dialog (for any graph box).</td>
</tr>
<tr>
<td>*.FRRT</td>
<td>Open/Save</td>
<td>A <em>report</em> file (*.FRRT) contains the report generated by the Cary Eclipse about the data.</td>
</tr>
<tr>
<td>*.FGRT</td>
<td>Open/Save</td>
<td>A <em>graph template</em> file (*.FGRT) contains information that sets up the layout of the Graphics area including any bitmaps as well as the range and style of the graph axes.</td>
</tr>
<tr>
<td>*.FBRT</td>
<td>Open/Save</td>
<td>A <em>batch file</em> (*.FBRT) contains the collected data that was Visible in the Trace Preferences dialog (for any graph box), the method by which the data was collected and the report and graphics template.</td>
</tr>
<tr>
<td>*.CSV</td>
<td>Open/Save</td>
<td>An <em>ASCII spreadsheet</em> file (*.FCSV) has the same extension for all applications. It consists of raw intensity data (that was currently Visible in the Trace Preferences dialog (for any graph box) saved as XY comma-delimited pairs. This file can be imported into a spreadsheet program such as Microsoft Excel.</td>
</tr>
<tr>
<td>Data.*</td>
<td>Open</td>
<td>A Cary <em>DOS</em> (Data.*) file is the equivalent of a Cary Windows Data file—it does not contain any Batch information.</td>
</tr>
<tr>
<td>*.ADL</td>
<td>Open only</td>
<td>An <em>ADL</em> file (*.ADL) is a program file generated in the Applications Development Language (ADL).</td>
</tr>
<tr>
<td>*.SPC</td>
<td>Open/Save</td>
<td>A Cary <em>GRAMS</em> file (*.SPC ) is the equivalent of a Cary Eclipse Windows Data file—it does not contain any Batch information.</td>
</tr>
</tbody>
</table>
An ASCII Spreadsheet file consists of raw intensity data saved as XY comma-delimited pairs. You can choose to import or export Cary Eclipse files to ASCII files, which can then be imported in spreadsheet programs such as Microsoft Excel.

The format is as follows:

- Line 1 contains the sample name plus a comma
- Line 2 contains X and Y labels. The name of the X label must be the same as that used to label the graph.
  For example, in Ratio, the graph label is Time (secs).
- Lines 3 and onward contain the X,Y data
- After the last line of X,Y pairs there must be a blank line separating the data from any text audit strings or the end of the file.

For example, if a Ratio file was collected as:

\[
\begin{align*}
  s &= 0.025, \text{ intensity } = 1.422 \\
  s &= 0.075, \text{ intensity } = 1.403 \\
  s &= 0.125, \text{ intensity } = 1.384 \\
  &\text{etc.}
\end{align*}
\]

It would appear as follows in the ASCII file:

```
Ratio,
Time (secs): Intensity (a.u.)
0.025, 1.422
0.075, 1.403,
0.125, 1.384,
Ratio,
Collection time;
etc.
```

Microsoft’s Excel generates a CSV file with the above format.

There is also an alternative format for CSV files. For example,
How to combine data files into a batch file—Ratio

To combine a number of data files into a batch file, do the following:

Step 1
Select **Open**… from the **File** menu to display the Windows **Open** dialog.

Step 2
A list of stored batch files will appear. Select the down arrow to the right of the **Files of type** field and select 'Data' to list all the data files.

Step 3
Select the **Overlay Data** check box.

Step 4
Highlight the data files that you require. Hold down the CTRL key to select multiple files.

Step 5
Select **Open**. The highlighted data files will load into the application and appear in the same graph box.

Step 6
Select **Save As**… from the **File** menu to display the Windows **Save As** dialog.

Step 7
A list of stored data files will appear. Select the down arrow at the right of the **Files of type** field and select 'Batch' to list all the batch files. Ensure that the **Save only focused trace** check box is cleared.

Step 8
In the **File name** field, type in the file name for your new batch file.
Step 9

Select **Save** to create the new batch file. The current method will be stored with the batch file.

All the data files are now combined into the one batch file.

**How to export collected data—Ratio**

If you want to export data from a Cary Eclipse application into a third-party application, then you simply need to save your data as an Spreadsheet ASCII (*.CSV) file or a Rich Text Format (*.RTF) file. Spreadsheet ASCII files are recommended if you are planning to use the data in a spreadsheet program such as Microsoft® Excel. Rich Text Format files are recommended if you are planning to open the data in a word processor program such as Microsoft® Word.

To export collected data:

Step 1

From the **File** menu, select **Save As**. The **Save As** dialog is displayed.

Step 2

Click on the down arrow next to the **Files of type** list box and select the Spreadsheet ASCII (*.CSV) or Rich Text Format (*.RTF) option.

Step 4

Type the name of your new file in the **File name** field.

Step 5

Select **Save** to export your data to this new file.

**Software and Screen features—Ratio**

- **Menu line**
- **Status display**
- **Status line**
- **Buttons**
- **Function keys**
- **Right mouse menu**
- **How to remove or display the command buttons**

**Menu line—Ratio**

- **File menu**
- **Edit menu**
- **View menu**
- **Commands menu**
- **Setup menu**
- **Graph menu**
- **Help menu**
Status Line—Ratio

The Status Line is displayed at the very bottom of the Ratio Application window. The line will display the following information (from left to right):

<table>
<thead>
<tr>
<th>Hardware Status</th>
<th>This field displays status messages indicating the current instrument activity.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sequence Information</td>
<td>This field displays status messages indicating the current system activity.</td>
</tr>
<tr>
<td>Current Results</td>
<td>This field displays information on the last reading.</td>
</tr>
</tbody>
</table>

Ratio Application window

Buttons—Ratio

The major commands in this application can be accessed both from the menu items at the top of the main window and from the application buttons. These buttons, which appear in the left side of the Ratio Application window, comprise the following:

Hot Tip
If you wish to access these commands from the main menus, rather than from these buttons, you can remove the Command buttons from your application window. To do this, clear the Buttons command in the View menu. This will enlarge the Graphics area, making viewing and selection of scans easier.

<table>
<thead>
<tr>
<th>Button</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Setup</td>
<td>Click this button to display the Setup dialog where you can set up your Cary Eclipse for the collection, storage and analysis of the ratio run. You can also set up any accessories that you may have and define the reporting options.</td>
</tr>
</tbody>
</table>
| Zero   | Select this to zero the current ordinate value. This command can also be accessed by pressing F5.  
  Note
  Before you select Zero, ensure that you have placed the blank solution in the cell positions that are currently in the cell path. |
| Set Temp | Click Set temp to open the Set Temperature dialog where you can change the temperature during a ratio run.  
  Q. Why is the Set Temp button unavailable?  
  A. The Set Temp button is only accessible if you are using a thermostatted accessory.  
  (On the Accessories page, the Temperature control option must be selected.) Once you have started a run, you can access the Set Temp button by selecting the Pause button to pause the run. You can then select the Set Temp button. |
| Stop Time | Click the Stop Time button to access the Stop Time dialog. Use this dialog to extend the length of a ratio run while the run is in progress. Extending a run may be necessary if you decide whilst a run is in progress that the data collection period is too short. |
| Clear Report | Select Clear Report to remove all of the text that appears in the Report area. If you have saved the report, you can view the information again. To do this, simply open the report file using the Open… command in the File menu. |
| Recalculate | Use the pages accessible from the Recalculate option to perform re-calculations on data that has been analyzed. |
| Print   | Select Print to display the Windows Print dialog where you can choose what printer the
Function keys—Ratio

The function keys at the top of your keyboard perform basic Cary Eclipse operations simply and quickly.

<table>
<thead>
<tr>
<th>Function key</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>This displays the Help for the current dialog or window.</td>
</tr>
<tr>
<td>F4</td>
<td>Press F4 to display the Go to Wavelength dialog.</td>
</tr>
<tr>
<td>F5</td>
<td>Press F5 to perform an instrument Zero. The current intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before selecting Start if you want your data to start at 0 intensity. This command is also accessible from the Commands menu or by selecting the Zero button. <strong>Note</strong> Before you select Zero, ensure that you have placed the blank solution in the cell positions that are currently in the cell path. Select the Clear Zero command from the Commands menu to remove zero correction.</td>
</tr>
<tr>
<td>F6</td>
<td>Press F6 to display the Rapid Result dialog where you can read a blank and multiple sample readings without changing the application method. This command is also accessible from the Commands menu.</td>
</tr>
<tr>
<td>F9</td>
<td>If the application is online, press F9 to start a data collection. This is equivalent to selecting the Start button. If the application is not online, you will need to press F9 to first connect to the Cary Eclipse. Then press F9 to start the data collection. This command is also accessible from the Commands menu or by selecting the Start button. <strong>Note</strong> You will not be able to perform a Connect if another application is collecting data.</td>
</tr>
<tr>
<td>F12</td>
<td>Select this to stop the current measurement. This is equivalent to selecting the Stop button. This command is also accessible from the Commands menu or by selecting the Stop button.</td>
</tr>
</tbody>
</table>

See also:  
Go to Wavelength  
Zero  
Rapid Result
Right mouse menu—Ratio

The following commands are accessible by clicking the right mouse button on editable fields in the Cary Eclipse software:

- **Undo** If available, this reverts to the previous value before the Edit on the field.
- **Cut** If available, this cuts the highlighted text to the clipboard.
- **Copy** If available, this copies the highlighted text to the clipboard.
- **Paste** If available, this pastes the text from the clipboard to the cursor position.
- **Delete** If available, this deletes the highlighted text/number.
- **Select All** If available, this selects all entries.

How to remove or display the command buttons—Ratio

The command buttons (Setup, Zero, Pre-scan, Set Temp, Stop Time, Clear Report, Recalculate and Print) appear on the left side of the Ratio Application window.

To remove the command buttons

Step 1

From the Ratio Application window, select the View menu.

Step 2

Clear the Buttons menu item.

The buttons will disappear from the application screen, giving you more room to display reports.

To show the command buttons

Step 1

From the Ratio Application window, select the View menu.

Step 2

Select the Buttons menu item.

The buttons will re-appear in the application screen.

ADL—Ratio

- ADL Command Line
- ADL Hooks

ADL Command Line—Ratio

Introduction to ADL
Use the ADL Command Line to run Single-line or Multi-line Applications Development Language (ADL) commands that enable you to perform functions over and above that of the existing program functionality.

ADL is not a general purpose programming language. It has been developed specifically by Varian to meet the needs of spectroscopists. It can be used to handle all the data collected from the Cary Eclipse—both photometric values and entire data collections. ADL programs that you develop should be stored with the extension 'ADL'.

To access the ADL Command Line, select the ADL Command item from the Commands menu. You can also press ALT+Y to open the dialog.

Note
Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.

Software controls

Single Line
Selecting this lets you enter ADL commands on a single line into the Command Line entry box.

Q. Can I enter more than one ADL command on a single line?
A. Yes, provided that the commands are separated by a semicolon ‘;’.

Multi-line
The Multi-line capability enlarges the Command Line entry box. Use the Multi-line capability to create a more complex ADL program by adding a series of commands over multiple lines. When Multi-line mode is selected, the Quick Reference option becomes available.

Quick Reference
Select Quick Reference to open the Quick Reference dialog where you can easily add a command in the correct syntax to your ADL program. This option is only available for Multi-line commands.

A detailed description of all the commands in the Quick Reference is available by pressing the Help button on the Quick Reference dialog.

Browse
Select Browse to access the Open dialog where you can browse for and select an existing ADL program to load and run.

Command Line (entry box)
This entry box enables you to use ADL commands. You can enter single ADL commands into this field, or you can enter a series of ADL commands. If you have the entry type set to Single Line, you can review previous ADL entries by selecting the down arrow to the right of the list box.

The ADL commands in the Command Line are executed when you select OK.

Note
On entering this window, the Cary Eclipse will recall the last command or program executed from the Command Line entry box. The ADL commands entered in the last executed program are typically stored in the Varian\Cary Eclipse WinFLR\ADL directory on the drive on which the Cary Eclipse software is installed.

Hot Tip
You can rerun the ADL commands in the Command Line entry box from any screen in
the Ratio application by pressing Alt-X or selecting **Re-run ADL** from the **Commands** menu.

See also:
- [ADL Hooks](#)

## ADL Hooks—Ratio

The **ADL Hooks** command is accessible from the **View** menu.

Instead of writing a full ADL program you can use ADL hooks to link into an existing application. This is particularly useful if you want to make small changes to the way an application works, e.g. you want to pop up a dialog for the user to enter some specific data before the run starts. ADL hooks are links at specific points in an application at which you can run an ADL program. For example, ADL hooks have been supplied to run the following ADL programs:

### Note
Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.

<table>
<thead>
<tr>
<th>Hook</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>RT_Startup.ADL</td>
<td>This is run when the Ratio application starts. For this to to work the application must be started from a batch or method file that has ADL Hooks on.</td>
</tr>
<tr>
<td>RT_PreSeq.ADL</td>
<td>This runs when the <strong>Start</strong> button is selected.</td>
</tr>
<tr>
<td>RT_PostSeq.ADL</td>
<td>This runs at the end of the <strong>Start</strong> button sequence.</td>
</tr>
<tr>
<td>RT_Online.ADL</td>
<td>This runs when Cary Eclipse is detected online.</td>
</tr>
<tr>
<td>RT_OffLine.ADL</td>
<td>This runs when Cary Eclipse goes offline.</td>
</tr>
<tr>
<td>RT_Zero.ADL</td>
<td>This runs when the Zero button is selected.</td>
</tr>
<tr>
<td>RT_Recalc.ADL</td>
<td>This runs when the <strong>Recalculate</strong> button is selected.</td>
</tr>
<tr>
<td>RT_Shutdown.ADL</td>
<td>This runs when the application is shutting down.</td>
</tr>
<tr>
<td>RT_PreRead.ADL</td>
<td>This runs just before a read is taken.</td>
</tr>
<tr>
<td>RT_PostRead.ADL</td>
<td>This runs just after a read is taken (It has the reading result stored in the <strong>UserResult</strong> variable.)</td>
</tr>
<tr>
<td>RT_PreSetup.ADL</td>
<td>This runs when the <strong>Setup</strong> button is selected.</td>
</tr>
<tr>
<td>RT_PostSetup.ADL</td>
<td>This runs when the <strong>OK</strong> button on the <strong>Setup</strong> dialog is selected.</td>
</tr>
<tr>
<td>RT_CellChange.ADL</td>
<td>This runs instead of the Cary Eclipse driving the Multicell Holder. It enables you to add your own instructions for the Cary Eclipse Multicell Holder or to use your own cell changing accessory.</td>
</tr>
<tr>
<td>RT_PreStage.ADL</td>
<td>This runs prior to each collection stage. It determines the stage by checking the <strong>UserResult</strong> variable as <strong>UserResult = stage number</strong>.</td>
</tr>
<tr>
<td>RT_PreUserRecalc.ADL</td>
<td>This runs just after the <strong>Recalculate</strong> button has been selected and performs any</td>
</tr>
</tbody>
</table>
Help and Safety information—Ratio

- Help menu
- Safety home page

Help menu—Ratio

Use the Help menu to view the online Help and software information for this application.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ratio Help</td>
<td>Select this to display the Ratio Home Page. This contains a list of online Help topics specific to the Ratio Application.</td>
</tr>
<tr>
<td>Help Topics</td>
<td>Select this to display the Cary Eclipse Windows Online Help Home Page.</td>
</tr>
<tr>
<td>Tip of the day</td>
<td>View useful tips about using your Cary Eclipse instrument and software.</td>
</tr>
<tr>
<td>Eclipse Tour</td>
<td>Select this to display the Cary Eclipse Multimedia Online Help. This will give you an animated introduction to the Cary Eclipse software and hardware.</td>
</tr>
<tr>
<td>About…</td>
<td>Select About to access a dialog that displays information about the application as well as Varian email and Internet contact information. To clear this About dialog, select OK.</td>
</tr>
</tbody>
</table>

Safety Home Page

Click on the desired index entry to view the Help.

- About Safety
- Lamp module
Ratio How To—Ratio

Select the procedure you require below for a step-by-step informative guide on running the application software.

Procedural

Calculations and reporting

- How to determine the autofluorescence of a reaction
- How to subtract autofluorescence from your reaction
- How to create a ratioed trace
- How to use the Gryniewicz equation
- How to use the ratio ruler
- How to generate a report from previously collected data
- How to use advanced reporting options

General

- How to email your results
- How to export collected data
- How to combine data files into a batch file
- How to remove or display the command buttons

See also:
- Ratio Home Page
- Graphics How To

Autofluorescence correction—Ratio

To determine the true fluorescence value of a reaction it is necessary to subtract the autofluorescence of the reaction. That is it is necessary to subtract the intrinsic cell fluorescence from the overall calculated fluorescence of the reaction.

To determine an autofluorescence value of a reaction:

If using **Sample Averaging**, place a sample in a cuvette and open **Advanced Reads** to take a reading to obtain your autofluorescence value.

**Alternatively**

If you are using **information from a continuum**, place your sample in a cuvette to be read over time and using
Kinetics to collect and average the data points get your autofluorescence value.

Note
If you are using multiwavelength analysis you will need to do this for both wavelengths. If you are using single wavelength analysis you will only be require to do this once and collect one autofluorescence value.

Status Display
Select this check box to display the Status Display window. This window allows you to view the status of various parameters during the data collection.

See also: Subtracting autofluorescence

Subtracting Autofluorescence—Ratio

To determine the true fluorescence value of a reaction it is necessary to subtract the autofluorescence of the reaction. That is it is necessary to subtract the intrinsic cell fluorescence from the overall calculated fluorescence of the reaction.

The Autofluorescence value has to be subtracted post run.

Step 1

Using the Trace Preferences option display the trace(s) from your run as separate graphs.

The individual traces will be listed as your substance run at a particular wavelength. Select the individual traces by clicking in the Visible column next to the trace.

Step 2

Return to the main graphics area and select the trace you wish to correct by subtracting an autofluorescence value.

Step 3

In the Setup dialog select the Analyze tab and check the Subtract autofluorescence check box.

Step 4

In the Enter value to subtract from trace entry field, type in the autofluorescence value obtained previously for the selected trace. Select OK.

Step 5

A new corrected trace will be generated showing the initial trace minus the autofluorescence value, and will be named as such _AF1. This trace will be listed in the Trace Preferences window but has not been saved. Save the new trace as a batch file.

Step 6
Repeat this procedure for each trace.

**Status Display**

Select this check box to display the Status Display window. This window allows you to view the status of various parameters during the data collection.

See also:
- Determining autofluorescence
- Using traces with autofluorescence subtracted to obtain a ratioed trace

**How to resize graph boxes—Ratio**

Step 1
Click on the graph that you want to resize so that the highlight box appears around it.

Step 2
Move the cursor to the edge of the highlight box and place it over one of the square marks (e.g. ■) in the box until it turns into a double headed arrow (↔).

Step 3
Click and hold the left mouse button and drag the box to the desired size.

Step 4
Release the left mouse button.

See also:
- How to alter the look of your graphs

**How to zoom a graph—Ratio**

To zoom a graph, do the following:

Step 1
Click on the button on the toolbar, or select **Cursor Mode** from the **Graph** menu. The **Cursor Modes** dialog will appear.

Step 2
Select **Free** and select the **OK** button.

Step 3
Click and hold down left mouse button in the highlighted graph box.

Step 4
Drag the mouse sideways and up or down. You will see a zoom box made up of a dotted line appear on the screen. (See picture below.)

The dotted line is a zoom box.

Hot Tip
To move the box on the screen, hold down the left and right mouse buttons together. You can then position the box anywhere in the graph box. Releasing the right mouse button will still allow you to resize the zoom box.

Step 5

Release the left mouse button. The area defined by the zoom box will be redrawn to fill the entire graph box, and the scans within the box will be enlarged accordingly. The abscissa (X) and ordinate (Y) coordinates displayed below the graph will also be changed to reflect the positions of the corners of the zoom box.

You can repeat this process if you need to zoom in even further.

Note
The system may limit the extent to which you can Zoom.

See also:
How to scale your graph manually
How to scale your graph automatically

How to take the derivative of a trace

Some uses of derivatives are:

- To resolve partly overlapping peaks
- To quantitatively determine trace compounds
- To characterize pure compounds
- Purity testing

To take the derivative of a collected trace and plot the result, follow these steps:

Step 1

Click on the trace to be smoothed. It will become red (highlighted).

Step 2

Click the Selected Trace or Focused Trace button. The name of the highlighted trace will appear in the calculation display field.

Step 3

In the Trace group, select your derivative order in the Operation field. Choose from derivatives 1 to 4 (Deriv1, Deriv2, Deriv3, Deriv4). You may have to scroll to find the derivative you require.
Step 4

Enter a Filter Size and an Interval in the corresponding fields.

Step 5

In the Display Options group, select New Graph to plot the result in a new graph box, or Selected Graph to plot the result in the current graph box.

Step 6

Click the Apply button in the Trace group. Your Deriv selection will be displayed before the trace name in the calculation display field.

Step 7

Click on the equals (=) button. The result will be displayed in the Graphics area.

See also:
About derivatives

How to smooth a trace

To smooth a collected trace and plot the result, follow these steps:

Step 1

Click on the trace to be smoothed. It will become red (highlighted).

Step 2

Click on the Maths button in the toolbar (it's on the far right) or select the Maths menu.

Step 3

Click the Selected Trace or Focused Trace button. The name of the highlighted trace will appear in the calculation display field.

Step 4

In the Trace group, select Smooth in the Operation field. (You may have to scroll to find it.)

Step 5

Enter a Filter Size and an Interval in the corresponding fields.

Step 6

In the Display Options group select New Graph to plot the result in a new graph box, or Selected Graph to plot the result in the current graph box.

Step 7
Click the Apply button in the Trace group. The word ‘Smooth’ will appear before the trace name in the calculation display field.

Step 8

Click on the equals (=) button. The result will be displayed in the Graphics area.
### Scan Home Page

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<th>Use the...</th>
<th>Show me...</th>
</tr>
</thead>
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<td>• Setup dialog</td>
<td></td>
</tr>
<tr>
<td>Performing scans</td>
<td>• Commands menu</td>
<td>• How to perform a fluorescence scan&lt;br&gt;• How to perform a phosphorescence scan&lt;br&gt;• How to perform a bio-/chemi-luminescence scan</td>
</tr>
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<td>• How to generate a report from previously collected data&lt;br&gt;• How to use advanced reporting options</td>
</tr>
<tr>
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<td>• Maths Calculator&lt;br&gt;• User Data Form</td>
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<tr>
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</tr>
<tr>
<td>Help and safety information</td>
<td>• Help menu&lt;br&gt;• Safety Home Page</td>
<td></td>
</tr>
</tbody>
</table>

See also: [Scan How To](#)

### Scan Application

#### Introduction

Use the Scan application to scan samples across a wavelength or wavenumber range and manipulate the collected data. You can choose various display modes for the collected data depending on the type of sample you are measuring and the Cary Eclipse accessories that you are using.

To access the Scan Application, click the Windows **Start** button, point to **Programs**, **Cary Eclipse** and then select **Scan**.
Software controls

The application comprises the following:

**Instrument Status Readings**

The Instrument Status Readings, which display the Y and X values of the graph axes are displayed in the top left and right corners of the Scan Application window.

- **Y value**
  Displays the current Y value, usually intensity. This value is continually updated during a data collection.

- **X value**
  Displays the current X values for both excitation and emission. You can choose to display these values as either Angstroms, Wavelengths, Wavenumbers or Electron Volts. Select how the values will be displayed on the Cary page of the Setup dialog.

**Start and Stop buttons**

The Start and Stop buttons at the top of the Scan Application window are used to start and stop instrument analyses. These buttons can also be accessed from the Commands menu.

Tell me more about the Start and Stop buttons

**Graphics area**

The Graphics area refers to the plotting area of the application. It can contain traces of collected data, labels and pictures. In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area defined by the axes is referred to as a graph box.

To display or hide the Graphics area, select the Graphics command on the View menu. Double-clicking on the Graphics area enables you to toggle between half or full size display sizes. To manually size the Graphics area, place the mouse cursor over the border between the Graph and Report areas until the cursor changes to a double arrow ( ). Then hold down the mouse and drag it up or down to resize the areas.

Tell me more about the Graphics area

**Report area**

Use the Scan Report area to view the current report. The report may include the results of your data collection, the method parameters used to collect the results and any graphics you have included. You can select the information to include in your report in the Reports tab of the Setup dialog.

To display or hide the Report area, select the Report command from the View menu. Double-clicking on the Report area enables you to toggle between half or full size display sizes. To manually size the Report area, place the mouse cursor over the border between the Graph and Report areas until the cursor changes to a double arrow ( ). Then hold down the mouse and drag it up or down to resize the areas.

You can copy the generated report onto the Windows clipboard. To do this, highlight the report (or a relevant section of it) and press CTRL+C. You can then paste the report into a word processor such as Microsoft® Word.

**Hot Tip**

You can edit the Scan Report by pressing CTRL+R or selecting the Edit Report command from the Edit menu.

**Commands**

The Scan Application window contains the following commands, all of which can also be accessed from the various menus in the application.
Click on the buttons to view information about them:

- Setup ...
- Zero
- Pre-scan ...
- G-Factor
- Clear report
- Create report
- Print ...

**Hot Tip**

By becoming familiar with these commands (Setup, Zero, Pre-scan, etc.), you can remove these buttons from the Scan window. This will enlarge the Graphics and Report areas and make it easier to view results. To do this, clear the Buttons item in the View menu.

**Menus**

The Scan Application contains the following menus:

- File
- Edit
- View
- Commands
- Setup
- Help

See also:

- Status Display
- Status Line
- Keyboard function keys
- Scan Home Page

**Getting ready for a run—Scan**

- **Setup dialog**
- Setup menu
- **Cary page**
  - Bio-/Chemi-luminescence and Phosphorescence Options dialog
  - Gate Time for Bio-/Chemi-luminescence
  - Gate Time for Phosphorescence
  - How many flashes?
- **Options page**
  - About smoothing
- **Accessories page**
  - Multicell/Temperature accessory
  - Multicell Changer
  - Wellplate Reader accessory
  - Display Options dialog
  - Wellplate Sample List
  - Go To Well dialog
  - Polarizer accessory
- **Reports page**
  - Report toolbar
  - Create Report
  - Clear Report
  - Label Options
  - Peak Labels
  - How to use the advanced reporting options
- **Auto Store page**
  - How to email the results

**Setup—Scan**

Use the Setup dialog to set up your instrument and accessories for a scan. You can also use the dialog to set up the Analysis, Report and Storage parameters associated with the run.
To access the **Setup** dialog select the **Setup** button from the Scan Application window or the **Setup** command from the **Setup** menu.

The dialog comprises the following tabbed pages:

- Cary
- Options
- Accessories
- Reports
- Auto Store

See also: [Scan Application window](#)

**Setup menu—Scan**

Use the **Setup** menu to access the parameters for setting up a Scan.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Setup</strong></td>
<td>Choose <strong>Setup</strong> to display the <strong>Setup</strong> dialog where you can set up your instrument for a Scan measurement. The Setup dialog can also be accessed from the Setup command button.</td>
</tr>
<tr>
<td><strong>Instrument</strong></td>
<td>Choose <strong>Instrument</strong> to display the <strong>Cary</strong> page. You can use this page to set up the instrument parameters associated with the data collection.</td>
</tr>
<tr>
<td><strong>Options</strong></td>
<td>Choose <strong>Options</strong> to display the <strong>Options</strong> page where you can set up your filters and set the detector voltage. You can also use this page to select the way the collected data is displayed in the Graphics area, and the type of smoothing applied (if any).</td>
</tr>
<tr>
<td><strong>Accessories</strong></td>
<td>Choose <strong>Accessories</strong> to display the <strong>Accessories</strong> page. You can use this page to set up the accessory parameters associated with the data collection.</td>
</tr>
<tr>
<td><strong>Reports</strong></td>
<td>Choose <strong>Reports</strong> to display the <strong>Reports</strong> page. You can use this page to set up the report parameters associated with data collection.</td>
</tr>
<tr>
<td><strong>Auto Store</strong></td>
<td>Choose <strong>Auto Store</strong> to display the <strong>Auto Store</strong> page where you can specify if and when data will be stored, and in what format.</td>
</tr>
</tbody>
</table>

See also:
- Setup
- Cary
- Options
- Accessories
- Reports
- Auto Store

**Cary—Scan**

Use the **Cary** page to set up the instrument parameters associated with the data collection.

To access the **Cary** page, select the **Cary** command from the **Setup** menu. You can also access the page from the **Setup** dialog, by selecting the **Cary** tab. To access the **Setup** dialog, select the **Setup** button from the Scan Application window or the **Setup** command from the **Setup** menu.

**Instrument setup**
**Data Mode**  
Select from one of the following three data modes:

*Fluorescence*
Fluorescence refers to the emission of light from an excited single state. The fluorescence signal is measured when the lamp flashes.

*Bio-/Chemi-luminescence*
Bio-/Chemi-luminescence refers to bio- or chemi-luminescence, where the emission is generated via a chemical or biochemical reaction.

*Phosphorescence*
Phosphorescence refers to the emission of light from an excited triplet state. Phosphorescence is measured after the lamp has stopped flashing.

**Options**  
Select **Options** to open the **Bio-/Chemi-luminescence/Phosphorescence Options** dialog where you can set parameters such as the Decay, Delay and Gate times for your sample.

This **Options** button is only visible if you have selected **Bio-/Chemi-luminescence** or **Phosphorescence** as the **Data Mode**.

**Scan Setup**

**Scan Mode**  
Select from one of the following three scan modes:

*Excitation*
Select **Excitation** to perform an excitation scan, which is a qualitative absorption spectrum. During the scan, the emission monochromator is set to a fixed wavelength and the excitation monochromator is scanned over a wavelength range. This is often referred to as the emission spectrum or the fluorescence spectrum. This option is not available if you have selected **Bio-/Chemi-luminescence** as the **Data Mode**.

**Example of an emission spectrum**

*Emission*
Select **Emission** to perform an emission scan. During the scan, the excitation monochromator is set to a fixed wavelength and the emission monochromator is scanned over a wavelength range. This results in a fluorescence excitation spectrum.

**Example of an excitation spectrum**

*Synchronous*
Select **Synchronous** to scan the excitation and emission monochromators synchronously with a fixed delta (which is a real number and may be zero), between both monochromators. Delta may be entered in wavelength (nm) or wavenumber (cm⁻¹) for the scan performed. This option is not available if you have selected **Bio-/Chemi-luminescence** as the **Data Mode**.

**Example of a synchronous scan**

**X Mode**  
To set the X axis data display, select from the following options, which are accessible from the drop down menu:

- Angstroms
- Wavelength
- Wavenumber
- Electron volts

This option is not available if you have selected Synchronous as the Scan Setup mode.

**Start**
Enter the value (in angstroms, wavelength, wavenumber or electron volts) at which you want the Cary Eclipse to start to collect data.

If you have selected Excitation as the Scan Setup mode, the Start value refers to the excitation monochromator.

If you have selected Emission as the Scan Setup mode, the Start value refers to the emission monochromator.

If you have selected Synchronous as the Scan Setup mode, the Start value refers to the excitation monochromator. Note that the emission monochromator always starts at a higher wavelength than the excitation monochromator (Start + Delta).

**Stop**
Enter the value (in angstroms, wavelength, wavenumber or electron volts) at which you want the Cary Eclipse to stop collecting data.

If you have selected Excitation as the Scan Setup mode, the Stop value refers to the excitation monochromator.

If you have selected Emission as the Scan Setup mode, the Stop value refers to the emission monochromator.

If you have selected Synchronous as the Scan Setup mode, the Stop value refers to the excitation monochromators. Note that the emission monochromator always ends at (Stop + Delta).

**Emission/ Excitation/ Delta**
Depending upon the Scan Setup mode selected, of the following options is displayed.

These options are not available if you have selected Bio-/Chemi-luminescence as the Data Mode.

**Emission**
This option is displayed if you select Excitation as the Scan Setup mode. Enter the value that the Excitation monochromator will be set to.

**Excitation**
This option is displayed if you select Emission as the Scan Setup mode. Enter the value that the Emission monochromator will be set to.

**Delta**
This option is displayed if you select Synchronous as the Scan Setup mode. Enter the Delta in wavelength (nm) or wavenumber (cm-1). This enables you to set a fixed difference between the wavelength (or wavenumber) at which the excitation and emission monochromators will be synchronously scanned. The Delta range is 0–500 nm (or -7575.76– wavenumbers). If Delta is set to zero, both monochromators will be scanned at the same wavelength (or wavenumber).

**Zero order**
When Excitation is selected as the Scan Setup mode, select this command to set the Emission monochromator to zero order (white light).

When Emission is selected as the Scan Setup mode, select this command to set the Excitation monochromator to zero order (white light).
This option is not available if you have selected **Synchronous** as the **Scan Setup** mode.

**Excitation Slit**

Set the slit width of the excitation monochromator. This option is not available if you have selected **Bio-/Chemi-luminescence** as the Data Mode.

*As a general rule:*

At a fixed emission slit width, increasing the excitation slit width will result in an increase in the fluorescence excitation intensity.

**Emission Slit**

Set the slit width of the emission monochromator.

*As a general rule:*

At a fixed excitation slit width, a two-fold increase in the emission slit width will result in a four-fold increase in the emission intensity. Accordingly, a two-fold decrease in the emission slit width will result in a four-fold decrease in the emission intensity.

**3-D Mode**

The 3-D Mode options are not available if you have selected **Bio-/Chemi-luminescence** as the Data Mode.

**Ex/Em/Delta Stop**

Depending upon the Scan Setup mode selected, of the following options is displayed:

**Em Stop**

This option is displayed if you select **Excitation** as the scan mode. Enter the stop wavelength of the Emission monochromator (in angstroms, wavelength, wavenumber or electron volts). The number of scans performed is equal to the \[(Excitation stop - Excitation)/(Excitation increment) + 1\].

**Ex Stop**

This option is displayed if you select **Emission** as the scan mode. Enter the stop wavelength of the Excitation monochromator (in angstroms, wavelength, wavenumber or electron volts). The number of scans performed is equal to the \[(Emission stop - Emission)/(Emission increment) + 1\].

**Delta Stop**

This option is displayed if you select **Synchronous** as the scan mode. Enter the **Delta Stop** value in cm\(^{-1}\) or nm. The **Delta** will be incremented after each scan by the value entered in the **Delta Increment** field until the **Delta Stop** value is reached. Therefore, the number of scans performed is equal to the \[(Delta stop - Delta)/(Delta increment) + 1\].

**Ex/Em/Delta Increment**

Depending upon the Scan Setup mode selected, of the following options is displayed:

**Em Increment**

This option is displayed if you select **Excitation** as the data mode. Enter the step, in nm, that the Emission monochromator moves prior to the next scan. For example, if you perform an excitation scan from 400–600nm with an emission wavelength of 350 nm, and an emission increment of 10 nm, then the next scan will be performed with an emission wavelength of 360 nm between 400–600nm. This is repeated until the Emission Stop wavelength is reached.

**Ex Increment**

This option is displayed if you select **Emission** as the data mode. Enter the step, in nm, that the Excitation monochromator moves prior to the next scan. For example, if you perform an emission scan from 400–600nm with an excitation wavelength of 350 nm, and an excitation increment of 10 nm, then the next scan will be performed with an excitation wavelength of 360 nm between 400–600nm. This is repeated until the Excitation Stop wavelength is reached.
wavelength is reached.

**Delta Increment**
This option is displayed if you select Synchronous as the data mode. Enter the Delta Increment, in nm or cm⁻¹. When you perform a series of synchronous scans, you can increment the Delta after each scan by the amount entered in this field. Both monochromators are scanned from the Start wavelength to the Stop wavelength and the Delta will be incremented until the Delta Stop has been reached.

Examples of Excitation, Emission and Synchronous 3D collects

Scan Control

**Speed**
These Speed options are only available if you have selected Angstroms or Wavelength as the X Mode.

*Slowest, Slow, Medium, Fast, Fastest, Survey*
These buttons control the speed at which the Cary Eclipse moves. They are available if you select an X Mode of Angstroms or Wavelength, and will automatically assign the values for the Scan rate, Averaging time and Data interval parameters. Click on the links below to view these preset values.

**Angstroms**

<table>
<thead>
<tr>
<th>Scan rate</th>
<th>Scan rate</th>
<th>Averaging time</th>
<th>Data interval</th>
</tr>
</thead>
<tbody>
<tr>
<td>Slowest</td>
<td>300 A/min</td>
<td>1.0000 s</td>
<td>5.0000 A</td>
</tr>
<tr>
<td>Slow</td>
<td>1200 A/min</td>
<td>0.5000 s</td>
<td>10.000 A</td>
</tr>
<tr>
<td>Medium</td>
<td>6000 A/min</td>
<td>0.1000 s</td>
<td>10.000 A</td>
</tr>
<tr>
<td>Fast</td>
<td>12000 A/min</td>
<td>0.1000 s</td>
<td>20.000 A</td>
</tr>
<tr>
<td>Fastest</td>
<td>96000 A/min</td>
<td>0.0125 s</td>
<td>20.000 A</td>
</tr>
<tr>
<td>Survey</td>
<td>240000 A/min</td>
<td>0.0125 s</td>
<td>50.000 A</td>
</tr>
</tbody>
</table>

**Wavelength (nm)**

<table>
<thead>
<tr>
<th>Scan rate</th>
<th>Scan rate</th>
<th>Averaging time</th>
<th>Data interval</th>
</tr>
</thead>
<tbody>
<tr>
<td>Slowest</td>
<td>30.000 nm/min</td>
<td>1.0000 s</td>
<td>0.5000 nm</td>
</tr>
<tr>
<td>Slow</td>
<td>120.000 nm/min</td>
<td>0.5000 s</td>
<td>1.0000 nm</td>
</tr>
<tr>
<td>Medium</td>
<td>600.000 nm/min</td>
<td>0.1000 s</td>
<td>1.0000 nm</td>
</tr>
<tr>
<td>Fast</td>
<td>1200.000 nm/min</td>
<td>0.1000 s</td>
<td>2.0000 nm</td>
</tr>
<tr>
<td>Fastest</td>
<td>9600.000 nm/min</td>
<td>0.0125 s</td>
<td>2.0000 nm</td>
</tr>
<tr>
<td>Survey</td>
<td>24000.000 nm/min</td>
<td>0.0125 s</td>
<td>5.0000 nm</td>
</tr>
</tbody>
</table>

**Manual**
Select Manual to precisely set the Cary Eclipse data collection. Use the Scan Rate, Averaging Time and Data Interval fields (which are inter-related) to do this. Note that setting a value in one field may affect one or both of the others.

**Scan Rate**
This sets the rate at which the fluorescence spectrophotometer scans the abscissa or X axis. As the Scan Rate and Data Interval values are inter-dependant, when you adjust the Scan Rate, the Data Interval will be adjusted accordingly. Therefore you can either set the Scan Rate manually or set the Data Interval rate and allow the software to calculate the corresponding Scan Rate.
The **Scan Rate** field will be unavailable in the following circumstances:

- You have selected **Bio-/Chemi-luminescence** or **Phosphorescence** as the Data Mode. In this case, the Scan Rate will be dependant on the values set for the Averaging Time and Data Interval, and will be adjusted accordingly.
- You have selected **Wavenumber** or **Electron Volts** as the X Mode. In this case, the Scan Rate will be dependant on the values set for the Averaging Time and Data Interval, and will be adjusted accordingly.
- You have selected one of the **Speed** settings (Slowest, Slow, Medium, Fast, Fastest, Survey). As a result, the Scan Rate will be preset, depending upon the setting you have chosen.

**Averaging Time**
Enter the signal averaging time to be used during data collection. The way in which the the **Averaging Time** is calculated depends upon the **Data Mode** you have chosen.

**Fluorescence**
In **Fluorescence** mode, the data point at each wavelength will be an average of the signals from a number of flashes that occur during this averaging time, that is: Averaging Time/0.0125. The range is 0.0125–1000 s.

**Phosphorescence**
In **Phosphorescence** mode, the data point at each wavelength will be the average of the signals from a number of gate time measurements that occur during this averaging time, that is: Averaging Time/Gate Time. These signals are accumulated from successive decay curves so that the actual time taken to generate a point at a single wavelength will be approximately: (Averaging Time/Gate Time x Total Decay Time). In phosphorescence mode, the Averaging Time ranges from the Gate Time to a maximum of 1000 s.

**For bio-/chemi-luminescence:**
In **Bio-/Chemi-luminescence** mode, the data point at each wavelength will the average of the signals from a number of gate time measurements that occur during this averaging time, that is: Averaging Time/Gate Time. In bioluminescence mode, the Averaging Time ranges from the Gate Time to a maximum of 1000 s.

**Understanding Average Time and Gate Time for Phosphorescence**

**Data Interval**
Enter the value at which to collect and display a data point. This value determines the size of the steps between each data point in the X range. The smaller the interval, the more detail there will be in the collected traces. Therefore, to obtain a plot with high resolution, you should set a small interval. With a large interval, the data points will be far apart and the scan will be completed more quickly.

**Status Display**

Select this check box to display the **Status Display** window where you can view the status of various parameters during the data collection.

See also:
- **Scan Application window**
- **Bio-/Chemi-luminescence/Phosphorescence Options**
- **Accessories**
Use the Bio-/Chemi-luminescence/Phosphorescence Options dialog to set up the parameters associated with these two measurement modes.

To access the Bio-/Chemi-luminescence/Phosphorescence dialog, select the Options button from the Cary page.

**Note**
The Options button is only visible on the Cary page if you have selected Bio-/Chemi-luminescence or Phosphorescence as the Data mode.

**Dialog controls**

The Bio-/Chemi-luminescence/Phosphorescence Options dialog comprises the following options:

- **Total decay time**: Enter, in seconds, the total decay time for your sample. The total decay time is the time that it takes for the phosphorescence signal to decay to dark levels (zero). The lamp will not flash again until the total decay time has elapsed. The range is 0.001–1800.000 s.
  
  **Note** This option is not available in Bio-/Chemi-luminescence mode.

- **No. of flashes**: Enter the number of lamp flashes required, which is how many times the lamp flashes consecutively before the delay time is counted.

  If you are measuring phosphorescence samples that have a relatively long decay time (that is, a number of seconds), you can setup the Cary Eclipse to pulse the lamp multiple times, as this will increase the intensity of the phosphorescent signal. For short lived phosphorescent samples, multiple flashes will not impact on the intensity of the signal, and therefore only one flash is required.

  **How many flashes should I perform?**

  **Note** This option is not available in Bio-/Chemi-luminescence mode.

- **Delay time**: Enter, in milliseconds, the delay time for your sample. The delay time is the time that elapses between the last flash and the beginning of data collection. The default value is 0.100 ms. Setting a delay time ensures that there is no lamp effects in the phosphorescence decay curve. The range is 0.000–5000.000 s.

  **Note** This option is not available in Bio-/Chemi-luminescence mode.

- **Gate time**: Enter, in milliseconds, the gate time for your sample. The gate time is the overall reading time of the emission signal. The range is from 10µs–10s.

Understanding Average Time and Gate Time for Phosphorescence scans
Understanding Gate Time for Bio-/Chemi-luminescence scans
Example of the Bio-/Chemi-luminescence/Phosphorescence Options

Gate Time is set on Options page, which is accessible from the Cary page.

The following diagram highlights the Gate Time setting.

The Gate Time is set to 10 and the Intensity is equal to $I_T$. 

See also:
Cary page
Understanding Average Time and Gate Time for Phosphorescence Scans—Scan

For Phosphorescence scans, there is a relationship between Average Time (the time for which data is averaged) and Gate Time (the overall reading time of the emission signal).

Averaging is calculated as follows:

The data point at each wavelength will be the average of the signals from a number of gate time measurements that occur during this averaging time, that is:

**Averaging Time/Gate Time**

These signals are accumulated from successive decay curves so that the actual time taken to generate a point at a single wavelength will be approximately:

**(Averaging Time/Gate Time x Total Decay Time)**

In phosphorescence mode, the Averaging Time ranges from the Gate Time to a maximum of 1000 s.

**Note:**
Gate Time is not used if you select Fluorescence as the Data Mode.
**Average Time** is set on the **Cary** page of the **Setup** dialog. **Gate Time** is set on **Options** page, which is accessible from the **Cary** page.

The following diagrams highlight the relationship between **Average Time** and **Gate Time**.

**Example 1**

When the Average Time and Gate Time are set to the same value:

<table>
<thead>
<tr>
<th>Average Time/Gate Time</th>
<th>1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intensity</td>
<td>1 (Area $I_A$, $I_B$)</td>
</tr>
</tbody>
</table>

where:

- Total Decay Time: 40 ms
- Delay Time: 10 ms
- Average Time: 20 ms
- Gate Time: 20 ms

**Example 2**

When the Average Time is set to three times the Gate Time:

<table>
<thead>
<tr>
<th>Average Time/Gate Time</th>
<th>3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intensity (Area $I_A$)</td>
<td>$I_{A1} + I_{A2} + I_{A3}$</td>
</tr>
</tbody>
</table>
where:

- Total Decay Time: 40 ms
- Delay Time: 10 ms
- Average Time: 60 ms
- Gate Time: 20 ms
How many flashes should I perform?—Scan

The Number of Flashes option is set in the Bio-/Chemi-luminescence/Phosphorescence Options dialog. To access the dialog, select the Options button from the Cary page.

Note
The Options button is only visible on the Cary page if you have selected Bio-/Chemi-luminescence or Phosphorescence as the Data mode.

If you are measuring phosphorescence samples that have a relatively long decay time (that is, a number of seconds), you can setup the Cary Eclipse to pulse the lamp multiple times, as this will increase the intensity of the phosphorescent signal (refer to Example 1, below). For short lived phosphorescent samples, multiple flashes will not impact on the intensity of the signal, and therefore only one flash is required (refer to Example 2, below).

Example 1
The above example shows that the sample does not decay before the instrument lamp flashes again. Therefore setting multiple flashes will result in an increase in the intensity of the signal.

Example 2
The above example shows that the sample decays in less time than the time it takes the instrument to flash the lamp again. Therefore, setting more than 1 flash, in the No. of Flashes field will not increase the intensity of the signal.
Options—Scan

Display Options

Overlay traces
Select this option to superimpose the collected data of each sample in the run in one graph box. If you select this option, a graph box is created at the start of the collect in the Graphics area and each sample’s data is overlaid in that box as the data is collected. Traces will always be overlaid in the same graph, regardless of what accessory is used. Clear this option to display all of the traces in individual graphs.

Y minimum
Enter the minimum Y value to be displayed on the graph axis during the data collection.

Y maximum
Enter the maximum Y value to be displayed on the graph axis during the data collection.

Note
This option is not available if you have selected Cycle mode or if you have selected the Wellplate on the Accessory page. Display Options for the Wellplate are specified on the Wellplate Accessory page.

S/N or CAT mode
Select from one of the following modes.

S/N mode
Select S/N mode to enable signal to noise mode and specify the minimum signal to noise performance acceptable during the measurement.

The Cary Eclipse instrument will control the Averaging Time by waiting at each data point and averaging the signal until the value set in the S/N field is obtained. This parameter is useful if you have areas of low transmission in your scan and wish to have the same level of 'noise' across the whole scan. Using signal-to-noise mode means that you don't have to lengthen the data collection time by setting a long Averaging Time for the whole scan range. The instrument will average for a short time in the areas where there is high light throughput and longer in the areas where the light throughput is less.

If the S/N cannot be obtained in the set S/N Time out, then the instrument will keep collecting data, but the Cary Eclipse will display an error message. (A # will appear next to the intensity value.) The S/N Time out should therefore be set to a large number (for example, 10 s) to ensure that the S/N is met.

S/N
This field sets the signal-to-noise ratio that the Cary needs to achieve before moving to the next data collection point.

Time out
This sets the time period for which you want the Cary to average the signal in an attempt to reach
the ratio set in the Acceptable S/N entry field. If the specified ratio is not met in this time period then the instrument will record an error message and continue onto the next data point.

**CAT mode**
Select CAT (Computer Averaging of Transients) mode to average multiple scans. This enables better signal to noise ratios to be obtained. If this mode is selected, a series of scans (set in the **No. of scans** field) will be collected and a trace of these scans will be displayed. At the end of each scan, the averaging algorithm is applied and the result is updated and stored in the **Trace Preferences** dialog, however individual scans are not stored in this dialog. The standard deviation, which is also calculated for each point in the resultant trace, is displayed in the Trace Preferences Audit Log.

You can use CAT mode in conjunction with multiple samples. When it is used with the plate reader or multicell holder, the number of scans are collected at each cell/well before moving on to the next scan.

**No. of scans**
Enter the number of scans to be used for the CAT.

**Cycle mode**
Selecting **Cycle mode** enables you to collect scans as a function of time to determine whether the spectrum is changing. (By contrast, the Kinetics application enables you to determine whether a single wavelength is changing over time.) When you select **Cycle mode** the following options are enabled:

**No. of scans**
Enter the number of scans to be performed per cell/well.

**Time**
The **Time** option enables you to set the time interval between each successive scan. If the time entered is less than the time required to complete a scan, the scans are performed continuously with no delay.

The time at which each data point is plotted is stored with the trace, so that a post-run 3-D plot of Intensity vs wavelength vs time can be generated. The time of the first data point of the trace should be displayed in the User Data Form alongside the trace name with the heading of 'Time (min)'.

**Smoothing**
Select Smoothing to apply smoothing to the ratioed intensity signal using the algorithms defined below. You can select one of the three types of smoothing that can be performed during a run. Only the smoothed trace is displayed during data collection, however the ratioed data is also saved with the experiment.

Smoothing is used to reduce variability in data or noise. If you select smoothing, there will be a time delay in displaying data as number of points equal to the filter size need to be collected before the first data point can be averaged and displayed.

**Type**

**Savitzky-Golay**
Savitzky-Golay smoothing is used to obtain good noise reduction with sharp peaks. However, Peak edges can be distorted with rapid intensity changes.

This algorithm will take the number of Filter points, apply the Savitzky and Golay smoothing algorithm and display the smoothed value at the middle X-point of the filter. The naming of the smoothed trace will be 'Sample name_SG'.

**Tell me more about Savitzky-Golay smoothing**

**Boxcar average**
Boxcar averaging can be used when the change in intensity is minimal.

This algorithm will take the number of points as defined by the Filter size, and average these, displaying the average at the middle X-point of the filter.
The naming of the smoothed trace will be 'Sample name_Box'.

Tell me more about Boxcar averaging

Moving average
Moving averaging is recommended for time scans. It provides good noise reduction by improving signal to noise levels.

This algorithm will take the number of points as defined by the Filter size, and average these, displaying the average at the middle X-point of the filter.
The naming of the smoothed trace will be 'Sample name_Mov'.

Tell me more about Moving average

Factor
Enter the Factor (also known as Filter size), which determines the number of points that will be used in the smoothing procedures. The higher the Factor (and thus the number of points used), the greater the amount of smoothing that will be performed.

Factors must be odd values. For Boxcar and Moving averages, the range is 3–101. For Savitzky-Golay smoothing, the range is 5–101.

Excitation filter
Set the excitation filter for the Scan measurement. To do this, select the down arrow to the right of the field and select an option from the list.

A range of filter settings are available:

Auto
If you select the Auto option, the filter wheel will be automatically moved to the appropriate position for the selected excitation wavelength. It is recommended that you set the filter to Auto (which is the default setting).

Open
If you select Open no filter is used.

Closed
If you select Closed, a blank filter is used to prevent light from getting through.

250–395, 335–620, 550–1110, 695–1110
If you are looking at a wavelength range that is within one of these filter ranges, you can select your filter manually. Otherwise it is recommended that you set the filter to Auto.

Emission filter
Set the emission filter for the Scan measurement. To do this, select the down arrow to the right of the field and select an option from the list.

A range of filter settings are available:

Open
If you select Open no filter is used. In order to minimize steps in the spectra, it is recommended that you set the filter to Open (which is the default setting).

Closed
If you select Closed, a blank filter is used to prevent light from getting through.
If you are looking at a wavelength range that is within one of these filter ranges, you can select your filter manually. Otherwise it is recommended that you set the filter to **Auto**.

**3%T Attenuator**
Select 3%T Attenuator to decrease the fluorescence intensity and thus the amount of light going to the emission PMT. This setting is useful for highly fluorescent samples when you are using wide slits.

**Auto**
If you select the **Auto** option, the filter wheel will be automatically moved to the appropriate position for the selected emission wavelength.

**PMT Detector Voltage**
Set the EHT or sample detector PMT. To do this, select one of the following:

**Low**
Sets the PMT Detector voltage to a pre-defined value of 400 v. Typically you would select this setting if you are reading highly emitting samples.

**Medium**
Sets the PMT Detector voltage to a pre-defined value of 600 v. Typically you would select this setting if you are reading moderately emitting samples.

**High**
Sets the PMT Detector voltage to a pre-defined value of 800 v. Typically you would select this setting if you are reading low emitting samples.

**Manual**
Enables you to enter any PMT voltage in the adjacent Volts field.

**Volts**
Enter the PMT voltage in the adjacent field. Note that this option is only available if you have selected Manual as the PMT Detector voltage.

**Tip**
Increasing or decreasing the PMT Detector Voltage by 10% will double or halve the fluorescence intensity observed.

**Corrected spectra**
Select this option to enable the Emission or Excitation spectrum to be corrected.
If you select this option you will be prompted when you close the Setup dialog to select a spectra to collect. To do this, from the **Commands** menu point to **Correction Curves**, and then choose from the options of Excitation (200…600 nm), Emission (200…600 nm) or Emission (>600 nm). The instrument will store the relevant Intensity/Wavelength data in the Varian/Cary Eclipse WinFLR/Correction Curves directory on the drive on which the Cary Eclipse software is installed. The file will be saved as either Excorr.csv, Emcorr_190 to 600.csv or Emcorr_>600.csv, depending upon the Correction Curve you have selected.

The Corrected Spectra option is not available if you have selected **Synchronous** as the **Scan mode** on the **Cary** page.

**Status Display**
Select this check box to display the **Status Display** window, where you can view the status of various parameters during the data collection.

See also:
- **Scan Application**
- **Setup**
- **Accessories**
- **Trace Preferences**
- **Status Display**
About Smoothing—Scan

Smoothing

Savitzky-Golay

Savitzky-Golay smoothing is used to obtain good noise reduction with sharp peaks. The equation constants are specific to Filter Size. The constants for each filter size are published in the Savitzky-Golay paper (A. Savitzky, M.J.E. Golay, Anal. Chem., 36 (1964) 1627). The equation also requires that Filter Sizes entered must be odd values, between 5 and 101.

Example

<table>
<thead>
<tr>
<th>Data</th>
<th>(x1, y1), (x2, y2), (x3, y3) … (xn,yn)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Filter size (Factor)</td>
<td>5</td>
</tr>
</tbody>
</table>

Result

The first smoothed data point will be:

\[(x3, \frac{Y1*a1+Y2*a2+Y3*a3+Y4*a4+y5*a5}{b}),\]

the next:

\[(x4, \frac{y2*a1+y3*a2+y4*a3+y5*a4+y6*a5}{b}) \ldots \]

\[(xn-2, \frac{sum(yn-4*a1+yn-3*a2+yn-2*a3+yn-1*a4+yn*a5)}{b}),\]

where \(a1..a5, b\) are constants defined by Savitzky and Golay.

Each smoothed point is calculated using the previous and next data points. For this reason, calculations cannot be performed on the very first and very last data points in a data set. Hence, smoothed traces will lack the first and last X values of the original trace.

Boxcar average

Boxcar averaging can be used when the change in intensity is minimal.
Example

Data \((x_1, y_1), (x_2, y_2), (x_3, y_3) \ldots (x_n, y_n)\)

Filter size (Factor) 5

Result

The first smoothed data point will be:
\((x_3, \text{sum}(Y_1:y_5)/5),\)

the next:
\((x_8, \text{sum}(y_6:y_10)/5) \ldots (x_{n-2}, \text{sum}(y_{n-4}:y_{n})/5).\)

If there are not enough points at the end of the data set to complete the filter size, then these points will be ignored, hence, the gap at the end may be larger than the gap in the beginning.

Moving average

Moving averaging is recommended for time scans. It provides good noise reduction by improving signal to noise levels.
Example

Data 
Filter size (Factor) 5

Result

The first smoothed data point will be:

(x3, sum(Y1:y5)/5),

the next:

(x4, sum(y2:y6)/5) … (xn-2, sum(yn-4:yn)/5).

Accessories—Scan

Use the Accessories page to enable you to set up the accessory parameters associated with the data collection.

To access the Accessories page, select the Accessories command from the Setup menu. You can also access the page from the Setup dialog, by selecting the Accessories tab. To access the Setup dialog, select the Setup button from the Scan Application window or the Setup command from the Setup menu.

The Accessories page will vary, depending upon what accessory button you have selected from the following:

- Multicell/Temperature
- Wellplate
- Polarizer

See also:
Cary
Scan Application
Multicell/Temperature accessories—Scan

The Multicell/Temperature accessory page is displayed when you select the Multicell/Temperature tab on the Accessories page.

The page comprises the following:

**Multicell Holder**

Select this option to use the Multicell Holder and cells with the Cary Eclipse.

**Note**

By selecting this accessory you will prohibit the use of some combinations of other accessories that are also controlled by the Accessory Controller Board.

**Multi-zero**

If you select Multi-zero, a Multi-zero will be performed when the Zero button is selected in the Scan Application window. A reading will be taken at the set Averaging Time at the specified wavelengths of one or both monochromators and at every selected cell position. These readings will be offset to zero and subsequent readings will have the corresponding offset value subtracted from each cell.

**Reset**

Select Reset to drive the Multicell Holder to cell position 1, which is its fully retracted or zero position.

**Go to cell...**

Select Go to cell to open the Multi Cell Changer dialog. You can use this dialog to manually move the Multicell Holder accessory to a new cell. To do this, simply select the corresponding button of the cell you require in the Multi Cell Changer dialog. The Multicell Holder will move the selected cell to the measurement position in the sample compartment.

The Multi Cell Changer dialog is ‘modeless’ which means that it can remain open even once you have cleared the Setup dialog. This enables you to use the Rapid Read command to perform a reading at the cell specified in the Cell Changer.

If you move to a new cell after you have performed a Multi-zero scan, the Cary Eclipse will display the intensity reading corrected by the determined zero-offset amount for that cell.

**Note**

You can use this option to check the intensity of a cell or perform a scan at a particular cell.

**Temperature Control**

Select Temperature Control to specify the temperature parameters for your analysis.

**Note**

The Temperature Control option does not control temperature accessories that are not listed in the Device field. For example, if you have selected the Temperature Control option and set the Device to None because you have a different temperature control accessory, the Cary Eclipse software will not control that accessory. However, it will monitor the temperature and prompt the software to start when the accessory reaches the temperature listed in the Temperature field.

**Device**

Select the device that will be used to control the water temperature.

The options available will depend upon whether or not you have selected the Multicell holder option.

If you have selected the Multicell holder, the following Device options will be available for the following Device options will be available...
Temperature Display

Select this option to view Temperature Display information in the Status Display window. The Temperature Display option is only available if you have selected the Temperature Control option.

**Block/ PCB 150**
Select this option to view the temperature of the Block or the PCB 150 in the Status Display window. This option is not available if you have selected None as the Device option.

Note that the PCB 150 option is only displayed if you have selected the PCB 150 Water Bath as the Device option without the Multicell holder.

**Probes**
Select one or more of these options to view the temperature of the Probes in the Status Display window.
Status Display

Select this check box to display the Status Display window. This window allows you to view the status of various parameters during the data collection.

See also:
- Multi Cell Changer
- PCB 150
- Cary page
- Scan Application
- Status Display window

Multi Cell Changer—Scan

Use the Multi Cell Changer dialog to manually move the Multicell Holder accessory to a new cell. To access the Multi Cell Changer dialog, select the Go to cell button from the Multicell/Temperature Accessories page.

The dialog comprises the following:

**Multicell changer**

To manually move the Multicell Holder accessory to a new cell, simply select the corresponding button of the cell you require in the Multi Cell Changer dialog. The Multicell Holder will move the selected cell to the measurement position in the sample compartment.

The Multi Cell Changer dialog is ‘modeless’ which means that it can remain open even once you have cleared the Setup dialog. This enables you to use the Rapid Read command to perform a reading at the cell specified in the Cell Changer.

If you move to a new cell after you have performed a Multi-zero scan, the Cary Eclipse will display the intensity reading corrected by the determined zero-offset amount for that cell.

**Note**

You can use this option to check the intensity of a cell or perform a scan at a particular cell.

See also:
- Multicell/Temperature Accessories
- Rapid Read

Wellplate accessory—Scan

The Wellplate accessory page is displayed when you select the Wellplate tab on the Accessories page.

The page comprises the following:

**Wellplate reader**

Select this option to use the wellplate reader with the Cary Eclipse.

**Q.** Why is the wellplate reader option unavailable?

**A.** The multicell holder, temperature control and/or polarization accessory has been selected. As you cannot place any of these accessories and the wellplate reader in the Cary Eclipse sample compartment at the same time, you must clear other accessory selections before selecting the wellplate reader.

**Plate format**

Select the type of wellplate to be used. The default options are 6 wells, 12 wells, 24 wells, 48 wells, 96 wells, 192 wells or 384 wells.

The graphical representation of the wellplate will change according to the number of wells chosen.
**Auto-zero**
Select this option to perform an auto-zero when the **Zero** button is selected from the Scan Application window. A zero reading will be taken at the selected well position using the current method setup. To select the well position to be zeroed, use the **Select Well** option.

These readings will be offset to zero and subsequent readings will have this single value subtracted from each well in the sequence. The reading is taken only once, stored, and subtracted from the ratioed intensity reading.

**Select well**
Choose **Select well** to open the **Go To well** dialog where you can select the well position to be zeroed. This option is only available if you have selected **Auto-zero**.

**Well**
Displays the well position of the well selected for auto-zeroing. This field is not editable.

**Display options**
Select this option to open the **Display Options** dialog where you can set up the display parameters associated with the wellplate reader.

**Sample names**
Select **Sample Names** to open the **wellplate Sample List** dialog where you can enter or modify the sample names. Highlight one or more wells on the wellplate reader diagram to view and change their sample information.

**Go to well**
Select this option to open the **Go To Well** dialog.

**Well position**
Displays the well position that the mouse cursor is pointing to. This field is not editable.

**Reset**
Select **Reset** to move the wellplate reader to the first well position defined by the read sequence.

**Park**
Select **Park** to move the wellplate reader to the Park position. The wellplate reader accessory is moved clear of the sample compartment to enable you to easily access all wells.

**Wellplate reader diagram**
The wellplate reader diagram represents the number of wells present for the chosen wellplate configuration.

The labels for the rows will be 1 .. 12 and for the columns A .. H (for a 96 wellplate). Similar labels will occur for different wellplates according. The sequence for numbering the read order will be from A1 to A12, B12 - B1, C1 - C12...etc for a 96 wellplate. A similar sequence will be for other plates.

To select or clear a single well, simply click or right-click on it.

To select or clear a block of wells, hold down the left mouse and drag around the block required. All wells within the area will be selected or cleared.

**Status Display**
Select this check box to display the **Status Display** window. This window allows you to view the status of various parameters during the data collection.

**Display Options—Scan**
Use the **Display Options** page to set up the display parameters associated with the Wellplate Reader.

The **Display Options** page is accessible from the **Wellplate Accessories** page by selecting the **Display Options** button.
The page comprises the following:

**Display**

**Individual graphs** Select this option to display the collected data of each sample in individual graph boxes, rather than overlaid on each other in one box.

At the start of a scan, a separate box is created for each sample. Similarly, the collected data for each sample is displayed in a separate graph box.

**Overlay all traces** Select this option to superimpose the collected data of each sample in the run in one graph box.

One box is created at the start of the collect in the Graphics area and each sample’s data is overlaid in that box as the data is collected.

**User defined** Select this option if you wish to specify the exact number of traces to be included on one graph.

**Number of traces per graph**
Enter the number of traces to be displayed per graph.

**Overlay rows** Select Overlay rows to view overlaid graphs of the wells you have selected. Wells in the same row of the wellplate reader will be overlaid. If you had selected wells as shown in the adjacent diagram, a total of seven graphs would be displayed (as seven rows contain selected wells). The first graph would contain overlaid scans of well A1 and A6, the second graph would contain a scan of well B8, etc.

**Overlay columns** Select Overlay columns to view overlaid graphs of the wells you have selected. Wells in the same column of the wellplate reader will be overlaid. If you had selected wells as shown in the adjacent diagram, a total of 11 graphs would be displayed (as 11 rows contain selected wells). The first graph would contain overlaid scans of well A1 and D1, the second graph would contain a scan of well H2, etc.

See also: [Wellplate Accessories](#)

**Wellplate Sample List—Scan**

Use the Wellplate Sample List dialog to enter or modify the sample names of each well in the Wellplate reader.

The Wellplate Sample List is accessible from the Wellplate Accessories page by selecting the Sample Names button.

The page comprises the following:

**No. of samples** Displays the total number of samples that have been selected from the Wellplate Reader diagram on the Wellplate Accessories page. This field is not editable.

**Sample table** The sample table lists the Sample Names and Well numbers of the selected samples.

**Sample Name**
Samples are named numerically, depending upon where they are located on the Wellplate Reader diagram. The sequence for numbering the Sample Name order is to read from A1 to A12, B1 to
Go To Well—Scan

You can use the Go To Well dialog to manually move the Wellplate Reader accessory to a new well. This enables you to check the intensity of a well or perform a read at a particular well.

The Go To Well page is accessible from the Wellplate Accessories page by selecting the Select well or Go To Well button.

The Go To Well dialog is ‘modeless’ which means that it can remain open even once you have cleared the Setup dialog. This enables you to use the Rapid Read command to perform a reading at the selected well.

If you move to a new well after you have performed a Multi-zero, the Cary Eclipse will display the intensity reading corrected by the determined zero-offset amount for that well.

The page comprises the following:

**Wellplate reader diagram** The Wellplate Reader diagram represents the number of wells present for the chosen wellplate configuration.
The Polarizer accessory page is displayed when you select the Polarizer tab on the Accessories page. The page comprises the following:

**Polarizer**

Select this option to use the Polarizer with the Cary Eclipse.

**Status Display**

The labels for the rows will be 1..12 and for the columns A..H (for a 96 wellplate). Similar labels will occur for different wellplates according. The sequence for numbering the read order will be from A1–A12, B12–B1, C1–C12, etc for a 96 wellplate. A similar sequence will be for other plates.

To move the Wellplate Reader accessory to a new well, simply click on the corresponding well on this diagram. The Wellplate Reader will move the selected well to the measurement position in the sample compartment.

To select or clear a single well, simply click or right-click on it.

To select or clear a block of wells, hold down the cursor and drag around the block required. All wells within the area will be selected or cleared.

<table>
<thead>
<tr>
<th>X, Y, Well Position</th>
<th>Displays the Well Position that the mouse cursor is pointing to. This field is not editable.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reset</td>
<td>Select Reset to move the Wellplate Reader to the first well position, which is well A1.</td>
</tr>
<tr>
<td>Park</td>
<td>Select Park to move the Wellplate Reader to the Park position. When the accessory is in the park position you have full access to the Wellplate Reader platform. This is useful for inserting or removing wellplates from the accessory.</td>
</tr>
</tbody>
</table>

**Q. Why is the Polarization option unavailable?**

**A.** The Wellplate accessory has been selected. As you cannot place both the Wellplate accessory and the Polarization accessory in the Cary Eclipse sample compartment at the same time, you can only select one of these accessories.

**Measurement**

Select from the following measurement types: Polarization, Anisotropy or Other.

**G Factor**

The G Factor is a value that reflects the polarization bias of the emission monochromator. It is dependant upon the emission wavelength. If you do not select the G Factor option, then a G-factor value must be collected. If you do select the G Factor option then the value entered is used at the emission wavelength in subsequent polarization or anisotropy calculations.

The G Factor option is not available if you select Other as the Measurement type.

**Determining the G Factor**

A check to see whether there is a valid G Factor that corresponds to the emission wavelength is performed when you select OK to exit the Setup dialog. If the G Factor is not valid, the following prompt is displayed:

‘The current G Factor is not valid at the emission wavelength. Press the G Factor button to determine a new grating factor.’

Select OK to clear this dialog and select the G Factor button from the Scan Application window.

**Scan Home Page**
Select this check box to display the Status Display window. Use this window to view the status of various parameters during the data collection.

See also:
PCB 150 Accessories page

Reports—Scan

Use the Reports page to set up the report parameters associated with data collection.

From the Setup menu, select Reports to access the Reports page. You can also access the page from the Setup dialog by selecting the Reports tab. To access the Setup dialog, select the Setup button from the Scan Application window.

The page comprises the following:

Operator

Name
Enter the instrument operator name in this text entry field. Use the arrow to the right of this field to view and select one of the last ten entries in this field.

Note
In GLP mode the system will automatically enter the operator’s name and this field will not be editable.

Comment
Enter operator details and comments in this text entry field.

Options

This group enables you to select the information to include in your report.

Auto Print
Select Auto Print to print a report automatically during the data collection. If Auto Print is selected, then the system will send the report information to the specified printer (s) as well as the Report area. However if Auto Print is not selected, the report will only be sent to the Report area and can be viewed by selecting Report in the View menu.

To automatically obtain a copy of the graphics, you will need to select both the Auto Print and Graph options (see below). In this case, all graphical information displayed on screen will be printed with the report. Therefore it is advisable to arrange the graphical information as you require on screen, prior to printing.

For example, if you require a single graph print out, then the Single/Multi Graph option in the Graph menu should be changed to the correct setting.

Parameters
Select this option to include the method setup parameters within your report.

User data form
Select this option to include in the report any information entered in the User Data Form.

Graph
Select Graph to include any graphs or graphic information associated with your report. If you require a printout of the displayed graphics only, you must ensure that only this option is selected. Press the Clear Report button to clear any information in the Report area before you print otherwise this will be included in your printout.

% Page Height
Enter the size at which you want the graph to be printed, as a percentage of total page height. For example, if you want graphical information to take up half of the page, enter 50% in this field.

Company Logo
Check this option to include your company logo in the report.
A company logo will only be included if you have added one on the Company page of the System Information Application.

**Hot Tip**
If you have not included a Company logo, select the Header/Footer button to open the System Information Application and use the Add Logo option to add a logo.

**Header/ Footer**
Select the Header/Footer button to open the System Information application where you can enter the Header and Footer information to be included in your report. You can also enter any additional comments, and add a logo to your report. Once you have made all your changes, select OK to exit the System Information application and return to the Reports page of the Scan application.

**Focused trace**
Select this option to apply the Report selections to the focused trace only.

**All traces**
Select this option to apply the Report selections to all visible traces.

**Peaks**

Use the Peak options to select how the traces will be labeled in your report.

**Maximum Peaks**
Select **Maximum** to calculate the maximum peak in the trace/s and include the peaks in your report.

**All peaks**
Select **All** to calculate all peaks above the peak threshold in the trace/s and include the peaks in your report.

**Label peaks**
Select this if you wish the peaks on the graph to be labeled with the co-ordinates selected in the Peak Labels dialog (X and Y, X only, Y only).

**Peak type**
This parameter selects the type of peaks that will be included in your report. Select from the following options:

**Peaks**
The Cary Eclipse will report maximum points only. A peak is defined by a maximum in the first derivative.

**Valleys**
The Cary Eclipse will report minimum points only. A valley is defined by a minimum in the first derivative.

**Peaks Valleys**
The Cary Eclipse will report both maximum and minimum points. It is mainly used in odd number derivatives where the X intercept defines a peak.

**Zero Cross**
The Cary Eclipse will report the points at which the trace crosses over the zero ordinate line. A zero-crossing peak type is where the continuum crosses the X axis, that is at Y=0. This choice is useful for odd derivative scans calculated in the Maths window. For example, the zero crossing points of a first derivative scan correspond to the peaks and valleys in the trace.

**Threshold**
Enter the Threshold value, which is the value that defines what peaks/valleys will be displayed in the report. The difference in intensity between a valley, the highest peak and the adjacent valley is measured and, if this difference is greater than the threshold it is reported on the trace.

**Label options**
Select this option to open the Label Options dialog where you can select how the labels will be displayed in the report.

**X-Y Pairs Table**
Select this if you want to print a table showing the X-Y pairs of the collected data in your report.

**Data Interval**

*Actual*
Select **Actual** to report the X-Y pairs table for the trace at the actual Data Interval at which the data was collected.

*Interpolated*
Select **Interpolated** to report the X-Y pairs table for the trace at an interpolated data interval. Enter this data interval in the adjacent field. If no data has been collected at the interval you enter, the Cary Eclipse software will interpolate the data to produce an X-Y result.

**Status Display**

Select this check box to display the **Status Display** window, where you can view the status of your instrument during the data collection.

**Hot Tip**
You can copy a picture, spreadsheet file or document from another application (for example, Microsoft Word or Microsoft Excel) into the report area of the Lifetimes application. To edit these objects, simply double click on them to open the associated application in which they were created.

See also:
- Scan Application
- View menu
- User Data Form
- Single/Multi Graph
- Peak Labels
- Label Options dialog
- Maths window

**Report Toolbar—Scan**

Use the Report toolbar buttons to gain quick access to many of the Edit menu options. You can also use the toolbar to cut, copy and paste information and to change the look of your report.

To display or hide the toolbar, point to the Toolbars option in the View menu and select Reports. To display or hide the toolbar's text labels, point to the Toolbar option in the View menu and select Text Labels.

**Hot Tip**
To see the name of a button, point to it with the mouse.

The following commands can be accessed from the Reports Toolbar:

<table>
<thead>
<tr>
<th>Button</th>
<th>Menu command</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image" alt="Edit Report" /></td>
<td><strong>Edit Report</strong></td>
<td>Click this button to edit the report. This command is also accessible by pressing CTRL+R.</td>
</tr>
<tr>
<td><img src="image" alt="Undo" /></td>
<td><strong>Undo</strong></td>
<td>If available, this undoes the last action. This command is also accessible by pressing CTRL+Z.</td>
</tr>
<tr>
<td><img src="image" alt="Cut" /></td>
<td><strong>Cut</strong></td>
<td>Click this button to remove the highlighted text and place it on the Windows Clipboard. This command is also accessible by pressing CTRL+X.</td>
</tr>
<tr>
<td><img src="image" alt="Copy" /></td>
<td><strong>Copy</strong></td>
<td>Click this button to copy the highlighted text onto the Windows Clipboard. This command is also accessible by pressing CTRL+C.</td>
</tr>
</tbody>
</table>
Create Report—Scan

Select the Create Report button to access Report options.

The Create Report button is accessible from the Scan Application window. When you select this option, the Setup dialog will appear, containing the following page:

- Reports

See also: Scan Application window

Setup

Clear Report—Scan

Select Clear Report to remove the contents of the Report area. If you have saved the report, selecting this option does not delete the contents of the report, the information is simply removed from the Report Area.

You can select the Clear Report button from the Scan Application window or the Clear Report command from Commands menu.

See also: Scan Application Commands menu

Label Options—Scan

Use the Label Options dialog to select how labels will be displayed in the report. To access the Label Options dialog, select Label Options from the Reports page of the Setup dialog.

The dialog comprises the following:

- X and Y Labels
- Y Label
- X Label

Select this if you wish the peaks on the graph to be labeled with both the X and Y co-ordinates.

Select this if you wish the peaks on the graph to be labeled with only the Y co-ordinates.

Select this if you wish the peaks on the graph to be labeled with only the X co-ordinates.
Peak Labels—Scan

Use the **Peak Labels** dialog to set up the peak information that will be included in your report.

From the **Graph** menu, select **Peak Labels** to access the dialog. Note that this option is only available when a trace is visible.

**Hot Tip**
The selections you make in the **Peak Labels** dialog are applied to the selected graph(s). To select multiple graphs, hold down the CTRL key and select the graphs. You can also use the SHIFT key to select a range of graphs. When the shift key is held down, all graphs in between the first and last graph chosen are selected.

The dialog comprises the following:

**Peak Style**  
This parameter selects the type of peaks that will be reported by the Cary Eclipse software. Select from the following options:

**Note**  
When determining **Peaks, Valleys** or **Peaks Valleys** the Cary Eclipse software nominates the highest data point of a peak as the peak position and the lowest data point of the valley as the valley point. It does not interpolate between points. Thus, if your data points are relatively far apart (large Data Interval), the reported peak or valley may not be entirely accurate.

However, when you use the zero crossing points to calculate the peak and valley positions, the data is interpolated and a more accurate peak position may be obtained.

- **Peaks**  
The Cary Eclipse will report maximum points only.

- **Valleys**  
The Cary Eclipse will report minimum points only.

- **Peaks Valleys**  
The Cary Eclipse will report both maximum and minimum points.

- **Zero Cross**  
The Cary Eclipse will report the points at which the trace crosses over the zero ordinate line. A zero-crossing peak type is where the continuum crosses the X axis, that is at Y=0. This choice is useful for odd derivative scans calculated in the **Maths** window. For example, the zero crossing points of a first derivative scan correspond to the peaks and valleys in the trace.

**Hot Tip**  
Taking a first derivative of a noisy scan will reduce the noise and assist in locating the peaks more accurately.

**Threshold**  
Enter the **Threshold** value, which is the value that defines what peaks/valleys will be displayed. The difference in intensity between a valley, the highest peak and the adjacent valley is measured.
and, if this difference is greater than the threshold it is reported on the trace. To include Peaks/Valleys in your reports, ensure that you have the Label Peaks option selected on the Reports page of the Setup dialog.

See also:

Peak Picking

Maximum

Select Maximum to calculate the maximum peak in the trace/s. Select one of the Label options (X and Y labels, Y label, X label) to display the maximum peak on the trace. To include the maximum peak in your report, ensure that you have the Maximum Peaks option selected on the Reports page of the Setup dialog.

All

Select All to calculate all peaks above the peak threshold in the trace/s. Select one of the Label options (X and Y labels, Y label, X label) to display the maximum peak on the trace. To include the maximum peak in your report, ensure that you have the All Peaks option selected on the Reports page of the Setup dialog.

X and Y Labels

Select this if you wish the peaks on the graph to be labeled with both the X and Y co-ordinates.

Y Label

Select this if you wish the peaks on the graph to be labeled with only the Y co-ordinates.

X Label

Select this if you wish the peaks on the graph to be labeled with only the X co-ordinates.

No Labels

When selected, no peaks will be labeled with co-ordinates.

Position

Use the slider bar underneath the graph to select the position of the label on the graph. This option is not available if you have selected No Labels.

Y Decimals

Use this option to select the number of decimal places to appear in your Y co-ordinates. This option is not available if you have selected No Labels.

Font

Select the Font button to access the Font dialog where you can set the size and style of the text that will appear on the labels. This option is not available if you have selected No Labels.

See also:

Maths window
Maths How To
Reports

How to use advanced reporting options—Scan

This outlines ways to fully customize your reports. Options available include:

- Including header and footer information
- Including a logo
- Editing the text in the report
- Viewing the report on screen before printing
- Cutting and pasting to a word processor

Including header and footer information

You can have text appearing at the top and bottom of every page in your report. To specify what the text should be, do the following:

Step 1

On the Windows Taskbar, click the Windows Start button, point to Programs, Cary Eclipse and then select System Information. The System Information application appears with the Company page displayed.

Step 2

Enter the header and footer information in the Header and Footer fields.
Step 3

Click **OK** to implement your changes.

**Including a logo**

You can include a logo on the first page of your report. To specify what the logo should be, do the following:

Step 1

On the Windows Taskbar, click the Windows **Start** button, point to **Programs, Cary Eclipse** and then select **System Information**. The System Information application appears with the Company page displayed.

Step 2

Enter a logo using the **Add Logo** button. This logo will always be included on the front page of reports.

Step 3

Click **OK** to implement your changes.

**Q.** Why isn't the logo I selected included in my report?

**A.** If you are running the Cary Eclipse software under Windows NT you need to be logged on as an Administrator in order to specify the logo in the System Information application.

**Editing the text in the report**

You can edit any of the text in the Report area. Just select **Edit Report** from the **Edit** menu. You can then add, delete and change text in the Report area.

**Q.** The Edit Report option is not available, why not?

**A.** You do not have reporting editing privileges set in the GLP Administration application. The Administrator of the system will have to change your privileges before you can edit reports.

**Viewing the report on the screen before printing**

You can view a report before printing it. To do this, from the **File** menu select **Print Preview**. You can then ensure that the report format is correct before you print it.

**Hot Tip**

Use the arrow keys at the bottom of the Print Preview screen to change pages.

**Cutting and pasting to a word processor**

To have complete control over how your report looks you may wish to copy it to a word processor program, such as Microsoft Word. To do this:

Step 1

Open a report.

Step 2

From the **Edit** menu, choose **Edit Report**.
Step 3

Click in the Report area and highlight the text you want to export to your word processor.

Step 4

Right-click on the Report area and select **Copy** from the menu.

Step 5

Start up your word processor and use the Paste function (usually under the **Edit** menu) to place the text into a document.

See also:
- How to add a label to a graph
- How to add a picture to a graph
- How to alter the look of your graphs
- How to display a trace that is not appearing

## Auto store—Scan

Use the **Auto store** page to specify if and when data will be stored, and in what format. You can also use this page to set up the system to automatically send an email at the end of a read.

To access the **Auto store** page, select the **Auto store** command from the **Setup** menu. You can also access the page from the **Setup** dialog, by selecting the **Auto store** tab. To access the **Setup** dialog, select the **Setup** button from the Scan Application window or the **Setup** command from the **Setup** menu.

The page comprises the following:

### Storage

- **Off**

  Select this if you do not wish to be automatically prompted to save collected data.

  **Note**
  
  If you wish to store your unsaved data manually, choose Save Data As… from the File Menu to save your data after the run is complete. You can save your data as a Batch or Data file.

- **On; prompt at start**

  Select this to display the Windows **Save as** dialog at the start of the collect where you can enter the Batch filename for your data.

- **On; prompt at end**

  Select this to display the Windows **Save as** dialog at the end of a collect where you can enter the Batch filename for your data.

### Auto convert

Use the Auto convert options to convert your data into an ASCII file format that can be opened in spreadsheet programs such as Microsoft Excel.

- **None**

  Select **None** if you do not need to generate an ASCII file.

- **ASCII (csv)**

  Select this option to save the data as a comma delimited ASCII Spreadsheet file (*.CSV) file. Only the currently collected data is converted.

  ASCII files are stored in the same folder as the data file from which they were generated and
Email

Select the Email check box and enter the Email details to send an email to a person once the read is completed. This useful facility can be used to alert you to the completion of the read, particularly if you are working away from the laboratory where the Cary Eclipse instrument is. You can also attach the collected data to the email.

**Note**

If you choose to auto-convert the data to an ASCII file but you have set the Storage option to Off, then the ASCII file will be saved with the following format:

DATE TIME. CSV

The date and time will appear in the same form that is set in the Windows Regional Settings Properties dialog that is accessed from the Windows Control Panel.

For example, 22_4_97 1;32;08.CSV

**Tell me more about ASCII Spreadsheet Files**

**Email**

Select the Email check box and enter the Email details to send an email to a person once the read is completed. This useful facility can be used to alert you to the completion of the read, particularly if you are working away from the laboratory where the Cary Eclipse instrument is. You can also attach the collected data to the email.

**Note**

You must set up the email Host and Port options on the Email options page of the System Information application in order for the email facility to work correctly.

**To** Enter the recipient's email address. You can enter more than one recipient in this field.

**From** Enter your email address (or the email address associated with the computer on which the Cary Eclipse software is installed).

**Subject** Enter the subject of your email (or leave this field blank if you do not wish to include a subject).

**Cc** Enter the email address of any recipient you want to copy the email to.

**Message** Enter your message.

**Attached…** Use the Attached button include a file attachment in your email. To do this, select Attached and browse your computer to select a file, using the Windows® Open dialog that appears.

**Attach current data** Select this option to attach the data that has been collected during the run to the email.

**Status display**

Select this check box to display the Status display window, where you can view the status of your instrument during the data collection.

**How to email your results—Scan**
You may wish to send an email to a person once the read is completed. This useful facility can be used to alert you to the completion of the read, particularly if you are working away from the laboratory where the Cary Eclipse instrument is.

To do this:

Step 1

Ensure that you have set up the Host and Port options on the Email options page of the System Information application in order for the email facility to work correctly.

Step 2

From the Setup menu, select Auto Store. The Auto Store page is displayed.

Step 3

Select the Email check box to enable the email options.

Step 4

In the To field, enter the recipient's email address (for example, rchandle@varianinc.com). To enter multiple recipients, simply type a comma ',' between each email address.

Step 5

In the From field, enter the email address of the computer on which the Cary Eclipse software is installed. This may be your email address, or a communal email address, if the Cary Eclipse software is installed on a computer that has multiple users.

Step 6

Enter information in the Subject and Message fields as required.

Step 7

Select Attach current data. The data that has been collected during the run will be attached to the email.

**Hot Tip**

You can attach other documents to your email. To do this, select the Attached button and browse your computer to select a file, using the Open dialog that is displayed.

Step 8

Select OK to clear the Auto Store dialog. Your email will be sent once the run has been completed.

See also:

Auto Store

Performing scans—Scan

- Starting a scan
- Sample Name dialog
Information about starting a scan—Scan

The sequence that occurs when you select **Start** to commence the scan depends upon your storage and accessory selections.

### Storage selections

<table>
<thead>
<tr>
<th>If you have selected … as the Storage option on the Auto Store page</th>
<th>The following occurs</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>On, prompt at start</strong></td>
<td>The Windows <em>Save As</em> dialog will appear as soon as you select <strong>Start</strong>. You can save the current method, collected data, report and graph template as a Batch file.</td>
</tr>
<tr>
<td><strong>On, prompt at end</strong></td>
<td>The Windows <em>Save As</em> dialog will appear at the end of the data collection. You can save the current method, collected data, report and graph template as a Batch file.</td>
</tr>
<tr>
<td><strong>Off</strong></td>
<td>The Windows <em>Save As</em> dialog will not appear.</td>
</tr>
</tbody>
</table>

### Options selections

<table>
<thead>
<tr>
<th>If you have selected … on the Options page</th>
<th>The following is displayed</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Cycle mode</strong></td>
<td>The <em>Sample Name</em> dialog will appear when you select <strong>Start</strong>, prompting you to load the sample and enter your sample name. Select <strong>OK</strong> to display the <em>Sync Start</em> dialog and commence the kinetics run.</td>
</tr>
</tbody>
</table>

### Accessory selections

<table>
<thead>
<tr>
<th>If you have selected … on the Accessories page</th>
<th>The following is displayed</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>No accessories</strong></td>
<td>The <em>Sample Name</em> dialog is displayed when you select <strong>Start</strong>, prompting you to load the sample and enter your sample name. Select <strong>OK</strong> to display the <em>Sync Start</em> dialog and commence the kinetics run.</td>
</tr>
</tbody>
</table>
| **Multicell Holder**                         | The *Cell Loading Guide* dialog is displayed. Enter the sample name of each activated cell then select **OK** to display the *Sync Start* dialog.  
When you select **OK** to clear the *Cell Loading Guide* dialog, the cell changer will move to cell position 4, enabling you to easily access all of the cell positions and load your samples. |
| **Wellplate Reader**                         |                            |
Sample Name—Scan

If you are using a single cell holder, the Sample Name dialog is displayed once you select the Start button to commence an analysis.

The page comprises the following:

**Sample Name**

Enter the name of the sample to be used in the scan run. Click on the arrow to the right of the field to view and select the most recently entered sample names. Once you have entered the Sample Name, select OK to commence the scan run.

If you have selected Cycle Mode on the Options page, the the Sync Start dialog will be displayed after the Sample Name dialog. In Cycle Mode, the Cary Eclipse will automatically increment the Sample Name.

See also:
- Sample Name
- Cell Loading Guide
- Sync Start

Cell Loading Guide—Scan

If you are using a Multicell holder, the Cell Loading Guide dialog appears once you click the Start button to commence an analysis.

The Cell Loading Guide dialog prompts you to insert blank samples in the Multicell Holder to collect the scans. You can also use the dialog to name the samples in each selected cell in the Multicell Holder.

The dialog comprises the following:

**Loading Guide/Sample Name entry**

Enter the names of the sample to be used in the scan run in each available field. This sample name will be used as the Name of Trace in the Trace Preferences dialog.

**Note**

The Cell Loading Guide dialog appears only if you have selected the Multicell Holder option on the Accessories page of the Setup dialog. Only cells selected in the Multicell Holder diagram on the Accessories page will be available in the Loading Guide.

In the adjacent example, cells 1—3 will be available in the Loading Guide dialog, as they have been selected in the Multicell Holder diagram.
Sync Start—Scan

Use the Sync Start dialog to synchronize the start of a reaction and the start of the a scan cycle run. The Cycle mode is selected on the Options page of the Setup dialog.

Before displaying the Sync Start dialog, the Cary Eclipse will set up all the instrument and graphing parameters so that data collection can start as soon as the countdown finishes or you select OK.

The sequence for a Sync Start is as follows:

- Set up your data collection parameters
- Select the Start button (and load the samples if necessary)
- Once the Sync Start dialog is displayed, add your active reagent
- Commence the data collection by selecting the OK button

The Sync Start dialog comprises the following:

Count Down  This field displays a two minute countdown in seconds. If you want to start your data collection before the two minute countdown ends, select the OK button. Alternatively, you can wait for the countdown to end and the data collection will start automatically.

Note  If you are using a Multicell Holder and you select OK, the system will automatically reset this accessory and immediately start the data collection.

Reset Slide  Click this button to drive the Multicell Holder to its zero point (Cell position 1).

Start/Stop instrument buttons—Scan

The following buttons may be displayed at the top of the Scan Application. The Start and Connect buttons will change depending upon the status of the instrument. For example, if the instrument is ready for a run, the Start button will be visible to indicate that the system is ready to commence a run.

Select Start to commence a scan run using the current Setup parameters. If any unsaved data exists in the Graphics area you will be prompted to save the data. If you chose not to save, any unsaved data, pictures and labels will be deleted when you start the scan.

If another application is already online with the Cary Eclipse, the Start button will change to Connect, and you will need to select Connect to bring this application online before the Start button will re-appear. (You will not be able to connect if the other application is collecting data.)

Select this to end a scan. If you have selected Storage On in the Auto Store page of the Setup dialog, all data collected is automatically stored in the file specified in the Batch name field of the Windows Save As dialog before or after the run commenced.
Pre-scan—Scan

Select Pre-scan to determine the optimum instrument parameters to be used for your sample. At the end of this collection sequence the software will report the optimum Ex and Em wavelengths and PMT voltage. Once you are satisfied with the parameters, the software will automatically update your method setup to reflect these new parameters.

To access the Pre-scan option, select the Pre-scan command from the Graph menu, or select the Pre-scan button from the Scan Application window.

The Pre-scan dialog comprises the following:

**Type of scatter**

Select the type of scatter you wish to search for from the following:

- **Rayleigh scatter**: Rayleigh scatter refers to the scatter of the excitation light. It is displayed at the peak of the excitation wavelength.
- **Raman scatter**: Raman scatter refers to scatter due to the solvent. Its peak is at a fixed energy from the excitation wavelength, and is solvent dependent. Thus, the type of solvent used must be selected from the Solvent list.
- **2nd order**: Second order scatter refers to the scatter of higher order excitation light. Its peak is a multiple of the excitation wavelength.

Go To Wavelength—Scan

Use the Go To Wavelength dialog to temporarily change the instrument to a new excitation or emission wavelength. You would mainly use this dialog if you want to quickly go to a wavelength and check the intensity reading of the sample. The lamp will flash continuously in this mode. This option is only available if the instrument is not currently collecting data.

To access the Go To Wavelength dialog, select the Go To Wavelength command from the Commands menu. You can also select F4 to open the dialog.

**Note**

This is not the wavelength used in the application method. If your method uses a wavelength that is different from the 'Go to wavelength' setting you select, then when you select Start the system will change to the wavelength selected (as set on the Cary page of the Setup dialog).

The Go to Wavelength page comprises the following:

- **Excitation Wavelength**: Enter the new wavelength in this numeric entry field. When you select Go To the instrument will change to the new value.

- **Zero Order**: Select Zero Order to drive the Cary Eclipse fluorescence spectrophotometer to 0 nm (white light).
G-Factor—Scan

Select G-Factor to calculate the G Factor. You will be prompted to move your polarizers (Em and Ex) and perform the IHV and IHH measurements. At the end of the sequence the system will calculate the subsequent polarization measurements which will be performed when you select the Start button.

The G-Factor command is only available if you have selected the Polarizer accessory on the Accessories page. Once this option is selected, you can then select the G-Factor button from the Scan Application window or the G-Factor command from Commands menu.

Rapid Result—Scan

Use the Rapid Result dialog to take a reading of a blank and sample/s, without having to change the current application method. You can use the Multi Cell Changer dialog to specify the cell you want to read. This means that between runs, while new samples are being prepared and loaded, other samples can quickly be measured. The result is printed in the Report area.

To access the Rapid Result dialog, select the Rapid Result command from the Commands menu or press F6.

Zero—Scan

You can then use this light for aligning accessories.

Emission

**Wavelength**

Enter the new wavelength in this numeric entry field. When you select Go To the instrument will change to the new value.

**Zero Order**

Select Zero Order to drive the Cary Eclipse fluorescence spectrophotometer to 0 nm (white light). You can then use this light for aligning accessories.

Go To

Click Go To once you have entered a new wavelength, to change the instrument to the entered value.

See also:

Function Keys

G-Factor—Scan

Select G-Factor to calculate the G Factor. You will be prompted to move your polarizers (Em and Ex) and perform the IHV and IHH measurements. At the end of the sequence the system will calculate the subsequent polarization measurements which will be performed when you select the Start button.

The G-Factor command is only available if you have selected the Polarizer accessory on the Accessories page. Once this option is selected, you can then select the G-Factor button from the Scan Application window or the G-Factor command from Commands menu.

Rapid Result—Scan

Use the Rapid Result dialog to take a reading of a blank and sample/s, without having to change the current application method. You can use the Multi Cell Changer dialog to specify the cell you want to read. This means that between runs, while new samples are being prepared and loaded, other samples can quickly be measured. The result is printed in the Report area.

To access the Rapid Result dialog, select the Rapid Result command from the Commands menu or press F6.

**Excitation**

**Wavelength**

Enter the wavelength at which you want to take a reading.

**Zero order**

Select this option to set the excitation monochromator to zero order (white light).

**Emission**

**Wavelength**

Enter the wavelength at which you want to take a reading.

**Zero order**

Select this option to set the excitation monochromator to zero order (white light).

**Zero**

Click Zero to change the wavelength to the entered value and measure a blank.

**Result**

Click Result to change the wavelength to the entered value and measure a sample. The X,Y results are corrected for the blank and appear in the reports area.

See also:

Multi Cell Changer
Commands menu

Zero—Scan

You can then use this light for aligning accessories.

Emission

**Wavelength**

Enter the new wavelength in this numeric entry field. When you select Go To the instrument will change to the new value.

**Zero Order**

Select Zero Order to drive the Cary Eclipse fluorescence spectrophotometer to 0 nm (white light). You can then use this light for aligning accessories.

Go To

Click Go To once you have entered a new wavelength, to change the instrument to the entered value.

See also:

Function Keys
Use the **Zero** button to zero the current intensity value. The intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before commencing a data collection if you want your data to start at 0 intensity. The Cary Eclipse will prompt you to load a blank before performing the zero.

Before you select **Zero**, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.

When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the Scan Application window.

Select the **Clear Zero** command from the **Commands** menu to remove zero correction and thus remove the subtraction effect.

To access the **Zero** option, select the **Zero** command from the **Commands** menu, or select the **Zero** button from the Scan Application window.

---

**See also:**
- *Scan Application window*
- *Commands menu*
- *Cell Loading Guide*

---

### Commands menu—Scan

Use the **Commands** menu to access major operational commands in the **Scan** application. The menu is accessible from the menu line at the top of the Scan Application window.

Alternatively you can use function key shortcuts for some of the commands. These appear next to the item in the **Commands** menu. Note that not all commands appear as both button and menu items.

---

**Hot Tip**

If you access these commands using the **Commands** menu, rather than from the application buttons, you can remove these buttons from your application window. To do this, clear the **Buttons** command in the **View** menu. This will enlarge the Graphics area, making viewing and selection of scans easier.

---

**Tell me how to remove or display the command buttons**

The following commands are accessible from the **Commands** menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Start</strong></td>
<td>Select this to start a data collection using the currently set up parameters. <em>Note</em> If the <strong>Connect</strong> command is displayed in place of the <strong>Start</strong> command, this indicates that another Cary Eclipse application is in use and has control of the Cary Eclipse instrument. To make the Scan application active so that you can perform a read, you will need to select <strong>Connect</strong>. However, you will not be able to select <strong>Connect</strong> if another application is collecting data.</td>
</tr>
<tr>
<td><strong>Stop</strong></td>
<td>Select this to stop the data collection. If you have selected <strong>Storage On</strong> in the <strong>Auto Store</strong> page of the <strong>Setup</strong> dialog, all data collected is automatically stored in the file specified in the Batch name field of the Windows <strong>Save As</strong> dialog before or after the run commenced.</td>
</tr>
<tr>
<td><strong>Clear Zero</strong></td>
<td>Select this option to clear the <strong>Zero</strong> command. When you select the <strong>Zero</strong> command (from the <strong>Commands</strong> menu or by pressing F5), the intensity at that time will be reset to zero and all subsequent results will have the intensity value that was reset to zero subtracted from them.</td>
</tr>
</tbody>
</table>
Selecting **Clear zero** cancels this zeroing (and thus subtraction) effect.

<table>
<thead>
<tr>
<th>Pre-scan</th>
<th>Select <strong>Prescan</strong> to determine the optimum instrument parameters to be used for your sample. At the end of this collection sequence the software will report the optimum Ex and Em wavelengths and PMT voltage. Once you are satisfied with the parameters, the software will automatically update your method setup to reflect these new parameters.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reset</td>
<td>Select this command to re-initialize the instrument. This will perform the same initializing procedure that is performed when the instrument is powered on.</td>
</tr>
<tr>
<td>Rapid Result</td>
<td>Select this to display the <strong>Rapid Result</strong> dialog. Use this dialog to take a reading of a blank and multiple sample readings without changing the application method. This means that between runs, while new samples are being prepared and loaded, other samples can quickly be measured. This command can also be accessed by pressing F6.</td>
</tr>
<tr>
<td>Zero</td>
<td>Select this to zero the current intensity value. The intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the <strong>Zero</strong> command before selecting <strong>Start</strong> if you want your data to start at 0 intensity. This command is also accessible by pressing F5. <strong>Note</strong> Before you select <strong>Zero</strong>, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.</td>
</tr>
<tr>
<td>Correction Curves</td>
<td>To collect correction factors, select from one of the following: <strong>Excitation (220...660 nm)</strong> If you select this option, the Cary Eclipse will do the following: 1. Set the emission mono to 640 nm and prompt you to insert a concentrated RhB solution 2. Perform an excitation scan from 220 to 600 nm The excitation correction factors will then be calculated and stored to the following file path on the directory on which the Cary Eclipse software is installed: Varian/Cary Eclipse WinFLR/Correction Curves/Excorr.csv. <strong>Emission (220...660 nm)</strong> If you select this option, the Cary Eclipse will do the following: 1. Prompt you to insert a diffuser 2. Perform a synchronous scan from 220 to 600 nm (with delta set to 0 nm) The emission correction factors will then be calculated and stored to the following file path on the directory on which the Cary Eclipse software is installed: Varian/Cary Eclipse WinFLR/Correction Curves/Excorr.csv. <strong>Emission (&gt;660 nm)</strong> An NIST calibrated source is required to perform this correction. Example of uncorrected and corrected scans.</td>
</tr>
<tr>
<td>Go to...</td>
<td>Select <strong>Go To...</strong> to display the <strong>Go To Wavelength</strong> dialog where you can change the instrument to a new wavelength. Use this command if you want to quickly go to a wavelength to check the intensity reading. This command is only available if the instrument is not currently collecting data. It can also be accessed by pressing F4. <strong>Note</strong> This is not the wavelength used in the application method. If your method uses a wavelength that is different from the 'Go To wavelength', then when you select Start the system will change to the wavelength selected. (In most applications, this is set in the Cary...</td>
</tr>
</tbody>
</table>
How to perform a fluorescence scan—Scan

This demonstrates how to perform a wavelength scan in fluorescence mode.

- Start the Scan application
- Set up data collection parameters
- Set up instrument parameters such as Data Mode and Scan Setup
- Set up visual system monitoring
- Set up the scan options
- Make sure that no accessories are selected
- Set up reporting and printing requirements
- Set up storage of collected data before run
- Set up email options
- Finish Setup
- Zero the instrument
- Start the Scan run
- Save the collected data

Start the Scan application

Step 1

Click the Windows Start button, point to Programs, Cary Eclipse and then select Scan. The Scan Application is displayed.

Note

If you are running a GLP system you may be prompted to enter a password before accessing the Scan Application.

Set up data collection parameters

Step 2 [Setup dialog]

Select the Setup button to display the Setup dialog and specify the method parameters for a new method.

Set up instrument parameters such as Data Mode and Scan Setup
Step 3 [Setup dialog | Cary page]

a. Set the **Data Mode** to **Fluorescence**.

b. Set the **Scan Setup** mode to **Emission**. During the emission scan, the excitation monochromator is set to a fixed wavelength and the emission monochromator is scanned over a wavelength range.

c. Set the **X Mode** to **Wavelength (nm)**.

d. Enter an **Excitation (nm)** value that is within the region where the fluorescent molecule to be scanned will absorb light. For purposes of this procedure, Anthracene will be used as an example and therefore a value of **360 nm** should be set.

e. Enter an **Excitation slit (nm)** value of 5 and an **Emission slit (nm)** value of 5. Slits determine the resolution of the spectrum and therefore are used in conjunction with the **PMT Detector Voltage** (set on the **Options** page) to determine concentration. If a compound is highly fluorescent and has reasonable signal intensity (as is the case for Anthracene) the slits can be set quite narrow.

f. Enter a **Start (nm)** value of 370 nm and enter a **Stop (nm)** value of 520 nm. The Start (nm) value should be set to the Excitation (nm) value plus the sum of the slits. In this example, as the Excitation value is 360 nm, and the sum of the slits is 10 nm, the Start (nm) value is set to 370. Typically the Stop (nm) value should be set to 150–200 nm greater than the Start (nm) value.

g. If selected, clear the **3-D Mode** check box.

h. Set the **Scan Control** to **Medium**.

**Set up visual system monitoring**

Step 4 [Setup dialog | Cary page]

Select the **Status Display** check box so that you can view various instrument parameters during the scan.

**Set up the scan options**

Step 5 [Setup dialog | Options page]

a. Select the **Options** tab to display the **Options** page.

b. In the **Display Options** group, select the **Overlay traces** check box to overlay the results of the traces on one graph.

c. If selected, clear the **CAT or S/N Mode, Cycle Mode** and **Smoothing** check boxes.

d. Set the **Excitation filter** to **Auto**. This will automatically move the filter wheel to the appropriate position for the selected excitation wavelength.

e. Set the **Emission filter** to **Open** in order to minimize steps in the spectra.

f. Set the **PMT Detector Voltage** to Medium. This setting can later be adjusted if results are over-range. If the signal is too high, decrease the PMT Detector Voltage. If the signal is too low, increase the PMT Detector Voltage.

**Make sure that no accessories are selected**

Step 6 [Setup dialog | Accessories page]
Ensure that no options are selected on any of the tabs of the **Accessories** page.

### Set up reporting and printing requirements

**Step 7** [Setup dialog | Reports page]

a. Select the **Reports** tab to display the **Reports** page where you can specify your reporting requirements for this method.

   i. Enter your name in the **Name** entry field.

   ii. If required, enter any comments relating to your experiment in the **Comment** field.

   iii. Set up your report style by selecting the appropriate check boxes in the **Options** group.

   For example, select the **Auto print** check box to obtain a printout of your report automatically.

   Select the **Parameters** check box to include your experimental parameters in the report.

   Select the **Graph** check box to include a graph in the generated report.

   ![Note](image)

   If **Auto print** is selected, the system will send the report information to the specified printer as well as displaying it in the Report area. However if **Auto print** is not selected, the report will be displayed in the Report area only. The Report area can be viewed by selecting the **Report** command from the **View** menu.

b. Set up the **Peaks** reporting options.

   i. Select **Maximum peaks** to report the peak with the largest peak threshold that exceeds the peak **Threshold** value.

   ii. Select **All peaks** to report all peaks meeting the **Peak type** criterion and exceeding the **Threshold** value.

   iii. Select the **Peak type** and specify the peak **Threshold**.

c. If required, select the **X-Y Pairs table** check box. You can use the **Actual Data Interval** by which the data was collected or you can make the Cary Eclipse Interpolate the points to a new interval.

### Set up storage of collected data before run

**Step 8** [Setup dialog | Auto Store page]

a. Select the **Auto Store** tab to display the **Auto store** page. Use this page to set up whether the collected data is to be saved, and if so, when the Cary Eclipse should store the information.

b. Select **Storage** option of **On; Prompt at end**.

c. Select the **Auto convert** option you require.

   If you select **Select for ASCII (csv)** or **Select for ASCII (csv) with Log**, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format in the current folder.

### Set up email options

**Step 9**

Refer to [How to email your results](#) for instructions on setting up the email options.

### Finish Setup
Step 10

Once you are satisfied with your method setup select **OK** to confirm any changes you have made and close the **Setup** dialog.

**Zero the instrument**

Step 11

a. Place the blank solution in the sample compartment of the Cary Eclipse instrument.

b. Click the **Zero** button to zero the system. Alternatively, select **Zero** from the **Commands** menu to perform a zero. When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the Scan Application window.

**Start the Scan run**

Step 12

a. Select the **Start** button to commence a data collection. Alternatively, select **Start** from the **Commands** menu. The **Sample Name** dialog is displayed.

b. Place the sample in the sample compartment of the Cary Eclipse instrument.

c. In the **Sample Name** dialog, enter the appropriate name for your sample and select **OK**. The scan will commence and the trace will appear in the Graphics area.

**Save the collected data**

Step 13

a. Once the run is finished, select the **Save As** command from the **File** menu.

b. In the **File name** field, enter a file name for this scan run.

c. Select **Save**. The data will be stored as a Batch file.

See also:

**Scan How To**

**How to perform a bio-/chemi-luminescene scan—Scan**

This demonstrates how to perform a wavelength scan in bio-/chemi-luminescene mode.

- **Start the Scan application**
- **Set up data collection parameters**
- **Set up instrument parameters**
- **Set up visual system monitoring**
- **Set up the display and filter options**
- **Make sure that no accessories are selected**
- **Set up reporting and printing requirements**
- **Set up storage of collected data before run**
- **Set up email options**
- **Finish Setup**
- **Zero the instrument**
- **Start the Scan run**
- Save the collected data

**Start the Scan application**

Step 1

Click the Windows **Start** button, point to **Programs, Cary Eclipse** and then select **Scan**. The Scan Application is displayed.

**Note**

If you are running a GLP system you may be prompted to enter a password before accessing the Scan Application.

**Set up data collection parameters**

Step 2 [Setup dialog]

Select the **Setup** button to display the **Setup** dialog and specify the method parameters for a new method.

**Set up instrument parameters such as Data Mode and Scan Setup**

Step 3 [Setup dialog] | Cary page

a. Set the **Data Mode** to **Bio-/Chemi-luminescence**.

b. Select the **Options** button to display the **Bio-/Chemi-luminescence Options** dialog. Enter the **Gate time** to **200 ms**. The Gate Time is the overall reading time of the emission signal and is typically set to between 100–500 ms.

c. Select **OK** to clear the **Bio-/Chemi-luminescence Options** dialog and return to the Cary page. Note that in the **Scan setup** group, the **Excitation** and **Synchronous** options are unavailable as the lamp does not flash for a Bio-/Chemi-luminescence scan. Therefore, it is set to **Emission** mode.

d. Set the **X Mode** to **Wavelength (nm)**.

e. Enter a **Start (nm)** value of **300 nm** and enter a **Stop (nm)** value of between **750 nm**. The Start (nm) value should be set to the Excitation (nm) value plus the sum of the slits.

f. Enter an **Emission slit (nm)** value of **10**. Slits determine the resolution of the spectrum and therefore are used in conjunction with the **PMT Detector Voltage** (set on the Options page) to determine concentration.

g. In the **Scan control** group, set the **Averaging time** to **0.20000 s** and the **Data interval** to **1.0000 nm**.

**Set up visual system monitoring**

Step 4 [Setup dialog | Cary page]

Select the **Status Display** check box so that you can view various instrument parameters during the scan.

**Set up the display and filter options**

Step 5 [Setup dialog | Options page]

a. Select the **Options** tab to display the **Options** page.
b. In the **Display Options** group, select the **Overlay traces** check box to overlay the results of the traces on one graph.

c. If selected, clear the **CAT or S/N Mode**, **Cycle Mode** and **Smoothing** check boxes.

d. Set the **Emission filter** to **Open** in order to minimize steps in the spectra.

e. Set the **PMT Detector Voltage** to **High**. A **High** setting is recommended as luminescence spectra are generally quite weak and therefore require a sensitive system to detect them. This setting can later be adjusted if results are over-range. If the signal is too high, decrease the **PMT Detector Voltage**.

**Make sure that no accessories are selected**

Step 6 [Setup dialog | Accessories page]

Make sure that no options are selected on any of the tabs in the **Accessories** page.

**Set up reporting and printing requirements**

Step 7 [Setup dialog | Reports page]

a. Select the **Reports** tab to display the **Reports** page where you can specify your reporting requirements for this method.

   i. Enter your name in the **Name** entry field.
   
   ii. If required, enter any comments relating to your experiment in the **Comment** field.

   iii. Set up your report style by selecting the appropriate check boxes in the **Options** group.

   For example, select the **Auto print** check box to obtain a printout of your report automatically. Select the **Parameters** check box to include your experimental parameters in the report. Select the **Graph** check box to include a graph in the generated report.

   **Note**

   If **Auto print** is selected, the system will send the report information to the specified printer as well as displaying it in the Report area. However if **Auto print** is not selected, the report will be displayed in the Report area only. The Report area can be viewed by selecting the **Report** command from the **View** menu.

b. Set up the **Peaks** reporting options.

   iii. Select **Maximum peaks** to report the peak with the largest peak threshold that exceeds the peak **Threshold** value.
   
   i. Select **All peaks** to report all peaks meeting the **Peak type** criterion and exceeding the **Threshold** value.
   
   ii. Select the **Peak type** and specify the peak **Threshold**.

c. If required, select the **X-Y Pairs table** check box. You can use the **Actual Data Interval** by which the data was collected or you can make the Cary Eclipse Interpolate the points to a new interval.

**Set up storage of collected data before run**

Step 8 [Setup dialog | Auto Store page]

a. Select the **Auto Store** tab to display the **Auto store** page. Use this page to set up whether the collected data is to be saved, and if so, when the Cary Eclipse should store the information.
b. Select **Storage** option of **On; Prompt at end**.

c. Select the Autoconvert option you require.
   If you select Select for ASCII (csv) or Select for ASCII (csv) with Log, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format in the current folder.

**Set up email options**

Step 9

Refer to [How to email your results](#) for instructions on setting up the email options.

**Finish Setup**

Step 10

Once you are satisfied with your method setup select **OK** to confirm any changes you have made and close the **Setup** dialog.

**Zero the instrument**

Step 11

a. Place the blank solution in the sample compartment of the Cary Eclipse instrument.

b. Click the **Zero** button to zero the system. Alternatively, select **Zero** from the **Commands** menu to perform a zero. When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the Scan Application window.

**Start the Scan run**

Step 12

a. Select the **Start** button to commence a data collection. Alternatively, select **Start** from the **Commands** menu. The **Sample Name** dialog is displayed.

b. In the **Sample Name** dialog, enter the appropriate name for you sample and select **OK**. The Scan run will commence and the corrected trace will appear in the Graphics area.

**Save the collected data**

Step 13

a. Once the run is finished, select the **Save As** command from the **File** menu.

b. In the **File name** field, enter a file name for this scan run.

c. Select **Save**. The data will be stored as a Batch file.

See also: [Scan How To](#)

**How to perform a phosphorescence scan—Scan**

This demonstrates how to perform a wavelength scan in phosphorescence mode.
Start the Scan application

Step 1

Click the Windows Start button, point to Programs, Cary Eclipse and then select Scan. The Scan Application is displayed.

Note

If you are running a GLP system you may be prompted to enter a password before accessing the Scan Application.

Set up data collection parameters

Step 2 [Setup dialog]

Select the Setup button to display the Setup dialog and specify the method parameters for a new method.

Set up instrument parameters such as Data Mode and Scan Setup

Step 3 [Setup dialog] | Cary page

a. Set the Data Mode to Phosphorescence.

b. Select the Options button to display the Phosphorescence Options dialog. The values you enter for the options in this dialog depend upon the sample you are scanning.

For example, if you were analysing a sample such as Europium (Eu3+), which has a short decay time, it is recommended that you enter the following values:

- Total decay time: 0.005 s
- No. of flashes: 1
- Delay time: 0.100 ms
- Gate time: 1.0 ms

However, if you were analysing a sample such as Naphthalene, which is known to have a phosphorescence lifetime in the seconds regime, it is recommended that you enter the following values:

- Total decay time: 2 s
- No. of flashes: 10
- Delay time: 0.100 ms
- Gate time: 500 ms
c. Once you have made entered the values in the **Phosphorescence options** dialog, select **OK** to clear the dialog and return to the **Cary page**.

d. Set the **Scan Setup** mode to **Emission**. During the emission scan, the excitation monochromator is set to a fixed wavelength and the emission monochromator is scanned over a wavelength range.

e. Set the **X Mode** to **Wavelength (nm)**. Set the following parameters for Europium**III**:

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Excitation (nm)</td>
<td>340</td>
<td>The <strong>Excitation (nm)</strong> value must be within the region where the phosphorescent molecule to be scanned will absorb light.</td>
</tr>
<tr>
<td>Excitation slit (nm)</td>
<td>20</td>
<td>Slits determine the resolution of the spectrum and therefore are used in conjunction with the <strong>PMT Detector Voltage</strong> (set on the <strong>Options</strong> page) to determine concentration. If a compound is not highly phosphorescent (as is the case for Europium) the slits need to be set quite wide.</td>
</tr>
<tr>
<td>Emission slit (nm)</td>
<td>20</td>
<td></td>
</tr>
<tr>
<td>Start (nm)</td>
<td>360</td>
<td>The <strong>Start (nm)</strong> value should be set to the <strong>Excitation (nm)</strong> value plus the sum of the slits. In this example, as the Excitation value is 340 nm, and the sum of the slits is 20 nm, the <strong>Start (nm)</strong> value is set to 360. Typically phosphorescence samples should be scanned to above 600 nm, and preferably to between 700–750 nm, even if they fluoresce at as low as 300 nm.</td>
</tr>
<tr>
<td>Stop (nm)</td>
<td>700-750</td>
<td></td>
</tr>
</tbody>
</table>

f. Set the following parameters for Naphthalene:

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Excitation (nm)</td>
<td>290</td>
<td>The <strong>Excitation (nm)</strong> value must be within the region where the phosphorescent molecule to be scanned will absorb light.</td>
</tr>
<tr>
<td>Excitation slit (nm)</td>
<td>10</td>
<td>Slits determine the resolution of the spectrum and therefore are used in conjunction with the <strong>PMT Detector Voltage</strong> (set on the <strong>Options</strong> page) to determine concentration. If a compound is not highly phosphorescent (as is the case for Europium) the slits need to be set quite wide.</td>
</tr>
<tr>
<td>Emission slit (nm)</td>
<td>20</td>
<td></td>
</tr>
<tr>
<td>Start (nm)</td>
<td>310</td>
<td>The <strong>Start (nm)</strong> value should be set to the <strong>Excitation (nm)</strong> value plus the sum of the slits. In this example, as the Excitation value is 340 nm, and the sum of the slits is 20 nm, the <strong>Start (nm)</strong> value is set to 360. Typically phosphorescence samples should be scanned to above 600 nm, and preferably to between 700–750 nm, even if they fluoresce at as low as 300 nm.</td>
</tr>
<tr>
<td>Stop (nm)</td>
<td>800</td>
<td></td>
</tr>
</tbody>
</table>

g. If selected, clear the **3-D Mode** check box.

**Set up visual system monitoring**

**Step 3 [Setup dialog | Cary page]**

Select the **Status Display** check box so that you can view various instrument parameters during the scan.

**Set up the display and filter options**
Step 4 [Setup dialog | Options page]

a. Select the **Options** tab to display the **Options** page.

b. In the **Display Options** group, select the **Overlay traces** check box to overlay the results of the traces on one graph.

c. If selected, clear the **CAT or S/N Mode**, **Cycle Mode** and **Smoothing** check boxes.

d. Set the **Excitation filter** to **Auto**. This will automatically move the filter wheel to the appropriate position for the selected excitation wavelength.

e. Set the **Emission filter** to **Open** in order to minimize steps in the spectra.

f. Set the **PMT Detector Voltage** to **High**. A **High** setting is recommended for low emitting samples. This setting can later be adjusted if results are over-range. If the signal is too high, decrease the **PMT Detector Voltage**.

**Make sure that no accessories are selected**

Step 5 [Setup dialog | Accessories page]

In this example we are not using any accessory so ensure that no options are selected on any of the tabs in the **Accessories** page.

**Set up reporting and printing requirements**

Step 6 [Setup dialog | Reports page]

a. Select the **Reports** tab to display the **Reports** page where you can specify your reporting requirements for this method.

   i. Enter your name in the **Name** entry field.
   
   ii. If required, enter any comments relating to your experiment in the **Comment** field.

   iii. Set up your report style by selecting the appropriate check boxes in the **Options** group.

      For example, select the **Auto print** check box to obtain a printout of your report automatically. Select the **Parameters** check box to include your experimental parameters in the report. Select the **Graph** check box to include a graph in the generated report.

      ![Note]

      If **Auto print** is selected, the system will send the report information to the specified printer as well as displaying it in the Report area. However if **Auto print** is not selected, the report will be displayed in the Report area only. The Report area can be viewed by selecting the **Report** command from the **View** menu.

b. Set up the **Peaks** reporting options.

   iii. Select **Maximum peaks** to report the peak with the largest peak threshold that exceeds the peak **Threshold** value.
   
   i. Select **All peaks** to report all peaks meeting the **Peak type** criterion and exceeding the **Threshold** value.
   
   ii. Select the **Peak type** and specify the peak **Threshold**.

c. If required, select the **X-Y Pairs table** check box. You can use the **Actual Data Interval** by which the data was collected or you can make the Cary Eclipse Interpolate the points to a new interval.
Set up storage of collected data before run

Step 7 [Setup dialog | Auto Store page]

a. Select the Auto Store tab to display the Auto store page. Use this page to set up whether the collected data is to be saved, and if so, when the Cary Eclipse should store the information.

b. Select Storage option of On; Prompt at end.

c. Select the Autoconvert option you require.
   If you select Select for ASCII (csv) or Select for ASCII (csv) with Log, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format in the current folder.

Set up email options

Step 8

Refer to How to email your results for instructions on setting up the email options.

Finish Setup

Step 9

Once you are satisfied with your method setup select OK to confirm any changes you have made and close the Setup dialog.

Zero the instrument

Step 10

Note:
This step is only necessary if there is background phosphorescence.

Note:
You MUST close the sample compartment prior to commencing a phosphorescence scan, as phosphorescence scanning is not immune to room light.

1. Place the blank solution in the sample compartment of the Cary Eclipse instrument.

2. Click the Zero button to zero the system. Alternatively, select Zero from the Commands menu to perform a zero. When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the Scan Application window.

Start the Scan run

Step 11

a. Select the Start button to commence a data collection. Alternatively, select Start from the Commands menu. The Sample Name dialog is displayed.

b. Place the sample in the sample compartment of the Cary Eclipse instrument.
c. In the **Sample Name** dialog, enter the appropriate name for your sample and select **OK**. The Scan run will commence and the corrected trace will appear in the Graphics area.

**Save the collected data**

**Step 12**

a. Once the run is finished, select the **Save As** command from the **File** menu.

b. In the **File name** field, enter a file name for this scan run.

c. Select **Save**. The data will be stored as a Batch file.

See also:

- Scan How To

**Viewing run progress and results—Scan**

- Reports page
- Reports toolbar
- Create Report
- Clear Report
- Label Options dialog
- Peak Labels dialog
- How to use the advanced reporting options
- View menu
- Edit menu
- Status display
- User Data Form
- Print Preview dialog
- Print

**View menu—Scan**

The View menu is accessible from the menu line at the top of the Scan Application window.

**Hot Tip**

Highlight a method or data file in Explorer and simply drag and drop it onto the application to open it. Any data will be displayed in the Graphics area.

The following commands can be accessed from the View menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>Use this to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Toolbars</td>
<td>Select this option to toggle the Graphics and Reports toolbar on and off. You can also select to display or hide the toolbar Text Labels.</td>
</tr>
<tr>
<td>Buttons</td>
<td>Select this option to toggle the left-hand button display (button area) on and off.</td>
</tr>
<tr>
<td>Graphics</td>
<td>Select this option to toggle the <strong>Graphics area</strong> on and off.</td>
</tr>
<tr>
<td>Report</td>
<td>Select this option to toggle the Report area on and off.</td>
</tr>
<tr>
<td>Report Vertical</td>
<td>If the Graphics option is selected, select <strong>Report Vertical</strong> to display the Graphics area, Report area and/or ADL Editor side by side vertically.</td>
</tr>
</tbody>
</table>
**Edit menu—Scan**

Use the **Edit** menu to copy, cut, paste and delete information. You can also use the menu to access the Report editing commands. The menu is accessible from the menu line at the top of the Scan Application window.

The following commands can be accessed from the **Edit** menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>Button</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Undo</strong></td>
<td><img src="image" alt="Undo" /></td>
<td>If available, this undoes the last action that was performed in the report area. This command is also accessible by pressing CTRL+Z.</td>
</tr>
<tr>
<td><strong>Cut</strong></td>
<td><img src="image" alt="Cut" /></td>
<td>Select this command to remove the highlighted text and place it on the Windows Clipboard. This command is also accessible by pressing CTRL+X.</td>
</tr>
</tbody>
</table>

See also:  
**ADL Hooks**  
How to insert ADL commands using the Quick Reference

---

**ADL**  
Select this option to toggle the ADL Editor on and off.

**ADL Vertical**  
If the ADL and Report options are selected, select **ADL Vertical** to display the ADL Editor vertically on the right hand side of the Report area.

**ADL Quick Reference**  
This displays the **Quick Reference** window which you can use to easily add a command in the correct syntax to your ADL program. A detailed description of all the commands in the Quick Reference is available by selecting the **Help** button on this window.

**ADL Hooks**  
Select this to activate a link (or hook) to an ADL program at a specific point in the application, and then run that ADL program. For example, ADL hooks have been supplied to run specific programs:

- on start-up of the Cary Eclipse software;
- when the system goes online or offline;
- when the Start button is selected;
- when a run finishes;
- when a zero is performed etc.

For the hook to work, ADL Hooks must be selected, and the appropriate ADL file for the application must be present in the Varian/Cary Eclipse WinFLR/ADL folder on the drive on which the Cary Eclipse software is installed.

**Note**  
Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.

**Hints**  
Select this option to toggle the Hints bubble text on and off. Hint text appears when you place the arrow cursor over a field, button or text entry field for a short period. It offers a quick explanation of the item’s function.

**Status Display**  
Select this option to display the **Status Display window**, where you can view the status of various parameters during the data collection.

---

file://C:\Documents and Settings\cary\Local Settings\Temp\~hhF1E6.htm
Use the **Status Display** window to view the status of your scan during the data collection. Depending upon settings you have for the scan, the following parameters may be displayed:

- Current Excitation Wavelength (nm)
- Current Emission Wavelength (nm)
- Excitation Slit Width (nm)
- Emission Slit Width
- PMT Voltage (V)

To access the **Status Display** window, select **Status Display** from the **View** menu, or from any page of the **Setup** dialog. You can choose to view the Status Display window at any time during a data collection.

### See also:
- **Status Line**
- **View menu**
- **Setup dialog**

---

<table>
<thead>
<tr>
<th>Command</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Copy</strong></td>
<td>Select this command to copy the highlighted text onto the Windows Clipboard. This command is also accessible by pressing CTRL+C.</td>
</tr>
<tr>
<td><strong>Paste</strong></td>
<td>Select this command to insert the contents of the Windows Clipboard (text only) at the cursor. Any highlighted text will be replaced. This command is also accessible by pressing CTRL+V.</td>
</tr>
<tr>
<td><strong>Delete</strong></td>
<td>Select this command to remove selected text in the Report area. This command is also accessible by pressing DEL.</td>
</tr>
<tr>
<td><strong>Select All</strong></td>
<td>Select this option to highlight all text in the Report area. This command is also accessible by pressing CTRL+A.</td>
</tr>
<tr>
<td><strong>Font</strong></td>
<td>This enables you to change the font of new or selected text in the report. For existing text, simply highlight the text in the report, select <strong>Font</strong> and make your changes in the <strong>Font</strong> dialog. When you select <strong>OK</strong>, the appearance of the highlighted text in the report will alter to reflect your font changes. This command is also accessible by pressing CTRL+F.</td>
</tr>
<tr>
<td><strong>Edit Report</strong></td>
<td>Select this option to edit the report. This command is also accessible by pressing CTRL+R.</td>
</tr>
<tr>
<td><strong>Clear Report</strong></td>
<td>Select this command to remove the contents of the Report area. If you have saved the report, selecting this option does not delete the contents of the report, the information is simply removed from the Report area.</td>
</tr>
</tbody>
</table>

**Hot Tip**
You can copy a picture, spreadsheet file or document from another application (for example, Microsoft Word or Microsoft Excel) into the report area of the of the Scan application. To edit these objects, simply double click on them to open the associated application in which they were created.

**See also:**
- **Scan Application**

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**User Data Form—Scan**
Use the User Data Form to enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data.

To access the User Data Form, select the User Data Form command from the Graph menu, or select the User Data Form button on the Graphics toolbar. The User Data Form command is only available if you have selected a graph box.

The dialog comprises the following:

**Data Name**  This column lists the names of each visible trace in the highlighted graph box. They will be labeled according their Sample Name.

**Other Columns**  Use these columns for recording additional information about the experimental conditions you used to collect the data. For example, if you are interested in time based data, you can record the time at which the sample was run. You do not need to include an entry for each corresponding sample name. Use the first row in the column to enter a name for the parameter (for example, 'Time').

You can choose to view these selected data points as a separate group of traces. To do this, open the Trace Preferences dialog and in the Select trace list, select the group of traces you wish to view. Trace groups will be named according to their column heading in the User Data Form (in the above example, it would be 'Time'). You can view traces as standard 2D graphs, or as contour or 3D graphs.

**Note**
You must enter a column heading, or else the data in that column will not be saved.

See also:
- Graph menu
- Graphics toolbar
- Trace Preferences
- Contour graphs
- 3D graphs

**Print Preview—Scan**

Use the Print Preview dialog to view the current report before you print it. From the File menu, select Print Preview to access the dialog.

The contents of the Graphics area appear first in the preview area followed by the contents of the Report area. You can reduce or enlarge the print preview on screen. To do this, click the left mouse button in the preview area to toggle through the five available views. Click the right mouse button in the preview area to return to the full page view.

The page comprises the following commands:

**First arrow**  Use this arrow to view the first page of the report.

**Previous arrow**  Use this arrow to view the previous page of the report.

**Next arrow**  Use this arrow to view the next page of the report.

**Last arrow**  Q. Why are the arrow buttons grayed out?
A. The buttons will be grayed out if the report has only one page of data.

**Print**  Use this to display the Windows Print dialog where you can choose the printer and pages to print as well as the number of copies you wish to print.
Print—Scan

Use the Print dialog to select the printer and pages to print of the currently displayed report as well as the number of copies you wish to print. From the File menu, select Print to access the dialog.

All graphical information displayed on screen will be printed with the report, therefore it is advisable to edit the report on the screen prior to printing. You can also preview the report prior to printing by using the Print Preview command.

If you require a printout of the graphics in the Graphics area only, select the Clear Report button to erase the contents of the Report area before you print.

Note
The options on the Report page are used to generate the contents of the Report area during a run or on a recalculation. If Auto Print has been selected the Cary Eclipse will automatically print a report at the end of a data collection.

Performing Calculations—Scan—Scan

- Maths Calculator
- Maths Operations
- Maths How To
- User Data Form

Maths window

Use the Maths window to perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator.

To access the Maths window, select the Calculator command from the Graph menu, or select the Calculator button on the Graphics toolbar.

The Maths window is only available in the Kinetics, Lifetimes, Scan and Validate applications.

The Maths window comprises the following:

- **Calculation display field**
  View the current calculation, including the trace/s being used, the trace operation being carried out and any functions that have been applied to the trace.

- **Selected Trace/Focused Trace**
  Choosing either of these options selects the highlighted (red) trace in the current Graph box for manipulation. When choosing between these options, use this simple rule:

  - Use the Selected Trace option when the selected calculation is only to be applied to one trace.

  Hot Tip
  Click on the down arrow adjacent to this field to view the last ten unique calculations that have taken place. Use this list to keep track of previous manipulations and repeat calculations quickly by simply selecting and then applying the desired calculation.
• Use the **Focused Trace** option when the same calculation is to be applied to a number of traces.

The following example illustrates the difference between these options.

To perform the same manipulation on several traces:

a. Highlight a trace by clicking on it with the mouse. The trace will become red.

b. Select **Selected Trace**, **Apply** and then **=** to plot the result of the calculation.

c. Highlight the next trace and repeat step b.

OR

a. Highlight the first trace.

b. Select **Focused Trace**, **Apply** and then **=** to plot the result of the calculation;

c. Highlight the next trace and select the **=** button to perform the same calculation on the newly selected trace.

**Trace**

Use the Trace group options to select the operation to be performed on the traces, the filter size and the interval (where applicable).

Click **Apply** to insert the selected operation into the equation.

Click **=** (equals) to display the result of the calculations.

**Operation**

Click on an operation to find out more about it:

- **Anisotropy**
- **Polarization**
- **Sum Function**
- **Mean**
- **Boxcar Averaging**
- **Moving Average**
- **Savitzky-Golay**
- **Spline**
- **Deriv 1**
- **Deriv 2**
- **Deriv 3**
- **Deriv 4**

**Display Options**

Select whether the results of the operation performed on the trace will be viewed in a New Graph box or in the Selected Graph box. If you choose the Selected Graph option, you will be able to directly compare the results with existing traces.

**Functions**

This group contains number and function buttons that can be used as a standard calculator in addition to being applied to traces.

To use the functions:

1. select a trace or value
2. select either a second trace or number
3. select the equals (**) button to display the result

All the basic mathematical functions are available (addition, subtraction, multiplication and division), as well as logarithm (**Log**) and square root (**Sqrt**) functions. When used as a calculator, the results of equations appear in the Report area.

**Additional buttons**

- **C**
- **CE**
Click **CE** to clear the last entry entered in the calculation display field.

=  
Click **(equals)** to start the operation shown in the calculation display field.

See also:
- [Maths Home Page](#)
- [Maths How To](#)

## Maths Operations

The following operations can be selected on the **Maths** window. To access the **Maths** window, select the **Calculator** command from the **Graph** menu, or select the **Calculator** button on the **Graphics** toolbar.

### Anisotropy, Polarization and Sum Function

The term **Anisotropy** refers to an unequal angular distribution about a point in space. Anisotropy also refers specifically to a simple expression for measuring the degree of polarization of the fluorescence. It is almost identical to the expression for polarization apart from having a factor of two in the denominator. **Anisotropy** is usually represented by the lowercase letter " \( r \) ", while polarization by an uppercase " \( P \) ".

The two expressions are listed below:

\[
\text{Anisotropy} \\
\begin{align*}
\text{r} &= \frac{I_{vv} - GI_{vh}}{I_{vv} + 2GI_{vh}} \\
\end{align*}
\]

\[
\text{Polarisation} \\
\begin{align*}
\text{P} &= \frac{I_{vv} - GI_{vh}}{I_{vv} + GI_{vh}} \\
\end{align*}
\]

where

- \( I_{vv} \) equals the intensity of fluorescence with excitation polarizer = vertical, emission polarizer = vertical
- \( I_{vh} \) equals the intensity of fluorescence with excitation polarizer = vertical, emission polarizer = horizontal
- \( G \) is a factor that accounts for the polarization bias of the instrument. In particular it accounts for the wavelength-dependent polarization bias of the emission side of the instrument (dominated by the polarization bias of the emission grating).

The \( G \) factor is given by:

\[
G = \frac{I_{hv}}{I_{hh}}
\]

where

- \( I_{hv} \) equals the intensity of fluorescence with excitation polarizer = horizontal, emission polarizer = vertical
- \( I_{hh} \) equals the intensity of fluorescence with excitation polarizer = horizontal, emission polarizer = horizontal

The expression for the anisotropy is a quotient. The denominator of the expression, as given by \( I_{vv} + 2GI_{vh} \) is the ‘sum’ function, and actually represents the total emission unbiased by any sample polarization effects.

### Mean

The **Mean** operation allows the average of two or more traces to be taken and the resulting trace to be plotted. The mean of the traces can also be used in conjunction with other functions.

The equation used in mean calculations is:
The calculation is performed over the common X range of the traces and the trace with the most
dense data is used to determine the data points. The other traces are interpolated at the relevant
data points.

**Boxcar Averaging**

This algorithm will take the number of points as defined by the Filter size, and average these,
displaying the average at the middle X-point of the filter.

For example, for the following:

- **Data:** \((x_1, y_1), (x_2, y_2), (x_3, y_3) \ldots (x_n, y_n)\)
- **Filer size:** 5

The first smoothed data point will be \((x_3, \text{sum}(Y_1:y_5)/5)\), the next \((x_8, \text{sum}(y_6:y_{10})/5) \ldots (x_{n-2}, \text{sum}(y_{n-4}:y_n)/5)\).

If there are not enough points at the end of the data set to complete the filter size, then these points
will be ignored, hence, the gap at the end may be larger than the gap in the beginning. There will
be a gap before the first point and after the last point of the smoothed continuum, with respect to
the original unsmoothed data. This algorithm is only valid for evenly x-spaced data points.

Within a time scan, (e.g., Advanced collect in kinetics) the data points may be unevenly spaced
between different stages. Each stage will be treated as a new collect, hence, there may be large
gaps between stages. If there are not enough points at the end of a stage to complete the filter
size, then these points are ignored, hence, there may be large gaps at the end of a stage. The filter
size will be limited to odd numbers, with the minimum of 3 to a maximum of 101.

**Moving Average**

This algorithm will take the number of points as defined by the Filter size, and average these,
displaying the average at the middle X-point of the filter.

For example, for the following:

- **Data:** \((x_1, y_1), (x_2, y_2), (x_3, y_3) \ldots (x_n, y_n)\)
- **Filer size:** 5

The first smoothed data point will be \((x_3, \text{sum}(Y_1:y_5)/5)\), the next \((x_4, \text{sum}(y_2:y_6)/5) \ldots (x_{n-2}, \text{sum}(y_{n-4}:y_n)/5)\). There will be a gap before the first point and after the last point of the smoothed continuum, with respect to the original unsmoothed data. This algorithm is only valid for evenly x-spaced data points. Within a time scan, (e.g., Advanced collect in kinetics), the data points may be unevenly spaced between different stages. This will be treated by interpolation of data from the previous stage so as to give evenly spaced data points into the next stage. If there is not enough data for interpolation, then the next stage will be treated as a new collect, hence, there may be gaps and further delays in displaying the point. The filter size will be limited to odd numbers, with the minimum of 3 to a maximum of 101.

**Savitzky-Golay**

The Savitzky-Golay technique operation enables you to reduce the noise in the currently selected
Maths scan by using smoothing.

When you select this operation you will be prompted to enter a Filter Size from 5 to 101. The
larger the Filter Size, the more data points that will be smoothed.

Q. I cannot enter a particular value; it keeps changing to another number — why?

A. The Savitzky-Golay technique requires an odd number to be entered, not an even number.

You will also need to enter an Interval. The Interval refers to how many of the averaged points
will be plotted on the smoothed trace. The smaller the interval, the more detail there will be in the trace.

Consider selecting an Interval of 1. This would mean that when the smoothed continuum is plotted, every smoothed point will be displayed.

Q. Why can't I see the Filter Size and Interval fields?
A. These will be displayed when you select Smooth in the Operation field.

**Spline**

**Deriv 1–4**
The Deriv operations allow you to obtain derivatives of a selected scan. The derivative can be of the orders 1 to 4. The resulting trace is then plotted. The Savitzky-Golay technique is used for derivative calculations.

On selection of the Deriv operation you will be prompted to enter a Filter Size from 5 to 101. The Filter Size refers to the number of points around a particular point that will be averaged to produce a new derived Y value. The lower the Filter Size, the more data points that will be used in the derivative trace.

Q. I cannot enter a particular value; it keeps changing to another number — why?
A. The Savitzky-Golay technique requires an odd number to be entered, not an even number.

You will also need to enter an Interval. The Interval refers to how many of the derived points will be plotted on the resulting trace. The smaller the Interval, the more detail there will be in the trace.

Q. Why can't I see the Filter Size and Interval fields?
A. These will be displayed when you select one of the ‘Derivs’ in the Operation field.

See also:
* About smoothing

**Maths How To…**

- How to perform a spectral subtraction
- How to perform a mean calculation
- How to smooth a trace
- How to derive a trace

See also:
* Maths Home Page

**Displaying data graphically—Scan**

- Graphics area
- Graphics How To
- **Graph menu**
- Graphics toolbar
- **Graph Labels**
- Add Label dialog
- Edit Label dialog
- How to add a label to a graph
- How to edit a label
- How to move a label
- How to resize a label
- How to delete a label
- **Graph Pictures**
Graphics area—Scan

The Graphics area of the Scan application appears in the top half of the Scan Application and is used to display traces of collected data. In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area surrounding the graph is referred to as the graph box. A graph will appear in the Graphics area when you select Start to commence a Scan.

To display or hide the Graphics area, select the Graphics command on the View menu. Double-clicking on the Graphics area enables you to toggle between half or full size display areas.

You can access the functions associated with the Graphics area by doing the following:

- selecting one of the Graphics toolbar buttons;
- selecting a command from the Graph menu; or
- clicking the right mouse button in the Graphics area and select a command from the menu

Note

Not all functions are available on every menu.

You can customize the graphics area to suit your requirements in the following ways:
<table>
<thead>
<tr>
<th>To…</th>
<th>Use the…</th>
<th>Tell me how…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Change the axes scaling automatically</td>
<td>Autoscale graph command</td>
<td>How to scale your graph automatically</td>
</tr>
<tr>
<td>Change the axes scaling manually</td>
<td>Axes Scales dialog</td>
<td>How to scale your graph manually</td>
</tr>
<tr>
<td>Modify the graph appearance (axes, trace)</td>
<td>Graph Preferences dialog</td>
<td>How to alter the look of your graphs</td>
</tr>
<tr>
<td></td>
<td></td>
<td>How to specify the number of tick marks and decimal places for your X and Y axes</td>
</tr>
<tr>
<td>Add a new graph to the display</td>
<td>Add graph command</td>
<td></td>
</tr>
<tr>
<td>Remove a graph from the display</td>
<td>Remove graph command</td>
<td></td>
</tr>
<tr>
<td>View single or multiple graphs</td>
<td>Single/multiple graph command</td>
<td></td>
</tr>
<tr>
<td>Resize a graph box</td>
<td>Cursor</td>
<td>How to resize graph boxes</td>
</tr>
<tr>
<td>Track the focused trace</td>
<td>Cursor Mode dialog</td>
<td>How to track the focused trace</td>
</tr>
<tr>
<td>Select the focused trace</td>
<td>Cursor</td>
<td>How to select the focused trace</td>
</tr>
<tr>
<td>Display a trace in the graph</td>
<td>Trace Preferences dialog</td>
<td>How to display a trace that is not appearing in a graph box</td>
</tr>
<tr>
<td>Remove a trace from a graph</td>
<td>Trace Preferences dialog</td>
<td>How to remove a trace from a graph</td>
</tr>
<tr>
<td>Remove a trace from the trace list</td>
<td>Trace Preferences dialog</td>
<td>How to remove a trace from the trace list</td>
</tr>
<tr>
<td>Zoom in and out on the graph</td>
<td>Zoom In and Zoom Out commands</td>
<td>How to zoom a graph</td>
</tr>
<tr>
<td>Add a label to the graph box</td>
<td>Add Label dialog</td>
<td>How to add a label to a graph</td>
</tr>
<tr>
<td>Edit labels in the graph box</td>
<td>Edit Label dialog</td>
<td>How to edit a label</td>
</tr>
<tr>
<td>Move labels in the graph box</td>
<td>Cursor</td>
<td>How to move a label</td>
</tr>
<tr>
<td>Resize labels in the graph box</td>
<td>Cursor</td>
<td>How to resize a label</td>
</tr>
<tr>
<td>Delete labels from the graph box</td>
<td>Delete Annotation command</td>
<td>How to delete a label</td>
</tr>
<tr>
<td>Add a picture to the graph box</td>
<td>Add Picture dialog</td>
<td>How to add a picture to a graph</td>
</tr>
<tr>
<td>Move pictures in the graph box</td>
<td>Cursor</td>
<td>How to move a picture</td>
</tr>
<tr>
<td>Resize pictures in the graph box</td>
<td>Cursor</td>
<td>How to resize a picture</td>
</tr>
<tr>
<td>Replace pictures in the graph box</td>
<td>Edit Picture dialog</td>
<td>How to replace a picture</td>
</tr>
<tr>
<td>Delete pictures from the graph box</td>
<td>Delete annotation command</td>
<td>How to delete a picture</td>
</tr>
<tr>
<td>Save the Graphics area you have set up as a graphics template</td>
<td>Save As dialog (from the File menu)</td>
<td>How to create a graphics template</td>
</tr>
</tbody>
</table>
Note
The red trace in any graph box is the one on which all information appears and the one used for the cursor tracking. It is called the focused trace.

See also:
Graphics Toolbar

Graphics How To—Scan

Graphics

- How to alter the look of your graphs
- How to create a graphics template

Traces

- How to create a contour graph
- How to create a slice from a contour graph
- How to display a trace
- How to track the focused trace
- How to select the focused trace
- How to remove a trace from a graph
- How to remove a trace from the trace List

Sizing and scale

- How to resize graph boxes
- How to scale your graph manually
- How to scale your graph automatically
- How to zoom a graph

Labels

- How to add a label
- How to edit a label
- How to delete a label
- How to move a label
- How to resize a label

Pictures

- How to add a picture
- How to move a picture
- How to delete a picture
- How to replace a picture
- How to resize a picture

Graph menu—Scan

Introduction

Use the Graph menu to view data in various graphical formats or in multiple graphs in the Graphics area. You can double-click on the Graphics area to toggle the display through graph and report to graph only.

In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The
rectangular area surrounding the graph is referred to as the graph box. A graph will appear in the Graphics area when you select Start to commence a scan.

The **Graph** menu can be accessed in the following ways:

- from the main menu at the top of the Scan Application window
- by clicking the right mouse button in a graph box or the Graphics area

**Note**

The red trace in any graph box is the one on which all information appears and the one used for the cursor tracking. It is called the focused trace.

**Menu commands**

The following commands can be accessed from the **Graph** menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>Button</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Print XY point</strong></td>
<td></td>
<td>Select this to print the cursor's current X and Y coordinates in the report area. This option is only available from the Graph menu that appears when you right-click in a graph box.</td>
</tr>
<tr>
<td><strong>Copy Graph</strong></td>
<td></td>
<td>Select Copy Graph to copy the entire Graphics area to the Windows Clipboard. All of the graphs in the Graphics area will be copied to the Clipboard.</td>
</tr>
<tr>
<td><strong>Paste to Graph</strong></td>
<td></td>
<td>Select Paste to graph to paste the contents of the Windows Clipboard into the Graphics area. Text is pasted as a label, graphics are pasted as pictures.</td>
</tr>
<tr>
<td><strong>Add Graph</strong></td>
<td>![button]</td>
<td>Select Add graph to add a new graph box to the Graphics area. You may want to do this if you have, for example, a graph containing existing data and you want to collect some new data in a new graph.</td>
</tr>
<tr>
<td><strong>Remove Graph</strong></td>
<td>![button]</td>
<td>Select Remove graph to delete the selected graph box from the Graphics area. The graph box is removed, but the data is not deleted and is still available from the Trace Preferences window. You may want to use remove graph if you have a large number of graph boxes open but only want to focus on some of these. <strong>Note</strong> You can remove multiple graph boxes by first holding down the CTRL key and selecting the graphs to be removed and then selecting the Remove Graph button or menu item. Use the SHIFT key to highlight a range of graphs for removal.</td>
</tr>
<tr>
<td><strong>Clear All Traces</strong></td>
<td>![button]</td>
<td>Select Clear All Traces to delete all traces from the graph/s. Note that selecting this option simply turns off the Visible property for the traces in the Trace Preferences window. You can redisplay the traces by making the traces Visible again in the Trace Preferences window.</td>
</tr>
<tr>
<td><strong>Add Label</strong></td>
<td>![button]</td>
<td>Select Add Label to display the Add Label dialog where you can enter a text label to be placed on a graph or in the background of the Graphics area.</td>
</tr>
<tr>
<td><strong>Add Picture</strong></td>
<td>![button]</td>
<td>Select Add Picture to display the Add Picture dialog where you can select a picture, bitmap or chemical structure to be placed on a graph or in the background of the Graphics area.</td>
</tr>
<tr>
<td><strong>Edit Annotation</strong></td>
<td>![button]</td>
<td>Select Edit Annotation to alter the content or look of the selected annotation, (either a label or a picture) in the Graphics area. <strong>Note</strong></td>
</tr>
<tr>
<td><strong>Delete Annotation</strong></td>
<td>Select <strong>Delete Annotation</strong> to remove an annotation, (such as a label or a picture) from the Graphics area. You can use this menu option to remove highlighted annotations in the Graphics area such as labels, pictures and ruler/fit information.</td>
<td></td>
</tr>
<tr>
<td>----------------------</td>
<td>---------------------------------------------------------------------------------------------------</td>
<td></td>
</tr>
<tr>
<td><strong>Trace Preferences</strong></td>
<td>Select this command to display the <strong>Trace Preferences</strong> dialog where you can select the trace(s) to display in the selected graph.</td>
<td></td>
</tr>
<tr>
<td><strong>Graph Preferences</strong></td>
<td>Select this command to display the <strong>Graph Preferences</strong> dialog where you can change the appearance of your graph. Any changes you make to the appearance of the graph will be applied to all graphs.</td>
<td></td>
</tr>
<tr>
<td><strong>Cursor Mode…</strong></td>
<td>Select this command to display the <strong>Cursor Modes</strong> dialog where you can choose the type of cursor to be used on the screen. For example, you can select to use a free cursor that can move anywhere in the graph or exactly trace the focused scan (red scan). Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the Least Squares method or the Point to Point method.</td>
<td></td>
</tr>
<tr>
<td><strong>Axes Scales…</strong></td>
<td>Select this command to display the <strong>Axes Scales</strong> dialog where you can set the scale range for the X and Y axes.</td>
<td></td>
</tr>
</tbody>
</table>
| **Auto scale graph** | To automatically scale the graph, point to one of the following options:

- **Autoscale[XY]**
  Select Autoscale[XY] to automatically scale collected data by scaling the Y height and X width to full screen.

- **Autoscale[X]**
  Select Autoscale[X] to automatically scale collected data by scaling the X width to full screen.

- **Autoscale[Y]**
  Select Autoscale[Y] to automatically scale collected data by scaling the Y height to full screen. |
| **Zoom Out** | Select **Zoom Out** to Zoom back to the original X and Y ranges which were used to collect the data (i.e. the method setup conditions of the focused scan). For example, the system will revert back to the X and Y ranges set in the method if you zoom in and out during a data collection. |
| **Single/Multi Graph** | Select **Single/Multi Graph** to toggle between displaying one or all graphs.

**Note**
The graph highlighted in the **Multi Graph** view will be displayed when you toggle the **Single Graph** option. |
| **Auto** | Select **Auto Arrange Graphs** to view and arrange all graphs simultaneously. |
| **Arrange Graphs** | **Note**
<table>
<thead>
<tr>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>You can move graph boxes manually by clicking the left mouse button outside the graph axes and dragging the graph box to a new position. You can also resize graphs.</td>
</tr>
</tbody>
</table>

| **User Data Form** | **Select User Data Form** to display the User Data Form where you can enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data, such as the pH at which data was collected. |

| **Peak Labels** | **Select this to the Peak Labels dialog where you can select various labeling and peak reporting functions.** |

| **Calculator** | **Select Calculator** to display the Maths window where you can perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator. |

| **Contour graph** | **Once you have generated a graph of data, you can select the Contour graph button to generate a Contour graph of this data.** |

| **XY slice** | **Once you have generated a Contour graph, you can generate an XY slice of the data, which will enable you to view a XY scan of a particular point of the XY graph.** |

| **YZ slice** | **Once you have generated a Contour graph, you can generate an YZ slice of the data, which will enable you to view a YZ scan of a particular point of the YZ graph.** |

| **3D graph** | **Once you have generated a graph of data, you can select the 3D graph button to generate a 3D graph of this data. The 3D Grams application by Galactic software® will be displayed.** |

| **History List** | **This displays a list of the current graphs.** |

---

**See also:**
- Add Label
- Add Picture
- Edit Label
- Edit Picture
- Trace Preferences
- Graph Preferences
- Cursor Modes
- Axes Scales
- Single/Multi Graph
- User Data Form
- Peak Labels
- Maths window
- Contour graphs
- 3D graph
- Graphics Toolbar
- Graphics area

**Graphics Toolbar—Scan**

Use the Graphics toolbar buttons to gain quick access to many of the Graph menu options. You can also use the toolbar to select the graph you want to view and to change the look of your graphs.

To display or hide the toolbar, point to the **Toolbars** option in the **View** menu and select **Graphics**. To display or hide the toolbar's text labels, point to the **Toolbars** option in the **View** menu and select **Text Labels**.

file://C:\Documents and Settings\cary\Local Settings\Temp\~hhF1E6.htm
The following commands can be accessed from the Graphics Toolbar:

<table>
<thead>
<tr>
<th>Button</th>
<th>Menu command</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Trace Preferences</td>
<td>Click this button to display the Trace Preferences dialog where you can select the trace (s) to display in the selected graph.</td>
<td></td>
</tr>
<tr>
<td>Graph Preferences</td>
<td>Click this button to display the Graph Preferences dialog where you can change the appearance of your graph. Any changes you make to the appearance of the graph will be applied to all graphs.</td>
<td></td>
</tr>
<tr>
<td>Cursor Mode</td>
<td>Click this button to display the Cursor Modes dialog where you can choose the type of cursor to be used on the screen. For example, you can select to use a free cursor that can move anywhere in the graph or exactly trace the focused scan (red scan). Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the Least Squares method or the Point to Point method.</td>
<td></td>
</tr>
<tr>
<td>Axes Scales…</td>
<td>Click this button to display the Axes Scales dialog where you can set the scale range for the X and Y axes.</td>
<td></td>
</tr>
<tr>
<td>Autoscale [XY]</td>
<td>Click the Autoscale[XY] button to automatically scale collected data by scaling the ordinate (Y) height and abscissa (X) width to full screen.</td>
<td></td>
</tr>
<tr>
<td>Autoscale[X]</td>
<td>Click the Autoscale[X] button to automatically scale collected data by scaling the abscissa (X) width to full screen.</td>
<td></td>
</tr>
<tr>
<td>Autoscale[Y]</td>
<td>Click the Autoscale[Y] button to automatically scale collected data by scaling the ordinate (Y) height to full screen.</td>
<td></td>
</tr>
<tr>
<td>Zoom Out</td>
<td>Click the Zoom Out button to Zoom back to the original X and Y ranges which were used to collect the data (i.e. the method setup conditions of the focused scan). For example, the system will revert back to the X and Y ranges set in the method if you zoom in and out during a data collection.</td>
<td></td>
</tr>
<tr>
<td>Add Label</td>
<td>Click this button to display the Add Label dialog where you can enter a text label to be placed on a graph or in the background of the Graphics area.</td>
<td></td>
</tr>
<tr>
<td>Add Picture</td>
<td>Click this button to display the Add Picture dialog where you can select a picture, bitmap or chemical structure to be placed on a graph or in the background of the Graphics area.</td>
<td></td>
</tr>
<tr>
<td>Single/Multi Graph</td>
<td>Select Single/Multi Graph to toggle between displaying one or all graphs.</td>
<td></td>
</tr>
<tr>
<td>Auto Arrange Graphs</td>
<td>Click the Auto Arrange Graphs button to view and arrange all graphs simultaneously.</td>
<td></td>
</tr>
<tr>
<td>Add Graph</td>
<td>Click this button to add a new graph box to the Graphics area. You may want to do this if you have, for example, a graph containing existing data is displayed and you want to collect some new data in a new graph.</td>
<td></td>
</tr>
</tbody>
</table>
| Remove Graph | Click this button to delete the selected graph box from the Graphics area. The graph box is removed, but the data is not deleted and is still available from the Trace Preferences.
Graph Labels—Scan

- Add Label dialog
- Edit Label dialog
- How to add a label to a graph
- How to edit a label
- How to move a label
- How to resize a label
- How to delete a label

Add Label—Scan

Use the Add Label dialog to place a label on your graph or into the background of the Graphics area.

To add a label to a graph box, you must have at least one trace Visible in the graph box. (You can select which traces you want to make visible in the Trace Preferences dialog.) If you are viewing multiple graphs, the label will be placed in the graph box that is currently selected. However, you can add a label outside of a graph box in the background of the Graphics area at any time.
Q. Can I add a label that will print with all my graphs.
A. Yes, you can add your label to the background of the Graphics area. To do this, right-click on a section of the background (gray area) and select Add Label from the menu. (You will notice that this menu has less options than the menu that appears when you right-click on a graph.) In the Add Label dialog, enter the label to be added to all graphs and select OK. The label will appear in the background of the Graphics area.

To access the Add Label dialog, select the Add Label command from the Graph menu, or select the Add Label button on the Graphics toolbar.

The Add Label dialog comprises the following:

- **To selected trace** Select this option to associate a label with the selected trace. When you associate a label with a trace and make the trace visible in the Graph box, the label will also be displayed with the trace.
- **To selected graph** Select this option to associate the label with the selected graph. When you associate a label with a graph, the label will always be displayed in the Graph box, regardless of what traces are visible.
- **Selected trace** This will identify the trace that is currently selected and display the name of the trace.
- **Font** Click Font to open the Windows Font dialog. In this dialog you can specify the font type, style and size to be used for the label.
- **Label** Use the Label field to enter the text for the label. You can enter as many lines of text as you require.

**Hot Tip**
If you wish to include certain XY co-ordinates on the graph as a label, position your cursor at the appropriate place, right-click and select Add Label from the menu. When the Add Label dialog appears, the co-ordinate positions will be added to the Label field.

**Note**
When you save the data (as a Batch or Data file) the label is saved with its associated trace or graph. Labels outside graph boxes can be saved in a graphics template. To do this, choose Graph Template as the Save As type in the Save As dialog. See 'Example of a Graphics Template'.

See also:
- Trace Preferences
- Graph menu
- Graphics toolbar
- How to add a label to a graph
- How to move a label
- How to edit a label
- How to resize a label
- How to delete a label

**Edit Label—Scan**

Use the Edit Label dialog to alter the content or look of your label.

To access the Edit Label dialog, right-click on the label you wish to change and select Edit Annotation from the menu.

The dialog comprises the following:

- **Font** Select Font to open the Font dialog, where you can specify the font type, style and size to be used for the label.
- **Label** Use the Label field to alter the text for the label.

See also:
- How to edit a label
How to add a label to a graph—Scan

You can add a label to your graph or Graphics area background. To do this:

Step 1

Click on the graph box you require or click outside the graph box(es) to choose the Graphics area background.

Note
There must be at least one trace Visible in the graph box before you can add a label to it.

Step 2

Click the Add Label button on the toolbar or right-click on the graph and select Add Label from the menu. The Add Label dialog will appear. The Add Label command is also accessible from the Graph menu.

Step 3

Click the Font button. The Windows Font dialog appears. Select the font type, size and style you require for your label. A sample of the font style appears in the Sample field. When you are satisfied with your selections, select OK to return to the Add Label dialog.

Step 4

Enter the label in the Label field and select OK. The label will be displayed on the selected graph. If necessary, you can move a label so that it is more appropriately placed on your graph.

See also:
How to move a label
How to edit a label
How to resize a label

How to edit a label—Scan

Step 1

Place the cursor on the label and click the left mouse button to select the label.

Hot Tip
If you are having trouble selecting the label it may be that a trace is interfering with the label selection. Try zooming in and clicking in a different part of the label.

Step 2

Click the right mouse button and select Edit Annotation from the drop-down menu. Alternatively you can select the Edit Annotation command from the Graph menu. The Edit Label dialog is displayed.

Step 3

Use the Edit Label dialog to change the label.
For example, if you want to change the font size of the label, select the Font button. In the Size field, scroll through the available sizes for the font and select the size you require. Alternatively, highlight the currently selected size in the Size entry field and enter the size you require. Click the OK button in this dialog to return to the Edit Label dialog.

Step 4

Click OK to clear the Edit Label dialog. Your label will appear with the new font size.

See also:
- How to add a label to a graph
- How to move a label
- How to resize a label
- How to delete a label

How to move a label—Scan

To move a label so that it is more appropriately placed on your graph or Graphics area background, do the following:

Step 1

Place the cursor on the label.

Step 2

Click and hold down the left mouse button.

Hot Tip
If you are having trouble selecting the label, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 3

Drag the label to the correct position and release the mouse button. The label will be displayed at the new position.

Note
Labels are transparent so as not to interfere with any traces on graphs.

See also:
- How to add a label to a graph
- How to edit a label
- How to resize a label
- How to delete a label

How to resize a label—Scan

To resize a label so that it is more appropriately sized on your graph or Graphics area background, do the following:

Step 1

Select the label by clicking on it with the mouse. A border will be displayed around the label to indicate it is selected.

Hot Tip
If you are having trouble selecting the label, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.
Step 2

Position the cursor over one of the corner edges of the label until the cursor becomes a diagonal double-headed arrow. Click and hold down the left mouse button.

Example of label showing the border that indicates it is selected and the arrow to be used for re-sizing

Step 3

Increase or decrease the size of the label by dragging the mouse. The label text will also be resized. Release the mouse button when the label and text are the correct size.

Note
Labels are transparent so as not to interfere with any traces on graphs.

See also:
How to add a label to a graph
How to edit a label
How to delete a label

How to delete a label—Scan

To delete a label from a graph or Graphics area background, do the following:

Step 1

Position the cursor on the label and click the left mouse button to select it.

Hot Tip
If you are having trouble selecting the label it may be that a trace is interfering with the label selection. Try zooming in and clicking in a different part of the label.

Step 2

Click with the right mouse button and select Delete Annotation from the drop-down menu displayed. Alternatively, you can select the Delete Annotation command from the Graph menu. The label will be deleted from the graph or Graphics area.

See also:
How to add a label to a graph
How to move a label
How to edit a label
How to resize a label

Graph pictures—Scan

- Add Picture dialog
- Edit Picture dialog
- How to add a picture
- How to replace a picture
- How to move a picture
- How to resize a picture
Add Picture—Scan

Use the Add Picture dialog to insert a picture or bitmap into the Graphics area. The picture can be added as a background picture or to a particular graph box.

To add a picture to a graph box you must have at least one trace **Visible** in the graph box. (You can select which traces you want to make visible in the Trace Preferences dialog.) If you are viewing multiple graphs, the picture will be placed in the graph box that is currently selected. However, you can add a picture outside of a graph box in the Graphics area at any time.

Q. Can I add a picture that will print with all my graphs.
A. Yes, you can add your picture to the background of the Graphics area. To do this, right-click on a section of the background and select Add Picture from the menu. (You will notice that this menu has less options than the menu that appears when you right-click on a graph.) In the Add Picture dialog, load the picture to be added to all graphs and select OK. The picture will appear in the background of the Graphics area.

To access the Add Picture dialog, select the Add Picture command from the Graph menu, or select the Add Picture button on the Graphics toolbar.

The Add Picture dialog comprises the following commands:

- **To selected trace**
  Select this option to associate a picture with the selected trace. When you associate a picture with a trace and make the trace visible in the Graph box, the label will also be displayed with the trace.

- **To selected graph**
  Select this option to associate the picture with the selected graph. When you associate a picture with a graph, the label will always be displayed in the Graph box, regardless of what traces are visible.

- **Load**
  Click Load to open the Windows Open dialog. Use this dialog to browse your computer for the picture you want to insert. The picture must be one of the following formats: DIB, JPG, PGE, BMP, EMF or WMF.

If you are viewing multiple graphs the picture will be placed on the graph that is currently selected. If no graph is selected, the picture will be placed on the Graphics area background.

When you save the data (as a Batch or Data file) the picture is saved with its associated trace or graph. Pictures outside graph boxes can be saved in a graphics template. To do this, choose Graph Template as the Save As type in the Save As dialog. See Example of a Graphics Template.

See also:
- How to add a picture to a graph
- How to move a picture
- How to resize a picture
- How to delete a picture
- How to replace a picture

Edit Picture—Scan

Use the Edit Picture dialog to change a picture you have inserted into the Graphics area.

To access the Edit Picture dialog, right-click on the picture you wish to change and select Edit Annotation from the menu.
The **Edit Picture** dialog comprises the following commands:

<table>
<thead>
<tr>
<th>Command</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>To selected</td>
<td>Displays whether you originally selected to add the picture to a trace or a graph. This option is not editable.</td>
</tr>
<tr>
<td>trace/ graph</td>
<td></td>
</tr>
<tr>
<td>Load</td>
<td>Click <strong>Load</strong> to open the Windows <strong>Open</strong> dialog. Use this dialog to browse your computer for the picture you want to insert. The selected picture will replace the current picture. The picture must be one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.</td>
</tr>
</tbody>
</table>

**See also:**
- [How to replace a picture](#)
- [How to add a picture](#)
- [How to move a picture](#)
- [How to resize a picture](#)
- [How to delete a picture](#)

### How to add a picture to a graph—Scan

You can add a picture to your graph or the Graphics area background. To do this:

Step 1

Click on the graph box you require or click outside the graph box(es) to choose the Graphics area background.

**Note**

There must be at least one trace Visible in the graph box before you can add a picture to it.

Step 2

Click the **Add Picture** button on the toolbar or right-click on the graph and select **Add Picture** from the menu. The **Add Picture** dialog will appear. The **Add Picture** command is also accessible from the **Graph** menu.

Step 3

Click the **Load** button. The Windows **Open** dialog appears.

Step 4

Browse through your directories to find the picture you want to insert. All pictures must be in the bitmap format and will have the extension .BMP.

Step 5

Highlight the .BMP that you want to insert and select **Open**.

Step 6

The **Add Picture** dialog will appear showing a preview of the bitmap. If the bitmap is the correct one select the **OK** button. The bitmap will be placed onto the graph.

**Note**

You should take care not to add a bitmap that is too big as it will obscure part or all of your graph.

You can move a picture so that it is more appropriately placed on your graph.
See also:
How to move a picture to a graph
How to delete a picture
How to replace a picture
How to resize a picture

How to replace a picture—Scan

To change a picture that you have added to a graph or Graphics area background do the following:

Step 1
Move the cursor to the picture and select the left mouse button to select it.

Hot Tip
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2
Click the right mouse button and select Edit Annotation from the drop-down menu or select Edit Annotation from the Graph menu. The Edit Picture dialog is displayed.

Step 3
From the Edit Picture dialog, select the Load button. The Windows Open dialog is displayed.

Step 4
Browse through your directories to find the picture to replace the current bitmap. Picture files must be in one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.

Step 5
Highlight the file that you want to insert and select Open to return to the Edit Picture dialog.

Step 6
The Edit Picture dialog will display a preview of the bitmap. If the bitmap is the correct one select the OK button. The bitmap will be placed onto the graph. If you need to, move the picture so that it is more appropriately placed on your graph.

Note
Do not to add a bitmap that is too big as it will obscure part or all of your graph.

See also:
How to add a picture to a graph
How to move a picture
How to delete a picture

How to move a picture—Scan

To move a picture so that it is more appropriately placed on your graph or Graphics area background, do the following:

Step 1
Place the cursor on the picture.

Step 2

Click and hold down the left mouse button.

**Hot Tip**

If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 3

Drag the picture to the correct position and release the mouse button. The picture will be displayed at the new position.

See also:
- How to add a picture to a graph
- How to delete a picture
- How to replace a picture
- How to resize a picture

**How to resize a picture—Scan**

To resize a picture so that it is more appropriately sized on your graph or Graphics area background, do the following:

Step 1

Select the picture by clicking on it with the mouse. A border will be displayed around the picture to indicate it is selected.

**Hot Tip**

If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Position the cursor over one of the corner edges of the picture until the cursor becomes a diagonal double-headed arrow. Click and hold down the left mouse button.

*Example of picture showing the border that indicates it is selected and the arrow to be used for re-sizing*

Step 3

Increase or decrease the size of the picture by dragging the mouse. The picture will also be resized. Release the mouse button when the picture is the correct size.

See also:
How to delete a picture—Scan

To delete a picture from a graph or Graphics area background, do the following:

Step 1

Position the cursor on the picture and click the left mouse button to select it.

_hot tip_
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Click with the right mouse button and select Delete Annotation from the drop-down menu displayed. Alternatively, you can select the Delete Annotation command from the Graph menu. The picture will be deleted from the graph or Graphics area.

see also:
How to add a picture to a graph
How to move a picture
How to zoom a graph
How to resize a picture

Trace Preferences—Scan

A single graph box can display many traces. Use the Trace Preferences dialog to select the trace(s) to display (make Visible) in the selected graph box.

To access the Trace Preferences dialog, select the Trace Preferences command from the Graph menu, or select the Trace Preferences button on the Graphics toolbar.

The dialog comprises the following:

_hot tip_
The Trace Preferences menu is also available on clicking the right mouse button anywhere in the trace listing.

Trace Preferences table

<table>
<thead>
<tr>
<th>Visible</th>
</tr>
</thead>
<tbody>
<tr>
<td>Use the Visible field to select which traces will appear on the selected graph. To select or clear the Visible field, simply click the left mouse button in this column. A tick will appear next to traces that are to be displayed in the selected graph.</td>
</tr>
</tbody>
</table>

If you are in single graph view, all selected traces will appear on the one graph. If you are in Multi Graph view the traces will appear on the graph that is currently selected.
Trace Options

Select Traces You can choose to include all traces of the graph you have produced in the Contours or 3D graph or a subset of these traces. The subsets you can choose from will depend upon the data points you have entered next to the samples in the User Data Form. For example, if you have included time based data points under the Data name of 'Time', this group of data points will be included in the Select trace list under the heading of 'Time'.

Note The following options are only visible if you have selected the Contour Graph button or the 3D Graph button to create a Contour or 3D graph.

Z Start Enter the value at which the Z axis will be started. Generally, this is the Start Excitation Wavelength that was entered on the Cary page of the Setup dialog. If you have selected All Traces, this option will not be available. Instead the axes will start at the first point you have defined for the selected traces. (These points are defined in the User Data Form.) For example, if you have selected to view Time traces only, the first data point for these traces will be used as the first point on the Z axis.

Z Interval Enter the interval, or the value between each point on the Z axis. Generally, this is the Delta that was entered on the Cary page of the Setup dialog. If you have selected All Traces, this option will not be available. Instead the intervals will be placed evenly amongst the data points you have defined for the selected traces. (These data points are defined in the User Data Form.) Note that the intervals may not correspond to actual data points.

Z Axis Enter a label for the Z axis. This label will be displayed in the graph area. If you have selected All Traces, this option will not be available. Instead the Data name you have assigned in the User Data Form will be used as the Z axis label.

Trace Audit Log
The **Trace Audit Log** displays information about the highlighted trace. This includes instrument parameters and data form information relating to the trace that has been selected. It also logs all information about data manipulation.

When more than one trace is highlighted, the **Trace Audit Log** will display information about the trace that was first selected.

---

**See also:**
- Trace Preferences menu
- Contour graphs
- Contour Graph Preferences
- 3D graphs
- User Data Form
- Graph menu
- Graphics toolbar

### Trace Preferences menu—Scan

The **Trace Preferences** menu is displayed when you click the right mouse anywhere on the Trace Preferences table.

The menu contains the following commands:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Select All</td>
<td>This selects or highlights all the traces in the listed on the Trace Preferences page.</td>
</tr>
<tr>
<td>Select None</td>
<td>This de-selects all the traces in the listing.</td>
</tr>
<tr>
<td>Show Traces</td>
<td>Select Show Traces to make all of the traces currently highlighted ‘Visible’, meaning that they will be displayed in the current graph box.</td>
</tr>
<tr>
<td>Hide Traces</td>
<td>Select Hide Traces to remove the ‘Visible’ property from all the traces currently highlighted, which will remove them from the current graph box.</td>
</tr>
<tr>
<td>Delete Traces</td>
<td>Select Delete Traces to delete all the highlighted traces in all graphs. Note that if you save the data file before you select Delete Traces, the traces are not deleted from the saved file, and they can be retrieved at a later date. Selecting Delete Traces will delete the traces from the application memory, and it is recommended that you select this option if you need to free up memory on your computer. <strong>Note</strong> If the application is controlled by the GLP Administration application, you will be prompted to save an unsaved data file before it is removed from the application.</td>
</tr>
<tr>
<td>Change Trace Color</td>
<td>Select Change Trace Color to display the Windows Color dialog where you can change the color of the currently highlighted trace(s). The focused trace will remain red while it retains the focus, and will revert to the selected color when the focus is moved.</td>
</tr>
</tbody>
</table>

---

**See also:**
- Trace Preferences page
- Graph Preferences page
- How to remove a trace from the trace list
- How to remove a trace from a graph

### How to select the focused trace—Scan

To select the scan that you want to be the focused (red) trace, do one the following:

- From the Graph, click the left mouse button on the trace that you want to make the focused trace.

- From the Graph Legend (if displayed), click the left mouse button on the trace that you want to make the focused trace.

- From the Trace Preferences dialog, click in the Color column of the trace that you want to make the focused trace.
**Note**
Select the **Show Legend** check box in the **Graph Preferences** dialog to display the **Graph Legend**.

**See also:**
*How to track the focused trace*
*How to view a particular trace*

### How to display a trace that is not appearing in the graph box—Scan

**Step 1**
Click on the graph in the Graphics area.

**Step 2**
Select the **Trace Preferences** button from the Graphics toolbar, or select **Trace Preferences** from the **Graph** menu. The **Trace Preferences** dialog is displayed.

**Step 3**
Find that trace that you wish to see and select the **Visible** option. A tick will appear in the **Visible** column to indicate that it will now be displayed.

If you wish to make multiple traces visible, do the following:

a. Hold down the CTRL key select all the traces that you want to display.

b. Click the right mouse button and select the **Show Traces** command from the drop-down menu. Ticks will appear next to the selected traces to indicate that they will now be displayed.

**Step 4**
Press **OK** to clear the **Trace Preferences** dialog. The selected trace(s) will be displayed.

**See also:**
*How to remove a trace from a graph*
*How to remove a trace from the trace list*

### How to remove a trace from a graph—Scan

**Step 1**
Select a graph in the Graphics area.

**Step 2**
Click the **Trace Preferences** button on the Graphics toolbar or select **Trace Preferences** from the **Graph** menu. The **Trace Preferences** dialog is displayed.

**Step 3**
Find that trace that you no longer wish to see and select the **Visible** option. The tick will be removed from the **Visible** column to indicate that it will not be displayed.

If you wish to make multiple traces visible, do the following:
a. Hold down the CTRL key select all the traces that you no longer want to display.

b. Click the right mouse button and select the **Hide Traces** command from the drop-down menu. The ticks will be removed from the selected traces to indicate that they will not be displayed.

Step 4

Select **OK** to clear the **Trace Preferences** dialog. The trace will be removed from the graph.

See also:
- How to display a trace that is not appearing in the graph box
- How to remove a trace from the trace list

**How to remove a trace from the trace list—Scan**

If your graphs contain unwanted traces, you may wish to delete them (rather than simply making them invisible) in order to free up application memory and aid system performance.

Step 1

Select a graph in the Graphics area.

Step 2

Click the **Trace Preferences** button on the Graphics toolbar or select **Trace Preferences** from the **Graph** menu. The **Trace Preferences** dialog is displayed.

Step 3

Select the trace to be removed by clicking on it.

Step 4

Click the right mouse button to display the **Trace Preferences** menu.

Step 5

Select **Delete Traces** to delete the sample from the trace list. You will be prompted to confirm this action.

Step 6

Select **OK** to clear the **Trace Preferences** dialog. The trace will be removed from all graphs.

See also:
- How to remove a trace from a graph
- How to display a trace that is not appearing in the graph box

**Graph Preferences—Scan**

Use the **Graph Preferences** dialog to change the appearance of your 2D or Contours graph.

Any changes you make to the appearance of the graph will apply to all graphs until the settings are changed again.

To access the **Graph Preferences** dialog, select the **Graph Preferences** command from the **Graph** menu, or select the **Graph Preferences** button on the Graphics toolbar.
The **Graph Preferences** dialog comprises the following tabbed pages:

- **2D Graph**
- **Contours**

See also:
- **Graph menu**
- **Graphics toolbar**
- **Graphics area**
- **Graphics How To**

## 2D Graph Preferences—Scan

Use the **2D Graph Preferences** dialog to change the appearance of your 2D graphs.

All graphs will appear labeled with the X axis label being horizontal and the Y axis vertical. You cannot change the orientation of these labels. However, you can alter the font and size of the axes and their labels. Any changes you make to the appearance of the graph will apply to all graphs until the settings are changed again.

To access the **2D Graph Preferences** page, select the **2D Graph** tab from **Graph Preferences** dialog. To access the **Graph Preferences** dialog, select the **Graph Preferences** command from the **Graph** menu, or select the **Graph Preferences** button on the Graphics toolbar.

The **2D Graph Preferences** dialog comprises the following:

### Axes Preferences

- **Axes Color**: Select the color that you want to assign to the axes of the graph/s from the drop down list.
- **Axes Width**: Select the width of the axes from the drop down list. The width, which is in pixels, ranges between one and five pixels.
- **Grid Style**: Select the style of grid for the graph from the drop down list. If you do not wish to include a grid on the graph, select **None**.
- **Plot Area Color**: Select the color to be used as the background of the graph from the drop down list. If you do not want a background color on the graph, select **White**.
- **Trace Width**: Select the width of the trace from the drop down list. The width is given in pixels and ranges between one and five pixels.
- **Font**: Click the **Font** button to display the Windows **Font** dialog where you can define the font style for the axes of the graph/s. The style and size of the font will be applied to all graph. The style, name and point size of the font appears on the font button.
- **Preview Box**: Preview any changes you make to the axes preferences in the preview box. The changes shown in this box will not be made until you select **OK**.

### Advanced Options

- **Auto tick marks**: The **Auto tick marks** option enables you to approximately set how many tick marks (or points) will be included on the graph axes. The tick marks generated will be whole, rounded, even numbers. The **Auto tick marks** option is not available if you have selected **Major ticks**.

**Y-axis**
Use the Y-axis slider bar to increase or decrease the number of tick marks (or points) to be included on the Y axis.

**X-axis**
Use the X-axis slider bar to increase or decrease the number of tick marks (or points) to be included on the X axis.

**Major ticks**
Use the **Major Ticks** option to enable you to set exactly how many ticks (or points) will be added to the X and Y axes. The number of marks you specify will be divided by the axis range, which can result in odd numbers on the axis. Note that any changes are not reflected in the graph bitmap at the top of the screen.

**Decimals**
Use the **Decimals** option to set the number of X and Y decimal places to be displayed on the axes. To set the number of decimal places, select the **Decimal** option and enter the number in the Y-axis and X-axis combo boxes. Note that any changes are not reflected in the graph bitmap at the top of the screen.

**Trace Style**

**Points**
Select **Points** to display each individual data point collected by the Cary Eclipse on the graph.

**Points and solid lines**
Select **Points and Solid Lines** to display each individual data point collected by the Cary Eclipse on the graph. Additionally, an interpolated line that connects the points will be drawn.

**Solid lines**
If you select **Solid Lines**, the Cary Eclipse system will interpolate the data between the actual, collected data points and show the data as a continuous line.

**Style cycles automatically**
If you select **Style Cycles Automatically**, the Cary Eclipse system will display each of the above three trace styles. This makes it easier to differentiate between traces, especially on black-and-white printers.

**Show Legend**

**Show Legend**
Select **Show Legend** to display a legend in the right side of each graph box. A single left click on a sample name or example line within the legend will make it the focused or red scan. Information about the trace will be displayed below the X axis.

**Hot Tip**
Double-clicking on the legend will automatically display the **Trace Preferences** window, allowing you to view more detailed information about your trace (e.g. audit log, File name, etc.).

See also:
- Graph Preferences
- Graph menu
- Graphics toolbar
- Graphics area
- Graphics How To

**Contours Graph Preferences—Scan**

Use the **Contours** page to change the appearance of your Contours graph.
Any changes you make to the appearance of the graph will apply to all graphs until the settings are changed again.

To access the Contours page, select the Contours tab from the Graph Preferences dialog. To access the Graph Preferences dialog, select the Graph Preferences command from the Graph menu, or select the Graph Preferences button on the Graphics toolbar.

The Contours page comprises the following:

### Axes Preferences

- **Axes Color**: Select the color that you want to assign to the axes of the graph/s from the drop down list.
- **Grid Style**: Select the style of grid for the graph from the drop down list. If you do not wish to include a grid on the graph, select None.
- **Plot Area Color**: Select the color to be used as the background of the graph from the drop down list. If you do not want a background color on the graph, select White.
- **Plot Axes Font**: Select the font size to be used for the plot axes. Select from Small, Medium or Large text.
- **Line colors**: If you choose to view the Contour graph as a series of lines rather than colors (by setting the Trace Style to Solid Lines), you can select the line color which will be displayed on the Contour graph. To do this, select a color from the drop down list.
- **Preview Box**: Preview any changes you make to the axes preferences in the preview box. The changes shown in this box will not be made until you select OK.

### Advanced Options

- **Major ticks**: Use the Major Ticks option to enable you to set exactly how many ticks (or points) will be added to the X and Y axes. The number of marks you specify will be divided by the axis range, which can result in odd numbers on the axis. Note that any changes are not reflected in the graph bitmap at the top of the screen.
- **Decimals**: Use the Decimals option to set the number of X and Y decimal places to be displayed on the axes. To set the number of decimal places, select the Decimal option and enter the number in the Y-axis and X-axis combo boxes. Note that any changes are not reflected in the graph bitmap at the top of the screen.

### Trace Style

- **Solid lines**: If you select Solid Lines, the Contours graph will be displayed as a series of lines. You can choose to label these lines by selecting the Data labels option.
- **Colors**: If you select Colors, the Contours graph will be displayed as a series of evenly spread colored bands. You can set the number of colors to be included, ranging from 2–200.
  - **Hot Tip**: Increasing the number of colors will increase the file size and printing time. If you are printing out Contour graphs, it is recommended that you limit the number of colors (for example, to 28).

### Legend

- **Position**: Select where to position the legend. Choose from Left, Right, Top or Bottom. Select None if you do not wish to display a legend.
Decimals

Select the number of decimal points to be included in points on the legend.

See also:
- Contour Graphs
- Contour Graph Preferences
- Graph menu
- Graphics toolbar
- Graphics area

How to alter the look of your graphs—Scan

Step 1

Click the Graph Preferences button on the Graphics toolbar, or select Graph Preferences from the Graph menu. The Graph Preferences dialog is displayed.

Step 2

Use the available options available to choose your preferred appearance for the graphs. A preview box in the top right-hand corner will demonstrate how the graphs will appear.

Step 3

Select OK to clear the Graph Preferences dialog and update all graphs to reflect your selections.

Note

You can also add labels and pictures to your graphs to alter their appearance.

Step 4

To re-use your graphics setup at a later date, save the setup as a graphics template.

See also:
- How to add a label to a graph
- How to add a picture to a graph
- How to move a label
- How to move a picture
- How to resize graph boxes
- How to make a graphics template

Cursor Modes—Scan

Use the Cursor Modes dialog to choose the type of cursor to be used on screen. For example, you can select to use a free cursor that can move anywhere in the graph.

To access the Cursor Modes dialog, select the Cursor Modes command from the Graph menu, or select the Cursor Modes button on the Graphics toolbar.

The dialog comprises the following:

Free

The Free cursor is not confined to a particular trace on the graph. It can move anywhere in a graph box, and its ‘X,Y’ position is shown in the bottom right-hand side of the Graphics area. You can use the Free cursor to zoom in on a particular part of a graph.

file://C:\Documents and Settings\cary\Local Settings\Temp~hhF1E6.htm
How to track the focused trace—Scan

To track the focused (red) trace in the graph:

Step 1

Click the Cursor Modes button from the Graphics toolbar, or select Cursor Mode from the Graph menu. The Cursor Modes dialog will appear.

Step 2

Select Track and select the OK button.

Step 3

Place the blue tracking cursor in the graph box you require.

Step 4

Move the cursor left and right across the graph box to trace the focused (red) scan. The cursor coordinates change (in the X, Y field at the bottom left hand corner of the Graphics area) as you move along the curve.

Hot Tip

If you wish to fix the cursor on a particular position on the curve, move the cursor to the required position. Click the left mouse button, press ALT on the keyboard, then release the mouse button.

See also:

How to zoom a graph
How to track the focused trace

Axes Scales—Scan

Use the Axes Scale dialog to manually change the axes range. This can assist in zooming in on a particular part of the
To access the Axes Scale dialog, select the Axes Scale command from the Graph menu, or select the Axes Scale button on the Graphics toolbar.

The dialog comprises the following:

**X Axis**

- **Mode**: View the current units for the X axis.
- **Scale**: Use the Scale field to enter a new range for the X axis, in the format n1 - n2 (where n1 is the start of the scale and n2 is the end of the scale).
  - **Hot Tip**: The Cary Eclipse will store the ten most recently entered ranges, which you can access by selecting the down arrow adjacent to the Scale field.

**Y Axis**

- **Mode**: View the current units for the X axis.
- **Scale**: Use the Scale field to enter a new range for the Y axis, in the format n1 - n2 (where n1 is the start of the scale and n2 is the end of the scale).
  - **Hot Tip**: The Cary Eclipse will store the ten most recently entered ranges, which you can access by selecting the down arrow adjacent to the Scale field.

**Z Axis**

- **Mode**: View the current units for the Z axis.
- **Scale**: Use the Scale field to enter a new range for the Y axis, in the format n1 - n2 (where n1 is the start of the scale and n2 is the end of the scale).

See also:

- How to scale your graph manually
- How to scale your graph automatically
- How to zoom a graph

**How to scale your graph manually—Scan**

**Step 1**

Click on the graph whose axes you want to change to make it the focused trace.

**Step 2**

Click the Axes Scales button on the toolbar, or select Axes Scales from the Graph menu. The Axes Scales dialog is displayed.

**Step 3**

Type in the new ranges for the X and Y axes in the format ‘n-n’, or choose an option for each from the drop-down list.

**Step 4**
Select **OK**. The axes on the selected graph will change to reflect your chosen settings.

**Hot Tip**
Select the **Zoom Out** command to resize the graph to its original settings.

**See also:**
- [How to zoom a graph](#)
- [How to scale a graph automatically](#)

**How to scale your graph automatically—Scan**

**Step 1**
Click on the graph whose axes you want to change to make it the focused trace.

**Step 2**
To autoscale the graph, select one of the following:

- Click the **Autoscale XY** button on the toolbar to autoscale both X and Y axes.
- Click the **Autoscale X** button on the toolbar to autoscale the X axis only.
- Click the **Autoscale Y** button on the toolbar to autoscale the Y axis only.

The focused trace will be scaled according to the option selected.

**Hot Tip**
Select the **Zoom Out** command to resize the graph to its original settings.

**See also:**
- [How to zoom a graph](#)
- [How to scale your graph manually](#)

**Viewing graphs—Scan**

- [Single/Multi Graph](#)
- [Individual Data and Overlay Data](#)

**Single/Multi Graph—Scan**

Select this option to toggle between a display of either one or multiple graph boxes in the Graphics area. Note that up to 100 graphs can be displayed in the Graphics area at the one time.

**Tip**
The **Single/Multi Graph** option is accessible from the **Graph** menu or the **Graphics** toolbar.
Example of a single graph box display

Example of a multi graph box display

See also:
Graph menu
Graphics toolbar
Graphics How To

**Individual Data and Overlay Data options—Scan**

Overlay Data selected
Peak Picking—Scan

With Peak Style set to 'Peaks' and Peak Threshold set to 'x', peak B is not detected. This is because the ordinate value on the right-hand side of peak B has not decreased by a minimum of 'x'.

The Peak Threshold, 'x' is reduced, but peak B is still not detected. This is because the ordinate value on the left-hand side of the peak does not decrease by a minimum of 'x'.

Peaks A and B are now detected. This is because the Peak Threshold value, 'x' is less than or equal to the decrease in the ordinate value on both sides of these peaks.

Contour Graphs—Scan

You can use the Cary Eclipse software to create contour graphs of your collected data traces. You can use contour graphs to quickly display maps of an excitation versus emission matrix for your sample of interest. Contour maps enable you to quickly identify peaks, whether you are looking at a single component with many peaks or a mixture of peaks.

Contour graphs can be viewed as lines or as bands of color. You can select the graph settings on the Contour page of the Graph Preferences dialog. You can choose to include all traces of the graph you have produced in the Contours
graph or a subset of these traces. The subsets you can choose from will depend upon the data points you have entered next to the samples in the **User Data Form**. For example, if you have included time based data points under the **Data name** of 'Time', this group of data points will be included in the **Select trace** list under the heading of 'Time'.

In the example below, the contour graph is being viewed as bands of color. With the use of the legend on the right, you can quickly identify the most intense by its color, in this case a deep red.

*An example of a contour graph, viewed as colored bands*

In the example below, the contour graph is being viewed as solid lines. In this case you would determine the most intense peak by firstly identifying the centres of contours and then comparing the intensity values for each of these centres. The centre with the highest intensity will be the highest peak.
An example of a contour graph, viewed as solid lines

To select a Contour Graph, simply click on any part of the graph (the plotting area). You can then apply many of the standard graph features available from Graphics toolbar. These include choosing a cursor mode, setting the X and Y axes scales, zooming the graph, viewing single or multiple graphs and deleting the graph. To move a contour graph, click on the graph box (the surrounding gray area), hold down your mouse and move the graph as required.

Once you have created a contour graph, you can also create XY and/or YZ slices of the contour graph at any point you select.

See also:
How to create a contour graph
How to create a slice from a contour graph
Contour Graph Preferences
Trace Preferences
User Data Form

How to create a contour graph—Scan

You can use the Cary Eclipse software to create contour graphs of your collected data traces.

To create a contour graph:

Step 1
Select an existing data trace.

Step 2
From the Graphics toolbar, select the Contour Graph button. The Trace Preferences dialog is displayed.

Step 3
In the Select Traces field of the Trace Preferences dialog, select the type of data traces you wish to display. By default, this will be set to All Traces, however you can select a subset of traces. The subsets you can choose from will depend upon the data points you have entered next to the sample names in the User Data Form. For example, if you have included time based data points under the Data name of 'Time', this group of data points will be included in the Select trace list under the heading of 'Time'.

Step 4
If you have selected All Traces, enter the Z Start and Z Interval values. These values correspond to the starting point of the graph and the interval between points.
Enter a label for the axis in the Z Axis field.

Step 5
Select OK to clear the Trace Preferences dialog. The contour graph will be displayed in the graphics area.

To select a contour graph, simply click on any part of the graph (the plotting area). You can then apply many of the standard graph features available from Graphics toolbar. These include choosing a cursor mode, setting the X and Y axes scales, zooming the graph, viewing single or multiple graphs and setting the graph preferences. To move a contour graph, click on the graph box (the surrounding gray area), hold down your mouse and move the graph as required.
How to create a slice from a contour graph—Scan

You can use the Cary Eclipse software to create a slice from a contour graph.

To create a slice of a contour graph:

Step 1
Select an existing contour graph. To do this, simply click in the plotting area of the graph.

Step 2
From the Graphics toolbar, select the **XY Slice** button. The **XY Slice** dialog is displayed.

or

From the Graphics toolbar, select the **YZ Slice** button. The **YZ Slice** dialog is displayed.

*Hot Tip:*
You can create both XY and YZ slices of the same point on the contour graph. To do this, select both the **XY Slice** button and **YZ Slice** button before selecting any points on the contour graph.

Step 3
The **XY Slice** and **YZ Slice** dialogs will prompt you to click on an area of the contour graph to create a slice. Select **OK** to clear this dialog. The Graphics display will be updated to include a blank Slice Graph.

Step 4
Click on the points of the contour graph that you want to add to the slice graph. A graph of the XY and/or YZ point will be added to the appropriate graph. You can add as many points as you like.

3D Graphs—Scan

The 3D Grams application by Galactic software® is included in the Cary Eclipse software. You can use the software to create 3D graphs of your collected data traces. 3D graphs are useful when you are analyzing an unknown sample. 3D graphs enable you to quickly produce distinct peaks of samples that may be emitting at a number of different wavelengths. Using the 3D graph, you will be able to determine the most suitable excitation and emission wavelengths for a sample.

You can choose to include all traces of the graph you have produced in the 3D graph or a subset of these traces. The subsets you can choose from will depend upon the data points you have entered next to the samples in the **User Data Form**. For example, if you have included time based data points under the **Data name** of 'Time', this group of data points will be included in the **Select trace** list under the heading of 'Time'.
An example of a 3D graph

See also:
- Trace Preferences
- User Data Form
- Contour Graphs

Examples of 3D collects—Scan

Excitation scan mode 3D example

Scan setup: Excitation scan mode

<table>
<thead>
<tr>
<th>X mode</th>
<th>nm</th>
</tr>
</thead>
<tbody>
<tr>
<td>Start</td>
<td>400</td>
</tr>
<tr>
<td>Stop</td>
<td>500</td>
</tr>
<tr>
<td>Em.</td>
<td>500 nm</td>
</tr>
</tbody>
</table>

3D mode

Em. stop = 550 nm, Ex. increment = 10 nm

The system will perform excitation scans at each of the following emission monochromator wavelengths:
Emission scan mode 3D example

Scan setup: Emission scan mode

<table>
<thead>
<tr>
<th>X mode</th>
<th>Ex.</th>
<th>300 nm</th>
</tr>
</thead>
<tbody>
<tr>
<td>Start</td>
<td>360</td>
<td></td>
</tr>
<tr>
<td>Stop</td>
<td>400</td>
<td></td>
</tr>
</tbody>
</table>

3D mode

Ex. stop = 360 nm, Ex. increment = 120 nm

The system will perform emission scans at each of the following excitation monochromator wavelengths:

Synchronous scan mode 3D example

Scan setup: Synchronous scan mode

<table>
<thead>
<tr>
<th>Start</th>
<th>Stop</th>
<th>Delta</th>
</tr>
</thead>
<tbody>
<tr>
<td>300</td>
<td>400</td>
<td>20 nm</td>
</tr>
</tbody>
</table>
3D mode

Delta stop = 40 nm, Increment = 5 nm

The system will perform five synchronous scans, using the following Delta nm values:

<table>
<thead>
<tr>
<th>Delta</th>
<th>Ex. monochromator: 300–400 nm</th>
<th>Em. monochromator: 320–420 nm</th>
</tr>
</thead>
<tbody>
<tr>
<td>20 nm</td>
<td>Ex. monochromator: 300–400 nm</td>
<td>Em. monochromator: 320–420 nm</td>
</tr>
<tr>
<td>25 nm</td>
<td>Ex. monochromator: 300–400 nm</td>
<td>Em. monochromator: 325–425 nm</td>
</tr>
<tr>
<td>30 nm</td>
<td>Ex. monochromator: 300–400 nm</td>
<td>Em. monochromator: 330–430 nm</td>
</tr>
<tr>
<td>35 nm</td>
<td>Ex. monochromator: 300–400 nm</td>
<td>Em. monochromator: 335–435 nm</td>
</tr>
<tr>
<td>40 nm</td>
<td>Ex. monochromator: 300–400 nm</td>
<td>Em. monochromator: 340–440 nm</td>
</tr>
</tbody>
</table>

See also:
Cary
Example of a synchronous scan

Example of a Graphics Template—Scan

By using the Add Label command in the Cary Eclipse graphics system, you can customize a graphics template such as the one shown below:
How to make a graphics template—Scan

A graphics template lets you set up and store the style of a graphics report for future use. You can do this by adding labels and pictures to the background of the Graphics area and altering any axes styles etc. One advantage of this over the application's built-in reporting features is that you have more flexibility as to where you can position graph boxes and any text that you may want to appear (e.g. operator name, batch number etc.) and also where you add pictures, e.g. company logos etc.

Step 1

Start the application and open the data file from which you want to make a graphics template. Alternatively, collect some new data.

Step 2

Make as many graph boxes as you require in your template by using the Add Graph function.

Step 3

Adjust the graph boxes to the correct size and position them as you want them to appear in the template.

Step 4

Do any of the following, if required:

- Change the appearance of the graph boxes in the Graph Preferences dialog
- Use the **Add Label** function to append text to the Graphics area
- Use the **Add Picture** function to add pictures, such as your company's logo to the Graphics area

**Step 5**

From the **File** menu, select **Print Preview**. Use the **Print Preview** dialog displayed to check the template that you have created.

**Step 6**

When you are satisfied with the appearance of the graphics, select the **Save As...** command from the **File** menu. The **Save As** dialog is displayed.

**Step 7**

A list of stored data files will appear. Select the down arrow at the right of the **Files of type** field and select 'Graph Template' to list all the template files. Ensure that the **Save only focused trace** check box is cleared.

**Step 8**

Enter a name for the template in the **File name** field.

**Step 9**

Select **OK** to save the graphic template. The template will be used each time you create a new file.

**Note**

You can also make a graphic template in a similar manner from existing data. No data will be saved in the template.

**See also:**
- Example of a Graphics Template
- How to add a label to a graph
- How to add a picture to a graph
- How to move a label
- How to move a picture

**Examples—Scan**

- Example of an emission scan
- Example of an excitation scan
- Example of a synchronous scan
- Examples of 3D collects

**Example of an Excitation Spectrum—Scan**

Select **Emission** to perform an emission scan. During the scan, the excitation monochromator is set to a fixed wavelength and the emission monochromator is scanned over a wavelength range. This results in a fluorescence excitation spectrum.

The excitation spectrum is a plot of intensity at a particular emission wavelength against the wavelengths of the exciting light. When collecting an excitation spectrum you need to determine where you have to excite the system to see fluorescence at the chosen emission wavelength.

The **Emission** selection is set on the **Cary** page. The following is an example of an Excitation Spectrum.
Example of an Emission Spectrum—Scan

Select **Excitation** to perform an excitation scan, which is a qualitative absorption spectrum. During the scan, the emission monochromator is set to a fixed wavelength and the excitation monochromator is scanned over a wavelength range. This is often referred to as the emission spectrum or the fluorescence spectrum.

Emission spectrums are characteristic of molecular structure and environment of material. The shape of the fluorescence spectrum is independent of the excitation wavelength because the emission always originates from the lowest excited state. The spectrum is often a “mirror image” of the absorption spectrum.

The **Excitation** selection is set on the **Cary** page. The following is an example of an emission spectrum.

See also:
- Example of an Emission Spectrum
- Cary page
Example of a synchronous scan—Scan

If you select to perform a Synchronous scan, the excitation and emission monochromators will be scanned synchronously with a fixed delta (which is a real number and may be zero), between both monochromators. The Synchronous Scan mode can be used to resolve multi-component mixtures.

This option is available from the Cary page of the Scan Application if you select Fluorescence or Phosphorescence as the Data Mode.

Flowchart of a Synchronous Scan
Example using a Fluorescein and RhB mixture

The emission scan of the mixture of Fluorescein and RhB

The separated scans of Fluorescin (red line) and RhB (blue line)
A synchronous scan of Fluorescein and RhB

See also:
- Cary page
- Examples of Excitation, Emission and Synchronous 3D collects

Example of a Corrected Spectra—Scan

The Correction Curves command is selected from the Commands menu. The following is an example of uncorrected and corrected spectra.

File Management—Scan

- File menu
- Open dialog
- Save As dialog
File menu—Scan

Use the File menu to open or save a method or data file, print or preview a report, or exit the Cary Eclipse application.

**Hot Tip**
Highlight a method or data file in Explorer and simply drag and drop it onto the application to open it. Any data will be displayed in the Graphics area.

The following commands can be accessed from the File menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>New</td>
<td>Select <strong>New</strong> to clear all method information and report information and create a new file containing the default settings for the application. The <a href="#">Save As dialog</a> will be displayed for you to enter the name of the new file and select a location.</td>
</tr>
<tr>
<td>Open Method…</td>
<td>Select <strong>Open Method</strong> to display the <a href="#">Open dialog</a> where you can search for and load a stored method.</td>
</tr>
<tr>
<td>Open…</td>
<td>Select <strong>Open</strong> to display the <a href="#">Open dialog</a> where you can search for and load method, data, report, graph template and batch files, as well as other file types compatible with the Cary Eclipse software. When you load a batch file, the method which collected the data is loaded as well as the associated report and graphics layout.</td>
</tr>
<tr>
<td>Save Method As…</td>
<td>Select this option to display the <a href="#">Save As dialog</a> and store the current method. You can use the Save As dialog to store the method, data, report, or graphics template separately or together as a Batch file. The data can also be saved as a comma delimited ASCII Spreadsheet file.</td>
</tr>
<tr>
<td>Save As…</td>
<td>Select this option to display the <a href="#">Save As dialog</a> and store the collected data. You can use the Save As dialog to store the method, data, report, or graphics template separately or together as a Batch file. The data can also be saved as a comma delimited ASCII file.</td>
</tr>
<tr>
<td>Print Setup…</td>
<td>Select this option to display the Windows Print Setup dialog where you can select the printer, printer page size and orientation.</td>
</tr>
<tr>
<td>Print Preview…</td>
<td>Select this option to display the <a href="#">Print Preview dialog</a> where you can view the current report as it will be printed.</td>
</tr>
<tr>
<td>Print…</td>
<td>Select this option to display the Windows Print dialog where you can choose the printer and pages to print as well as the number of copies you wish to print.</td>
</tr>
<tr>
<td>Explorer…</td>
<td>Select this option to run Windows Explorer. This may help you locate method and data files.</td>
</tr>
<tr>
<td>History List</td>
<td>This displays a list of the four most recently opened files.</td>
</tr>
<tr>
<td>Exit</td>
<td>Select this option to exit the Scan application.</td>
</tr>
</tbody>
</table>

See also:
- [Filename Extensions](#)
- [ASCII Spreadsheet File](#)

Open—Scan

Use this dialog to open a file previously saved by the application. To access the Open dialog, select **Open** or **Open**.
Method from the File menu.

Look in
The Look In field lists the available folders and files. Click the down arrow on the right of this field to view the location of the current folder in your computer's hierarchy. You can also select a new location simply by selecting a different folder. The folders and files in the selected folder are displayed the list box below the Look In field.

Note
To open the folder in the level that is one higher than the current folder, select on the toolbar.

Look in list box
The Look In List Box, which is directly below the Look In field, displays the folders and files in the selected location. To view the contents of a folder, double-click it.

- Click to create a new folder. Type the name of the new folder in the highlighted text entry field that appears and press Enter. Then double-click on the new folder to open it.
- Click to display the directory structure of the drive.
- Click to display the files as a list.
- Click to display files as a detailed list including the file size, file type and the date the file was last modified.
- Click to preview the selected trace before you load it into the application.

File name
Enter the name of the file you want to open in this field. The easiest way to do this is to find the file in the Look in list box and then select the file. However, you can type the full path of a file and then select Open to open the file.

Files of type
Click on the arrow on the right of this field to view a list of the different types of application files that can be opened in the list box.

- **Methods (*.FMSW)** Choose this file type to open a Method file. Only the method is loaded. No data is loaded.
- **Data (*.FDSW)** Choose this file type to open a Data file. Only the data is loaded. The method that was used to collect the data is not loaded. The last used method or default method will be displayed.
- **Report (*.FRSW)** Choose this file type to open a Report file. You can view the report in the Report area.
- **Graph Template (*.FGSW)** Choose this file to open a Graph Template file. The file will set up the layout of the Graphics area including any bitmaps as well as the range and style of the graph axes.
- **Batch (*.FBSW)** Choose this file type to open a Batch file. A Batch file includes data, the method used to collect the data, report and graphics setup.
- **Spreadsheet Ascii (*.CSV)** This will open ASCII files that contain single comma delimited files with two or more columns.
- **GRAMS (*.SPC)** Choose this file type to open a Cary Eclipse GRAMS data file. This allows you to use data files collected using a Cary Eclipse GRAMS system. The method that was used to collect the data is not loaded.
- **ADL (*.ADL)** This will open and run program files generated in the Applications Development Language (ADL).
- **All files (*.*)** This enables you to view all files in the directory so that you can open any compatible file.
**Overlay Data**  Select this check box to overlay individual data files in one graph box. If this is not checked the data from each file will appear in individual graph boxes. This option is only available if the Open dialog has been accessed by selecting 'Open...' from the File menu.

![Hot Tip]
Use the SHIFT or Control key to select multiple data files.

**Open**  This button opens the highlighted file/s or the files listed in the File name field.

See also:  
[File menu](#)  
[ASCII Spreadsheet File](#)

**Save As—Scan**

Use the Save As dialog to save the data, method, report or graphics template individually or together as a Batch file.

**Note**  This dialog appears when you select either the Save Method As... or Save Data As... menu option in the File menu. When you select Save Method As... this dialog appears with 'Methods' displayed in the Files of type field. When you select Save Data As... this dialog appears with 'Batch' displayed in the Files of type field.

Traces (or data) that are currently ‘Visible’ in the Graphics area are saved, regardless of how they are displayed (full screen graph box or tile).

For example, if six graph boxes are displayed in the Graphics area and each graph box has one Visible trace then all six traces will be saved.

If you do not wish to save particular traces, make them not Visible in the Trace Preferences dialog. To save only the focused trace, select the **Save only focused trace** check box.

The Save As dialog comprises the following:

**Look In**  The Look In field lists the available folders and files. Click the down arrow on the right of this field to view the location of the current folder in your computer's hierarchy. You can also select a new location simply by selecting a different folder.

The folders and files in the selected folder are displayed the list box below the Look In field.

![Note]
To open the folder in the level that is one higher than the current folder, select on the toolbar.

**Look in list box**  The Look In List Box, which is directly below the Look In field, displays the folders and files in the selected location. To view the contents of a folder, double-click it.

- Click to create a new folder. Type the name of the new folder in the highlighted text entry field that appears and press Enter. Then double-click on the new folder to open it.
- Click to display the directory structure of the drive.
- Click to display the files as a list.
- Click to display files as a detailed list including the file size, file type and the date the file was last modified.
- Click to preview the selected trace before you load it into the application.
Filename extensions—Scan

The Cary Eclipse system uses the filename extension to distinguish between data types (e.g. method or report) and also to differentiate applications. (e.g. Concentration or Kinetics).

The Syntax for file extensions is:

<table>
<thead>
<tr>
<th>Letter</th>
<th>Represents</th>
<th>Available Options</th>
</tr>
</thead>
<tbody>
<tr>
<td>First</td>
<td>Type of analysis</td>
<td>F Fluorescence</td>
</tr>
</tbody>
</table>
| Second | Type of file | B Batch  
          |              | D Data    
          |              | G Graph Template |
The following files may be opened and saved in the Scan Application:

**Note**
Selecting *All Files* in the Windows Open dialog will list all available file types.

<table>
<thead>
<tr>
<th>Extension</th>
<th>Available action</th>
<th>File type</th>
</tr>
</thead>
<tbody>
<tr>
<td>*.FMSW</td>
<td>Open/Save</td>
<td>A <em>method</em> file (*.FMSW) consists of the software setup by which data is collected.</td>
</tr>
<tr>
<td>*.FDSW</td>
<td>Open/Save</td>
<td>A <em>data</em> file (*.FDSW) contains data collected that is currently visible in the Trace Preferences dialog (for any graph box).</td>
</tr>
<tr>
<td>*.FRSW</td>
<td>Open/Save</td>
<td>A <em>report</em> file (*.FRSW) contains the report generated by the Cary Eclipse about the data.</td>
</tr>
<tr>
<td>*.FGSW</td>
<td>Open/Save</td>
<td>A <em>graph template</em> file (*.FGSW) contains information that sets up the layout of the Graphics area including any bitmaps as well as the range and style of the graph axes.</td>
</tr>
<tr>
<td>*.FBSW</td>
<td>Open/Save</td>
<td>A <em>batch file</em> (*.FBSW) contains the collected data that was Visible in the Trace Preferences dialog (for any graph box), the method by which the data was collected and the report and graphics template.</td>
</tr>
<tr>
<td>*.CSV</td>
<td>Open/Save</td>
<td>An <em>ASCII spreadsheet</em> file (*.CSV) has the same extension for all applications. It consists of raw intensity data (that was currently Visible in the Trace Preferences dialog (for any graph box) saved as XY comma-delimited pairs. This file can be imported into a spreadsheet program such as Microsoft Excel.</td>
</tr>
<tr>
<td>Data.*</td>
<td>Open</td>
<td>A <em>Cary DOS</em> (Data.*) file is the equivalent of a Cary Windows Data file—it does not contain any Batch information.</td>
</tr>
<tr>
<td>*.ADL</td>
<td>Open</td>
<td>An <em>ADL</em> file (*.ADL) is a program file generated in the Applications Development Language (ADL).</td>
</tr>
<tr>
<td>*.SPC</td>
<td>Open/Save</td>
<td>A <em>Cary GRAMS</em> file (*.SPC) is the equivalent of a Cary Eclipse Windows Data file—it does not contain any Batch information.</td>
</tr>
<tr>
<td>*.FADL</td>
<td>Open</td>
<td>An <em>Eclipse ADL</em> (*.FADL) file is a program file generated in the Applications Development Language (ADL) specifically for the Cary Eclipse. Eclipse ADL files will be associated with the relevant Eclipse application. When creating ADL files for the Cary Eclipse, it is recommended that you save them with the FADL extension.</td>
</tr>
</tbody>
</table>
An ASCII Spreadsheet file consists of raw intensity data saved as XY comma-delimited pairs. You can choose to import or export Cary Eclipse files to ASCII files, which can then be imported in spreadsheet programs such as Microsoft Excel.

The format is as follows:

- Line 1 contains the sample name plus a comma
- Line 2 contains X and Y labels. The name of the X label must be the same as that used to label the graph. For example, in Scan, the graph label is Wavelength (nm).
- Lines 3 and onward contain the X,Y data
- After the last line of X,Y pairs there must be a blank line separating the data from any text audit strings or the end of the file.

For example, if a scan file was collected as:

<table>
<thead>
<tr>
<th>nm</th>
<th>intensity</th>
</tr>
</thead>
<tbody>
<tr>
<td>700</td>
<td>0.1392</td>
</tr>
<tr>
<td>698</td>
<td>0.1388</td>
</tr>
<tr>
<td>696</td>
<td>0.1245</td>
</tr>
</tbody>
</table>

etc.

It would appear as follows in the ASCII file:

```
Scan,
Wavelength (nm); Intensity (a.u.)
700, 0.1392,
698, 0.1388,
696, 0.1245,
```

Microsoft’s Excel generates a CSV file with the above format.

There is also an alternative format for CSV files. For example,

```
Scan
Wavelength (nm); Intensity (a.u.)
700,0.1392
698,0.1388
696,0.1245
```

See also: Open, Save As

How to export collected data—Scan
If you want to export data from a Cary Eclipse application into a third-party application, then you simply need to save your data as an Spreadsheet ASCII (*.CSV) file or a Rich Text Format (*.RTF) file. Spreadsheet ASCII files are recommended if you are planning to use the data in a spreadsheet program such as Microsoft® Excel. Rich Text Format files are recommended if you are planning to open the data in a word processor program such as Microsoft® Word.

To export collected data:

Step 1

From the File menu, select Save As. The Save As dialog is displayed.

Step 2

Click on the down arrow next to the Files of type list box and select the Spreadsheet ASCII (*.CSV) or Rich Text Format (*.RTF) option.

Step 4

Type the name of your new file in the File name field.

Step 5

Select Save to export your data to this new file.

**How to combine data files into a batch file—Scan**

To combine a number of data files into a batch file, do the following:

Step 1

Select Open… from the File menu to display the Windows Open dialog.

Step 2

A list of stored batch files will appear. Select the down arrow to the right of the Files of type field and select 'Data' to list all the data files.

Step 3

Select the Overlay Data check box.

Step 4

Highlight the data files that you require. Hold down the CTRL key to select multiple files.

Step 5

Select Open. The highlighted data files will load into the application and appear in the same graph box.

Step 6

Select Save As… from the File menu to display the Windows Save As dialog.
A list of stored data files will appear. Select the down arrow at the right of the **Files of type** field and select 'Batch' to list all the batch files. Ensure that the **Save only focused trace** check box is cleared.

**Step 8**

In the **File name** field, type in the file name for your new batch file.

**Step 9**

Select **Save** to create the new batch file. The current method will be stored with the batch file.

All the data files are now combined into the one batch file.

**Software and Screen features—Scan**

- **Menu line**
- **Status display**
- **Status line**
- **Buttons**
- **Function keys**
- **Right mouse menu**
- **How to remove or display the command buttons**

**Menu line—Scan**

- **File menu**
- **Edit menu**
- **View menu**
- **Commands menu**
- **Setup menu**
- **Graph menu**
- **Help menu**

**Status line—Scan**

The Status Line is displayed at the very bottom of the **Scan Application window**. The line will display the following information (from left to right):

- **Hardware Status**
- **Sequence Information**
- **Current Results**

**Buttons—Scan**

The major commands in this application can be accessed both from the menu items at the top of the main window and from the application buttons. These buttons, which appear in the left side of the Scan Application window, comprise the following:

- **Hot Tip**

  If you wish to access these commands from the main menus, rather than from these buttons, you can remove the Command buttons from your application window. To do this, clear the **Buttons** command in the **View** menu. This will enlarge the Graphics area, making viewing and selection of scans easier.
<table>
<thead>
<tr>
<th>Button</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Setup</strong></td>
<td>Click this button to display the <strong>Setup</strong> dialog where you can set up the collection, storage and analysis parameters for the scan. You can also set up any accessories that you may have and define the reporting options.</td>
</tr>
<tr>
<td><strong>Zero</strong></td>
<td>Select this to zero the current ordinate value. This command can also be accessed by pressing F5.</td>
</tr>
<tr>
<td><strong>Note</strong></td>
<td>Before you select <strong>Zero</strong>, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.</td>
</tr>
<tr>
<td><strong>Pre-scan</strong></td>
<td>Select this to determine the optimum instrument parameters to be used for your sample. At the end of this collection sequence the software will report the optimum Ex and Em wavelengths and PMT voltage. Once you are satisfied with the parameters, the software will automatically update your method setup to reflect these new parameters.</td>
</tr>
<tr>
<td><strong>Clear Report</strong></td>
<td>Select <strong>Clear Report</strong> to remove all of the text that appears in the Report area. If you have saved the report, you can view the information again. To do this, simply open the report file using the <strong>Open…</strong> command in the <strong>File</strong> menu.</td>
</tr>
<tr>
<td><strong>Create report</strong></td>
<td>Select <strong>Create Report</strong> to display the <strong>Reports</strong> page of the <strong>Setup</strong> dialog. You can use this page to enter and select information that will appear in the report.</td>
</tr>
<tr>
<td><strong>Print</strong></td>
<td>Select <strong>Print</strong> to display the Windows <strong>Print</strong> dialog where you can choose what printer the report will be printed to. You can also select what pages to print as well as the number of copies you wish to print. This command can also be accessed by pressing CTRL+P.</td>
</tr>
<tr>
<td><strong>Note</strong></td>
<td>The options on the <strong>Reports</strong> page are used to generate the contents of the Report area during a run or on a recalculation. If <strong>Auto Print</strong> has been selected the Cary Eclipse will automatically print a report at the end of a data collection.</td>
</tr>
</tbody>
</table>

---

See also:
- Commands menu
- Setup dialog
- Reports page

**Function keys—Scan**

The function keys at the top of your keyboard perform basic Cary Eclipse operations simply and quickly.

<table>
<thead>
<tr>
<th>Function key</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>F1</strong></td>
<td>This displays the Help for the current dialog or window.</td>
</tr>
<tr>
<td><strong>F4</strong></td>
<td>Press F4 to display the <strong>Go To Wavelength</strong> dialog.</td>
</tr>
<tr>
<td><strong>F5</strong></td>
<td>Press F5 to perform an instrument <strong>Zero</strong>. The current intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the <strong>Zero</strong> command before selecting <strong>Start</strong> if you want your data to start at 0 intensity. This command is also accessible from the <strong>Commands</strong> menu or by selecting the <strong>Zero</strong> button.</td>
</tr>
<tr>
<td><strong>Note</strong></td>
<td>Before you select <strong>Zero</strong>, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.</td>
</tr>
<tr>
<td></td>
<td>Select the <strong>Clear Zero</strong> command from the <strong>Commands</strong> menu to remove zero correction.</td>
</tr>
<tr>
<td><strong>F6</strong></td>
<td>Press F6 to display the <strong>Rapid Result</strong> dialog where you can read a blank and multiple sample readings</td>
</tr>
</tbody>
</table>
The following commands are accessible by clicking the right mouse button on editable fields in the Cary Eclipse software.

- **Undo**
  - If available, this reverts to the previous value before the Edit on the field.

- **Cut**
  - If available, this cuts the highlighted text to the clipboard.

- **Copy**
  - If available, this copies the highlighted text to the clipboard.

- **Paste**
  - If available, this pastes the text from the clipboard to the cursor position.

- **Delete**
  - If available, this deletes the highlighted text/number.

- **Select All**
  - If available, this selects all entries.

### How to remove or display the command buttons—Scan

The command buttons (Setup, Zero, Pre-scan, Clear Report, Create Report and Print) appear on the left side of the Scan Application window.

**To remove the command buttons**

1. From the Scan Application window, select the *View* menu.

2. Clear the *Buttons* menu item.

   The buttons will disappear from the application screen, giving you more room to display reports.

**To show the command buttons**

1. From the Scan Application window, select the *View* menu.

2. Without changing the application method, this command is also accessible from the *Commands* menu.
Select the **Buttons** menu item.

The buttons will re-appear in the application screen.

## ADL—Scan

- [ADL Command Line](#)
- [ADL Hooks](#)
- [How to insert ADL commands](#)

### ADL Command Line—Scan

#### Introduction to ADL

Use the ADL Command Line to run Single-line or Multi-line Applications Development Language (ADL) commands that enable you to perform functions over and above that of the existing program functionality.

ADL is not a general purpose programming language. It has been developed specifically by Varian to meet the needs of spectroscopists. It can be used to handle all the data collected from the Cary Eclipse—both photometric values and entire data collections. ADL programs that you develop should be stored with the extension 'ADL'.

To access the ADL Command Line, select the ADL Command item from the **Commands** menu. You can also press ALT+Y to open the dialog.

**Note**

Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.

### Software controls

**Single Line**  
Selecting this lets you enter ADL commands on a single line into the Command Line entry box.

- **Q.** Can I enter more than one ADL command on a single line?  
  - **A.** Yes, provided that the commands are separated by a semicolon ‘;’.

**Multi-line**  
The Multi-line capability enlarges the Command Line entry box. Use the Multi-line capability to create a more complex ADL program by adding a series of commands over multiple lines. When Multi-line mode is selected, the Quick Reference option becomes available.

**Quick Reference**  
Select Quick Reference to open the Quick Reference dialog where you can easily add a command in the correct syntax to your ADL program. This option is only available for Multi-line commands.

- ![Show me How to insert ADL commands using the Quick Reference.](#)

A detailed description of all the commands in the Quick Reference is available by selecting the Help button on the Quick Reference dialog.

**Browse**  
Select **Browse** to access the Open dialog where you can browse for and select an existing ADL program to load and run.

**Command Line (entry box)**  
This entry box enables you to use ADL commands. You can enter single ADL commands into this field, or you can enter a series of ADL commands. If you have the entry type set to Single Line, you can review previous ADL entries by selecting the down arrow to the right of the list box.
The ADL commands in the Command Line are executed when you select OK.

**Note**
On entering this window, the Cary Eclipse will recall the last command or program executed from the Command Line entry box. The ADL commands entered in the last executed program are typically stored in the Varian\Cary Eclipse WinFLR\ADL directory on the drive on which the Cary Eclipse software is installed.

**Hot Tip**
You can rerun the ADL commands in the Command Line entry box from any screen in the Scan application by pressing ALT-X or selecting Re-run ADL from the Commands menu.

See also:
ADL Hooks

### ADL Hooks—Scan

The ADL Hooks command is accessible from the View menu.

Instead of writing a full ADL program you can use ADL hooks to link into an existing application. This is particularly useful if you want to make small changes to the way an application works, e.g. you want to pop up a dialog for the user to enter some specific data before the run starts. ADL hooks are links at specific points in an application at which you can run an ADL program. For example, ADL hooks have been supplied to run the following ADL programs:

**Note**
Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.

<table>
<thead>
<tr>
<th>Hook</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>SW_Startup.ADL</td>
<td>This is run when the Scan application starts. For this to to work the application must be started from a batch or method file that has ADL Hooks on.</td>
</tr>
<tr>
<td>SW_PreSeq.ADL</td>
<td>This runs when the Start button is selected.</td>
</tr>
<tr>
<td>SW_PostSeq.ADL</td>
<td>This runs at the end of the Start button sequence.</td>
</tr>
<tr>
<td>SW_Online.ADL</td>
<td>This runs when Cary Eclipse is detected online.</td>
</tr>
<tr>
<td>SW_OffLine.ADL</td>
<td>This runs when Cary Eclipse goes offline.</td>
</tr>
<tr>
<td>SW_Zero.ADL</td>
<td>This runs when the Zero button is selected.</td>
</tr>
<tr>
<td>SW_Recalc.ADL</td>
<td>This runs when the Recalculate button is selected.</td>
</tr>
<tr>
<td>SW_Shutdown.ADL</td>
<td>This runs when the application is shutting down.</td>
</tr>
<tr>
<td>SW_PreRead.ADL</td>
<td>This runs just before a read is taken.</td>
</tr>
<tr>
<td>SW_PostRead.ADL</td>
<td>This runs just after a read is taken (It has the reading result stored in the UserResult variable.)</td>
</tr>
<tr>
<td>SW_PreSetup.ADL</td>
<td>This runs when the Setup button is selected.</td>
</tr>
<tr>
<td>SW_PostSetup.ADL</td>
<td>This runs when the OK button on the Setup dialog is selected.</td>
</tr>
<tr>
<td>SW_CellChange.ADL</td>
<td>This runs instead of the Cary Eclipse driving the Multicell Holder. It enables you to add your own instructions for the Cary Eclipse Multicell Holder or to use your own cell changing accessory.</td>
</tr>
</tbody>
</table>
How to insert ADL commands using the Quick Reference—Scan

The Quick Reference window is accessed from the ADL Command window. It enables you to easily add a command in the correct syntax to your ADL program. To do this:

Step 1

Press Alt-Y to open the ADL Command Line window.

Step 2

Select the Multi-line option. The Command Line entry box will expand to take multiple lines of ADL commands and the Quick Reference check box is displayed.

Step 3

Select the Quick Reference check box. The Quick Reference window is displayed.

Step 4

If necessary, use the Go To field to jump to the appropriate part of the Quick Reference listing.

Step 5

Highlight the command you require in the Quick Reference list.

Step 6

Click the Insert in ADL button. The selected ADL command will appear in the Command Line entry box.

Step 7

Where necessary, alter the defaults supplied by the inserted Quick Reference command in the Command Line entry box to suit your program.

Step 8

Repeat steps 4–7 to add more commands as required.
Help and Safety information—Scan

- Help menu
- Safety home page

Help menu—Scan

Use the Help menu to view the online Help and software information for this application.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Scan Help</td>
<td>Select this to display the Scan Home Page. This contains a list of online</td>
</tr>
<tr>
<td></td>
<td>Help topics specific to the Scan Application.</td>
</tr>
<tr>
<td>Help Topics</td>
<td>Select this to display the Cary Eclipse Windows Online Help Home Page.</td>
</tr>
<tr>
<td>Tip of the day</td>
<td>View useful tips about using your Cary Eclipse instrument and software.</td>
</tr>
<tr>
<td>Eclipse Tour</td>
<td>Select this to display the Cary Eclipse Multimedia Online Help. This will</td>
</tr>
<tr>
<td></td>
<td>give you an animated introduction to the Cary Eclipse software and hardware.</td>
</tr>
<tr>
<td>About…</td>
<td>Select About to access a dialog that displays information about the application as well as Varian email and Internet contact information. To clear this About dialog, select OK.</td>
</tr>
</tbody>
</table>

Safety Home Page

Click on the desired index entry to view the Help.

- About Safety
- Lamp module
- Ultraviolet radiation
- Electrical hazards — Cary Eclipse
- Panels, covers and modules
- Other precautions
- Warnings and cautions
- Warning symbols
- Color coding
- Information symbols
- FCC advisory
- CE compliant products

Scan How To—Scan

Select the procedure you require below for a step-by-step informative guide on running the application software.

Setup, calculations and reports

- How to perform a fluorescence scan
- How to perform a fluorescence scan in cycle mode
- How to perform a fluorescence scan in signal-to-noise mode
- How to perform a phosphorescence scan
- How to perform a phosphorescence scan in cycle mode
How to perform a phosphorescence scan in signal-to-noise mode
How to perform a bio-/chemi-luminescence scan
How to perform a bio-/chemi-luminescence scan in cycle mode
How to perform a bio-/chemi-luminescence scan in signal-to-noise mode
How to use advanced reporting options
How to generate a report from previously collected data

Other

How to email your results
How to export collected data
How to combine data files into a batch file
How to import sample names
How to remove or display the command buttons
How to insert ADL commands using the Quick Reference

See also:
Scan Application Home Page
Graphics How To

How to perform a fluorescence scan in cycle mode—Scan

This demonstrates how to perform a fluorescence wavelength scan in cycle mode.

- Start the Scan application
- Set up data collection parameters
- Set up instrument parameters such as Data Mode and Scan Setup
- Set up visual system monitoring
- Set up the cycle mode and filter options
- Make sure that no accessories are selected
- Set up reporting and printing requirements
- Set up storage of collected data before run
- Set up email options
- Finish Setup
- Zero the instrument
- Start the Scan run
- Save the collected data

Start the Scan application

Step 1

Click the Windows Start button, point to Programs, Cary Eclipse and then select Scan. The Scan Application is displayed.

Note
If you are running a GLP system you may be prompted to enter a password before accessing the Scan Application.

Set up data collection parameters

Step 2 [Setup dialog]

Select the Setup button to display the Setup dialog and specify the method parameters for a new method.

Set up instrument parameters such as Data Mode and Scan Setup
Step 3 [Setup dialog | Cary page]

a. Set the **Data mode** to **Fluorescence**.

b. Set the **Scan setup** mode to **Emission**. During the emission scan, the excitation monochromator is set to a fixed wavelength and the emission monochromator is scanned over a wavelength range.

c. Set the **X Mode** to **Wavelength (nm)**.

d. Enter an **Excitation (nm)** value that is within the region where the fluorescent molecule to be scanned will absorb light. For purposes of this example, Anthracene will be used as an example and therefore a value of **360 nm** should be set.

e. Enter an **Excitation slit (nm)** value or 5 and an **Emission slit (nm)** value of 5. Slits determine the resolution of the spectrum and therefore are used in conjunction with the **PMT Detector Voltage** (set on the **Options** page) to determine concentration. If a compound is highly fluorescent and has reasonable signal intensity (as is the case for Anthracene) the slits can be set quite narrow. If

f. Enter a **Start (nm)** value of **370 nm** and enter a **Stop (nm)** value of **520 nm**. The **Start (nm)** value should be set to the **Excitation (nm)** value plus the sum of the slits. In this example, as the **Excitation** value is **360 nm**, and the sum of the slits is **10 nm**, the **Start (nm)** value is set to **370**.

Typically the **Stop (nm)** value should be set to **150–200 nm** greater than the **Start (nm)** value.

g. If selected, clear the **3-D Mode** check box.

**Set up visual system monitoring**

Step 4 [Setup dialog | Cary page]

Select the **Status Display** check box so that you can view various instrument parameters during the scan.

**Set up the cycle mode and filter options**

Step 5 [Setup dialog | Options page]

a. Select the **Options** tab to display the **Options** page.

b. Select the **Cycle mode** option. This option enables you to collect scans as a function of time to determine whether the spectrum is changing. (By contrast, the Kinetics application enables you to determine whether a single wavelength is changing over time.) If this option is not available, clear the **CAT or S/N mode** check box.

c. Enter a **No. of scans** value of **15** to set the number of scans to be performed per cell/well.

d. Enter a **Time** value of **1.0000** to set the interval between each successive scan. If the time entered is less than the time required to complete a scan, the scans are performed continuously with no delay.

e. Set the **Excitation filter** to **Auto** and the **Emission filter** to **Open**.

f. Set the **PMT Detector voltage** to **Medium**. This setting can later be adjusted if results are over-range. If the signal is too high, decrease the PMT Detector Voltage. If the signal is too low, increase the PMT Detector Voltage.

**Make sure that no accessories are selected**

Step 6 [Setup dialog | Accessories page]
Make sure that no options are selected on any of the tabs of the **Accessories** page.

**Set up reporting and printing requirements**

Step 7 [Setup dialog | Reports page]

a. Select the **Reports** tab to display the **Reports** page where you can specify your reporting requirements for this method.

   i. Enter your name in the **Name** entry field.
   ii. If required, enter any comments relating to your experiment in the **Comment** field.

   iii. Set up your report style by selecting the appropriate check boxes in the **Options** group.

   For example, select the **Auto print** check box to obtain a printout of your report automatically. Select the **Parameters** check box to include your experimental parameters in the report. Select the **Graph** check box to include a graph in the generated report.

   ![](http://C:\Documents and Settings\cary\Local Settings\Temp\~hhF1E6.htm)

   **Note**
   If **Auto print** is selected, the system will send the report information to the specified printer as well as displaying it in the **Report** area. However if **Auto print** is not selected, the report will be displayed in the area only. The **Report** area can be viewed by selecting the **Report** command from the **View** menu.

b. Set up the **Peaks** reporting options.

   i. Select **Maximum peaks** to report the peak with the largest peak threshold that exceeds the peak **Threshold** value.

   ii. Select **All peaks** to report all peaks meeting the **Peak type** criterion and exceeding the **Threshold** value.

   iii. Select the **Peak type** and specify the peak **Threshold**.

c. If required, select the **X-Y Pairs table** check box. You can use the **Actual Data interval** by which the data was collected or you can make the Cary Eclipse Interpolate the points to a new interval.

**Set up storage of collected data before run**

Step 8 [Setup dialog | Auto Store page]

a. Select the **Auto Store** tab to display the **Auto store** page. Use this page to set up whether the collected data is to be saved, and if so, when the Cary Eclipse should store the information.

b. Select **Storage option of On; prompt at end**.

c. Select the **Auto convert** option you require.

   If you select Select for ASCII (csv) or Select for ASCII (csv) with Log, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format in the current folder.

**Set up email options**

Step 9

Refer to [How to email your results](http://C:\Documents and Settings\cary\Local Settings\Temp\~hhF1E6.htm) for instructions on setting up the email options.

**Finish Setup**
Step 10

Once you are satisfied with your method setup select **OK** to confirm any changes you have made and close the **Setup** dialog.

**Zero the instrument**

Step 11

a. Place the blank solution in the sample compartment of the Cary Eclipse instrument.

b. Click the **Zero** button to zero the system. Alternatively, select **Zero** from the **Commands** menu to perform a zero. When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the Scan Application window.

**Start the Scan run**

Step 12

a. Select the **Start** button to commence a data collection. Alternatively, select **Start** from the **Commands** menu. The **Sample name** dialog is displayed.

b. In the **Sample name** dialog, enter the appropriate name for your sample and select **OK**. The **Sync Start** dialog is displayed.

c. The **Sync Start** dialog displays a two minute countdown in seconds. If you want to start your data collection before the two minute countdown ends, add your active reagent and select the **OK** button. Alternatively, you can add your active reagent and wait for the countdown to end. The data collection will start automatically.

**Save the collected data**

Step 13

a. Once the run is finished, select the **Save as** command from the **File** menu.

b. In the **File name** field, enter a file name for this scan run.

c. Select **Save**. The data will be stored as a Batch file.

See also:
- [Scan How To](#)
- [Cary](#)
- [Options](#)
- [Reports](#)
- [Auto store](#)

**How to perform a fluorescence scan in signal-to-noise mode—Scan**

This demonstrates how to perform a fluorescence wavelength scan in signal-to-noise mode.

- **Start the Scan application**
- **Set up data collection parameters**
- **Set up instrument parameters such as Data Mode and Scan Setup**
- **Set up visual system monitoring**
- **Set up the display, signal-to-noise and filter options**
- **Make sure that no accessories are selected**
- Set up reporting and printing requirements
- Set up storage of collected data before run
- Set up email options
- Finish Setup
- Zero the instrument
- Start the Scan run
- Save the collected data

**Start the Scan application**

Step 1

Click the Windows **Start** button, point to **Programs, Cary Eclipse** and then select **Scan**. The Scan Application is displayed.

**Note**
If you are running a GLP system you may be prompted to enter a password before accessing the Scan Application.

**Set up data collection parameters**

Step 2 [Setup dialog]

Select the **Setup** button to display the **Setup** dialog and specify the method parameters for a new method.

**Set up instrument parameters such as Data Mode and Scan Setup**

Step 3 [Setup dialog | Cary page]

a. Set the **Data mode** to **Fluorescence**.

b. Set the **Scan setup** mode to **Emission**. During the emission scan, the excitation monochromator is set to a fixed wavelength and the emission monochromator is scanned over a wavelength range.

c. Set the **X Mode** to **Wavelength (nm)**.

d. Enter an **Excitation (nm)** value that is within the region where the fluorescent molecule to be scanned will absorb light. For purposes of this example, Anthracene will be used as an example and therefore a value of **360 nm** should be set.

e. Enter an **Excitation slit (nm)** value or **5** and an **Emission slit (nm)** value of **5**. Slits determine the resolution of the spectrum and therefore are used in conjunction with the **PMT Detector Voltage** (set on the **Options** page) to determine concentration. If a compound is highly fluorescent and has reasonable signal intensity (as is the case for Anthracene) the slits can be set quite narrow. If

f. Enter a **Start (nm)** value of **370 nm** and enter a **Stop (nm)** value of **520 nm**. The Start (nm) value should be set to the Excitation (nm) value plus the sum of the slits. In this example, as the Excitation value is 360 nm, and the sum of the slits is 10 nm, the **Start (nm)** value is set to 370. Typically the Stop (nm) value should be set to 150–200 nm greater than the Start (nm) value.

g. If selected, clear the **3-D Mode** check box.

**Set up visual system monitoring**

Step 4 [Setup dialog | Cary page]
Select the **Status Display** check box so that you can view various instrument parameters during the scan.

### Set up the display, signal-to-noise and filter options

**Step 5 [Setup dialog | Options page]**

- a. Select the **Options** tab to display the **Options** page.

- b. In the **Display options** group, select the **Overlay traces** check box to overlay the results of the traces on one graph.

- c. Select the **CAT or S/N mode** option. If this option is not available, clear the **Cycle mode** and/or **Smoothing** check boxes.

- d. Set the **Mode** to **S/N mode**. You will be informed that the **Scan control** on the Cary page will be set to **Manual**. Select **OK** to continue.

- e. Set the **S/N** value to **600** and the **Time out (s)** value to **1.000**. The Cary Eclipse instrument will control the **Averaging Time** by waiting at each data point and averaging the signal until the value set in the **S/N** field is obtained. This parameter is useful if you have areas of low transmission in your scan and wish to have the same level of 'noise' across the whole scan.

- f. Set the **Excitation filter** to **Auto** and the **Emission filter** to **Open**.

- g. Set the **PMT Detector voltage** to **Medium**. This setting can later be adjusted if results are over-range. If the signal is too high, decrease the PMT Detector Voltage. If the signal is too low, increase the PMT Detector Voltage.

### Make sure that no accessories are selected

**Step 6 [Setup dialog | Accessories page]**

Make sure that no options are selected on any of the tabs of the **Accessories** page.

### Set up reporting and printing requirements

**Step 7 [Setup dialog | Reports page]**

- a. Select the **Reports** tab to display the **Reports** page where you can specify your reporting requirements for this method.

  i. Enter your name in the **Name** entry field.

  ii. If required, enter any comments relating to your experiment in the **Comment** field.

  iii. Set up your report style by selecting the appropriate check boxes in the **Options** group.

    For example, select the **Auto print** check box to obtain a printout of your report automatically. Select the **Parameters** check box to include your experimental parameters in the report. Select the **Graph** check box to include a graph in the generated report.

    **Note**

    If **Auto print** is selected, the system will send the report information to the specified printer as well as displaying it in the Report area. However if **Auto print** is not selected, the report will be displayed in the Report area only. The Report area can be viewed by selecting the **Report** command from the **View** menu.

- b. Set up the **Peaks** reporting options.
i. Select **Maximum peaks** to report the peak with the largest peak threshold that exceeds the peak **Threshold** value.

ii. Select **All peaks** to report all peaks meeting the **Peak type** criterion and exceeding the **Threshold** value.

iii. Select the **Peak type** and specify the peak **Threshold**.

c. If required, select the **X-Y Pairs table** check box. You can use the **Actual Data interval** by which the data was collected or you can make the Cary Eclipse Interpolate the points to a new interval.

### Set up storage of collected data before run

#### Step 8 [Setup dialog | Auto Store page]

a. Select the **Auto Store** tab to display the **Auto store** page. Use this page to set up whether the collected data is to be saved, and if so, when the Cary Eclipse should store the information.

b. Select **Storage option of On; Prompt at end**.

c. Select the **Auto convert** option you require.

If you select **Select for ASCII (csv) or Select for ASCII (csv) with Log**, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format in the current folder.

### Set up email options

#### Step 9

Refer to [How to email your results](#) for instructions on setting up the email options.

### Finish Setup

#### Step 10

Once you are satisfied with your method setup select **OK** to confirm any changes you have made and close the **Setup** dialog.

### Zero the instrument

#### Step 11

a. Place the blank solution in the sample compartment of the Cary Eclipse instrument.

b. Click the **Zero** button to zero the system. Alternatively, select **Zero** from the **Commands** menu to perform a zero. When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the Scan Application window.

### Start the Scan run

#### Step 12

a. Select the **Start** button to commence a data collection. Alternatively, select **Start** from the **Commands** menu. The **Sample name** dialog is displayed.

b. In the **Sample name** dialog, enter the appropriate name for you sample and select **OK**. The scan will
commence and the trace will appear in the Graphics area.

Save the collected data

Step 13

a. Once the run is finished, select the Save as command from the File menu.

b. In the File name field, enter a file name for this scan run.

c. Select Save. The data will be stored as a Batch file.

See also:
Scan How To
Cary
Options
Reports
Auto store

How to perform a phosphorescence scan in cycle mode—Scan

This demonstrates how to perform a phosphorescence wavelength scan in cycle mode.

- Start the Scan application
- Set up data collection parameters
- Set up instrument parameters
- Set up visual system monitoring
- Set up the cycle mode and filter options
- Make sure that no accessories are selected
- Set up reporting and printing requirements
- Set up storage of collected data before run
- Set up email options
- Finish Setup
- Zero the instrument
- Start the Scan run
- Save the collected data

Start the Scan application

Step 1

Click the Windows Start button, point to Programs, Cary Eclipse and then select Scan. The Scan Application is displayed.

Note
If you are running a GLP system you may be prompted to enter a password before accessing the Scan Application.

Set up data collection parameters

Step 2 [Setup dialog]

Select the Setup button to display the Setup dialog and specify the method parameters for a new method.

Set up instrument parameters such as Data Mode and Scan Setup
Step 3 [Setup dialog | Cary page]

a. Set the **Data Mode** to **Phosphorescence**.

b. Select the **Options** button to display the **Phosphorescence Options** dialog. The values you enter for the options in this dialog depend upon the sample you are scanning.

For example, if you were analysing a sample such as Europium (Eu3+), which has a very short phosphorescence lifetime, it is recommended that you enter the following values:

- Total decay time: 0.005 s
- No. of flashes: 1
- Delay time: 0.100 ms
- Gate time: 5.000 ms

However, if you are analysing a sample such as Napthalene, which is known to have a standard phosphorescence lifetime, it is recommended that you enter the following values:

- Total decay time: 1 s
- No. of flashes: 10
- Delay time: 0.100 ms
- Gate time: 5.000 ms

c. Once you have made entered the values in the **Phosphorescence options** dialog, select **OK** to clear the dialog and return to the **Cary** page.

b. Set the **Scan Setup** mode to **Emission**. During the emission scan, the excitation monochromator is set to a fixed wavelength and the emission monochromator is scanned over a wavelength range.

d. Set the **X Mode** to **Wavelength (nm)**.

e. Enter an **Excitation (nm)** value of **340 nm**. The **Excitation (nm)** value must be within the region where the phosphorescent molecule to be scanned will absorb light.

f. Enter an **Excitation slit (nm)** value of **10** and an **Emission slit (nm)** value of **10**. Slits determine the resolution of the spectrum and therefore are used in conjunction with the **PMT Detector Voltage** (set on the **Options** page) to determine concentration. If a compound is not highly phosphorescent (as is the case for Europium) the slits need to be set quite wide.

g. Enter a **Start (nm)** value of **360 nm** and enter a **Stop (nm)** value of between **700** to **750 nm**. The Start (nm) value should be set to the Excitation (nm) value plus the sum of the slits. In this example, as the Excitation value is 340 nm, and the sum of the slits is 20 nm, the Start (nm) value is set to 360.

Typically phosphorescence samples should be scanned to above 600 nm, and preferably to between 700–750 nm, even if they fluoresce at as low as 300 nm.

h. If selected, clear the **3-D Mode** check box.

### Set up visual system monitoring

Step 3 [Setup dialog | Cary page]

Select the **Status Display** check box so that you can view various instrument parameters during the scan.

### Set up the cycle mode and filter options
Step 5 [Setup dialog | Options page]

a. Select the Options tab to display the Options page.

b. Select the Cycle mode option. This option enables you to collect scans as a function of time to determine whether the spectrum is changing. (By contrast, the Kinetics application enables you to determine whether a single wavelength is changing over time.)
   If this option is not available, clear the CAT or S/N mode check box.

c. Enter a No. of scans value of 15 to set the number of scans to be performed per cell/well.

d. Enter a Time value of 1.0000 to set the interval between each successive scan. If the time entered is less than the time required to complete a scan, the scans are performed continuously with no delay.

e. Set the Excitation filter to Auto. This will automatically move the filter wheel to the appropriate position for the selected excitation wavelength.

f. Set the Emission filter to Open in order to minimize steps in the spectra.

g. Set the PMT Detector Voltage to High. A High setting is recommended for low emitting samples. This setting can later be adjusted if results are over-range. If the signal is too high, decrease the PMT Detector Voltage.

Make sure that no accessories are selected

Step 5 [Setup dialog | Accessories page]

Ensure that no options are selected on any of the tabs in the Accessories page.

Set up reporting and printing requirements

Step 6 [Setup dialog | Reports page]

a. Select the Reports tab to display the Reports page where you can specify your reporting requirements for this method.
   i. Enter your name in the Name entry field.
      ii. If required, enter any comments relating to your experiment in the Comment field.
   iii. Set up your report style by selecting the appropriate check boxes in the Options group.

   For example, select the Auto print check box to obtain a printout of your report automatically.
   Select the Parameters check box to include your experimental parameters in the report.
   Select the Graph check box to include a graph in the generated report.

   **Note**
   If Auto print is selected, the system will send the report information to the specified printer as well as displaying it in the Report area. However if Auto print is not selected, the report will be displayed in the Report area only. The Report area can be viewed by selecting the Report command from the View menu.

b. Set up the Peaks reporting options.
   iii. Select Maximum peaks to report the peak with the largest peak threshold that exceeds the peak Threshold value.
      i. Select All peaks to report all peaks meeting the Peak type criterion and exceeding the Threshold value.
ii. Select the **Peak type** and specify the peak **Threshold**.

c. If required, select the **X-Y Pairs table** check box. You can use the **Actual Data Interval** by which the data was collected or you can make the Cary Eclipse Interpolate the points to a new interval.

### Set up storage of collected data before run

**Step 7 [Setup dialog | Auto Store page]**

a. Select the **Auto Store** tab to display the **Auto store** page. Use this page to set up whether the collected data is to be saved, and if so, when the Cary Eclipse should store the information.

b. Select **Storage option of On; Prompt at end**.

c. Select the Autoconvert option you require. If you select Select for ASCII (csv) or Select for ASCII (csv) with Log, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format in the current folder.

### Set up email options

**Step 8**

Refer to [How to email your results](#) for instructions on setting up the email options.

### Finish Setup

**Step 9**

Once you are satisfied with your method setup select **OK** to confirm any changes you have made and close the **Setup** dialog.

### Zero the instrument

**Step 10**

**Note:**

You MUST close the sample compartment prior to commencing a phosphorescence scan, as phosphorescence scanning is not immune to room light.

1. Place the blank solution in the sample compartment of the Cary Eclipse instrument.

2. Click the **Zero** button to zero the system. Alternatively, select **Zero** from the **Commands** menu to perform a zero. When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the Scan Application window.

### Start the Scan run

**Step 11**

a. Select the **Start** button to commence a data collection. Alternatively, select **Start** from the **Commands** menu. The **Sample name** dialog is displayed.

b. In the **Sample name** dialog, enter the appropriate name for your sample and select **OK**. The **Sync Start** dialog is displayed.
c. The **Sync Start** dialog displays a two minute countdown in seconds. If you want to start your data collection before the two minute countdown ends, add your active reagent and select the **OK** button. Alternatively, you can add your active reagent and wait for the countdown to end. The data collection will start automatically.

### Save the collected data

#### Step 12

a. Once the run is finished, select the **Save As** command from the **File** menu.

b. In the **File name** field, enter a file name for this scan run.

c. Select **Save**. The data will be stored as a Batch file.

**See also:**
[Scan How To]

### How to perform a phosphorescence scan in signal-to-noise mode—Scan

This demonstrates how to perform a phosphorescence wavelength scan in signal-to-noise mode.

- **Start the Scan application**
- **Set up data collection parameters**
- **Set up instrument parameters**
- **Set up visual system monitoring**
- **Set up the display, signal-to-noise and filter options**
- **Make sure that no accessories are selected**
- **Set up reporting and printing requirements**
- **Set up storage of collected data before run**
- **Set up email options**
- **Finish Setup**
- **Zero the instrument**
- **Start the Scan run**
- **Save the collected data**

### Start the Scan application

#### Step 1

Click the Windows **Start** button, point to **Programs, Cary Eclipse** and then select **Scan**. The Scan Application is displayed.

**Note**

If you are running a GLP system you may be prompted to enter a password before accessing the Scan Application.

### Set up data collection parameters

#### Step 2 [Setup dialog]

Select the **Setup** button to display the **Setup** dialog and specify the method parameters for a new method.

### Set up instrument parameters such as Data Mode and Scan Setup

#### Step 3 [Setup dialog] | Cary page
a. Set the **Data Mode** to **Phosphorescence**.

b. Select the **Options** button to display the **Phosphorescence Options** dialog. The values you enter for the options in this dialog depend upon the sample you are scanning.

For example, if you were analysing a sample such as Europium (Eu³⁺), which has a very short phosphorescence lifetime, it is recommended that you enter the following values:

- **Total decay time**: 0.005 s
- **No. of flashes**: 1
- **Delay time**: 0.100 ms
- **Gate time**: 5.000 ms

However, if you are analysing a sample such as Napthalene, which is known to have a standard phosphorescence lifetime, it is recommended that you enter the following values:

- **Total decay time**: 1 s
- **No. of flashes**: 10
- **Delay time**: 0.100 ms
- **Gate time**: 5.000 ms

c. Once you have made entered the values in the **Phosphorescence options** dialog, select **OK** to clear the dialog and return to the **Cary** page.

d. Set the **Scan Setup** mode to **Emission**. During the emission scan, the excitation monochromator is set to a fixed wavelength and the emission monochromator is scanned over a wavelength range.

e. Set the **X Mode** to **Wavelength (nm)**.

f. Enter an **Excitation (nm)** value of **340 nm**. The **Excitation (nm)** value must be within the region where the phosphorescent molecule to be scanned will absorb light.

g. Enter an **Excitation slit (nm)** value of **10** and an **Emission slit (nm)** value of **10**. Slits determine the resolution of the spectrum and therefore are used in conjunction with the *PMT Detector Voltage* (set on the **Options** page) to determine concentration. If a compound is not highly phosphorescent (as is the case for Europium) the slits need to be set quite wide.

h. Enter a **Start (nm)** value of **360 nm** and enter a **Stop (nm)** value of between 700 to 750 nm. The **Start (nm)** value should be set to the Excitation (nm) value plus the sum of the slits. In this example, as the Excitation value is 340 nm, and the sum of the slits is 20 nm, the **Start (nm)** value is set to 360. Typically phosphorescence samples should be scanned to above 600 nm, and preferably to between 700–750 nm, even if they fluoresce at as low as 300 nm.

i. If selected, clear the **3-D Mode** check box.

**Set up visual system monitoring**

Step 3 [Setup dialog | Cary page]

Select the **Status Display** check box so that you can view various instrument parameters during the scan.

**Set up the display, signal-to-noise and filter options**

Step 5 [Setup dialog | Options page]
a. Select the **Options** tab to display the **Options** page.

b. In the **Display options** group, select the **Overlay traces** check box to overlay the results of the traces on one graph.

c. Select the **CAT or S/N mode** option. If this option is not available, clear the **Cycle mode** and/or **Smoothing** check boxes.

d. Set the **Mode** to **S/N mode**.
   You will be informed that the **Scan control** on the **Cary** page will be set to **Manual**. Select **OK** to continue.

e. Set the S/N value to **600** and the **Time out (s)** value to **1.000**. The Cary Eclipse instrument will control the **Averaging Time** by waiting at each data point and averaging the signal until the value set in the S/N field is obtained. This parameter is useful if you have areas of low transmission in your scan and wish to have the same level of 'noise' across the whole scan.

f. Set the **Excitation filter** to **Auto**. This will automatically move the filter wheel to the appropriate position for the selected excitation wavelength.

g. Set the **Emission filter** to **Open** in order to minimize steps in the spectra.

h. Set the **PMT Detector Voltage** to **High**. A **High** setting is recommended for low emitting samples. This setting can later be adjusted if results are over-range. If the signal is too high, decrease the **PMT Detector Voltage**.

**Make sure that no accessories are selected**

Step 5 [Setup dialog | Accessories page]

Ensure that no options are selected on any of the tabs of the **Accessories** page.

**Set up reporting and printing requirements**

Step 6 [Setup dialog | Reports page]

a. Select the **Reports** tab to display the **Reports** page where you can specify your reporting requirements for this method.

   i. Enter your name in the **Name** entry field.
   ii. If required, enter any comments relating to your experiment in the **Comment** field.
   iii. Set up your report style by selecting the appropriate check boxes in the **Options** group.

   For example, select the **Auto print** check box to obtain a printout of your report automatically. Select the **Parameters** check box to include your experimental parameters in the report. Select the **Graph** check box to include a graph in the generated report.

   **Note**
   If **Auto print** is selected, the system will send the report information to the specified printer as well as displaying it in the Report area. However if **Auto print** is not selected, the report will be displayed in the Report area only. The Report area can be viewed by selecting the **Report** command from the **View** menu.

b. Set up the **Peaks** reporting options.

   iii. Select **Maximum peaks** to report the peak with the largest peak threshold that exceeds the peak
Threshold value.
   i. Select All peaks to report all peaks meeting the Peak type criterion and exceeding the Threshold value.
   ii. Select the Peak type and specify the peak Threshold.

c. If required, select the X-Y Pairs table check box. You can use the Actual Data Interval by which the data was collected or you can make the Cary Eclipse Interpolate the points to a new interval.

Set up storage of collected data before run

Step 7 [Setup dialog | Auto Store page]

a. Select the Auto Store tab to display the Auto store page. Use this page to set up whether the collected data is to be saved, and if so, when the Cary Eclipse should store the information.

b. Select Storage option of On; Prompt at end.

c. Select the Autoconvert option you require.
   If you select Select for ASCII (csv) or Select for ASCII (csv) with Log, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format in the current folder.

Set up email options

Step 8

Refer to How to email your results for instructions on setting up the email options.

Finish Setup

Step 9

Once you are satisfied with your method setup select OK to confirm any changes you have made and close the Setup dialog.

Zero the instrument

Step 10

Note:
You MUST close the sample compartment prior to commencing a phosphorescence scan, as phosphorescence scanning is not immune to room light.

   a. Place the blank solution in the sample compartment of the Cary Eclipse instrument.

   b. Select the Zero button to zero the system. Alternatively, select Zero from the Commands menu to perform a zero. The system will perform an instrument zero on the blank solution. When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the Scan Application window.

Start the Scan run

Step 11

a. Select the Start button to commence a data collection. Alternatively, select Start from the Commands menu. The Sample Name dialog is displayed.
b. Place the sample in the sample compartment of the Cary Eclipse instrument.

c. In the Sample Name dialog, enter the appropriate name for your sample and select OK. The Scan run will commence and the corrected trace will appear in the Graphics area.

Save the collected data

Step 12

a. Once the run is finished, select the Save As command from the File menu.

b. In the File name field, enter a file name for this scan run.

c. Select Save. The data will be stored as a Batch file.

See also:
Scan How To

How to perform a bio-/chemi-luminescence scan in cycle mode—Scan

This demonstrates how to perform a bio-/chemi-luminescence wavelength scan in signal-to-noise mode.

- Start the Scan application
- Set up data collection parameters
- Set up instrument parameters
- Set up visual system monitoring
- Set up the display, cycle mode and filter options
- Make sure that no accessories are selected
- Set up reporting and printing requirements
- Set up storage of collected data before run
- Set up email options
- Finish Setup
- Zero the instrument
- Start the Scan run
- Save the collected data

Start the Scan application

Step 1

Click the Windows Start button, point to Programs, Cary Eclipse and then select Scan. The Scan Application is displayed.

Note
If you are running a GLP system you may be prompted to enter a password before accessing the Scan Application.

Set up data collection parameters

Step 2 [Setup dialog]

Select the Setup button to display the Setup dialog and specify the method parameters for a new method.

Set up instrument parameters such as Data Mode and Scan Setup

Step 3 [Setup dialog] | Cary page
a. Set the **Data Mode** to **Bio-/Chemi-luminescence**.

b. Select the **Options** button to display the **Bio-/Chemi-luminescence Options** dialog. Enter the **Gate time** to **200 ms**. The Gate Time is the overall reading time of the emission signal and is typically set to between 100–500 ms.

c. Select **OK** to clear the **Bio-/Chemi-luminescence Options** dialog and return to the **Cary** page. Note that in the **Scan setup** group, the **Excitation** and **Synchronous** options are unavailable as the lamp does not flash for a Bio-/Chemi-luminescence scan. Therefore, it is set to **Emission** mode.

d. Set the **X Mode** to **Wavelength (nm)**.

e. Enter a **Start (nm)** value of **300 nm** and enter a **Stop (nm)** value of between **750 nm**. The Start (nm) value should be set to the Excitation (nm) value plus the sum of the slits.

f. Enter an **Emission slit (nm)** value of **10**. Slits determine the resolution of the spectrum and therefore are used in conjunction with the **PMT Detector Voltage** (set on the **Options** page) to determine concentration.

### Set up visual system monitoring

**Step 4** [Setup dialog | Cary page]

Select the **Status Display** check box so that you can view various instrument parameters during the scan.

### Set up the display, cycle mode and filter options

**Step 5** [Setup dialog | Options page]

a. Select the **Options** tab to display the **Options** page.

b. In the **Display options** group, select the **Overlay traces** check box to overlay the results of the traces on one graph.

c. Select the **CAT or S/N mode** option. If this option is not available, clear the **Cycle mode** and/or **Smoothing** check boxes.

d. Set the **Mode** to **S/N mode**.
   You will be informed that the **Scan control** on the **Cary** page will be set to **Manual**. Select **OK** to continue.

e. Set the **S/N** value to **600** and the **Time out (s)** value to **1.000**. The Cary Eclipse instrument will control the **Averaging Time** by waiting at each data point and averaging the signal until the value set in the **S/N** field is obtained. This parameter is useful if you have areas of low transmission in your scan and wish to have the same level of 'noise' across the whole scan.

f. Set the **Emission filter** to **Open** in order to minimize steps in the spectra.

g. Set the **PMT Detector Voltage** to **High**. A **High** setting is recommended for low emitting samples. This setting can later be adjusted if results are over-range. If the signal is too high, decrease the **PMT Detector Voltage**.

### Make sure that no accessories are selected

**Step 6** [Setup dialog | Accessories page]

Make sure that no options are selected on any of the tabs in the **Accessories** page.
Set up reporting and printing requirements

Step 7 [Setup dialog | Reports page]

a. Select the **Reports** tab to display the **Reports** page where you can specify your reporting requirements for this method.
   i. Enter your name in the **Name** entry field.
   ii. If required, enter any comments relating to your experiment in the **Comment** field.
   iii. Set up your report style by selecting the appropriate check boxes in the **Options** group.

   For example, select the **Auto print** check box to obtain a printout of your report automatically. Select the **Parameters** check box to include your experimental parameters in the report. Select the **Graph** check box to include a graph in the generated report.

   **Note**
   If **Auto print** is selected, the system will send the report information to the specified printer as well as displaying it in the Report area. However if **Auto print** is not selected, the report will be displayed in the Report area only. The Report area can be viewed by selecting the **Report** command from the **View** menu.

b. Set up the **Peaks** reporting options.

   iii. Select **Maximum peaks** to report the peak with the largest peak threshold that exceeds the peak **Threshold** value.
   i. Select **All peaks** to report all peaks meeting the **Peak type** criterion and exceeding the **Threshold** value.
   ii. Select the **Peak type** and specify the peak **Threshold**.

c. If required, select the **X-Y Pairs table** check box. You can use the **Actual Data Interval** by which the data was collected or you can make the Cary Eclipse Interpolate the points to a new interval.

Set up storage of collected data before run

Step 8 [Setup dialog | Auto Store page]

a. Select the **Auto Store** tab to display the **Auto store** page. Use this page to set up whether the collected data is to be saved, and if so, when the Cary Eclipse should store the information.

b. Select **Storage** option of **On; Prompt at end**.

   c. Select the Autoconvert option you require.
   If you select Select for ASCII (csv) or Select for ASCII (csv) with Log, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format in the current folder.

Set up email options

Step 9

Refer to **How to email your results** for instructions on setting up the email options.

Finish Setup

Step 10
Once you are satisfied with your method setup select **OK** to confirm any changes you have made and close the **Setup** dialog.

**Zero the instrument**

**Step 11**

a. Place the blank solution in the sample compartment of the Cary Eclipse instrument.

b. Click the **Zero** button to zero the system. Alternatively, select **Zero** from the **Commands** menu to perform a zero. When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the Scan Application window.

**Start the Scan run**

**Step 12**

a. Select the Start **button** to commence a data collection. Alternatively, select **Start** from the **Commands** menu. The **Sample Name** dialog is displayed.

b. In the **Sample Name** dialog, enter the appropriate name for you sample and select **OK**. The Scan run will commence and the corrected trace will appear in the Graphics area.

**Save the collected data**

**Step 13**

a. Once the run is finished, select the **Save As** command from the **File** menu.

b. In the **File name** field, enter a file name for this scan run.

c. Select **Save**. The data will be stored as a Batch file.

---

**How to perform a bio-/chemi-luminescence scan in signal-to-noise mode—Scan**

This demonstrates how to perform a bio-/chemi-luminescence wavelength scan in signal-to-noise mode.

- **Start the Scan application**
- **Set up data collection parameters**
- **Set up instrument parameters**
- **Set up visual system monitoring**
- **Set up the display, signal-to-noise and filter options**
- **Make sure that no accessories are selected**
- **Set up reporting and printing requirements**
- **Set up storage of collected data before run**
- **Set up email options**
- **Finish Setup**
- **Zero the instrument**
- **Start the Scan run**
- **Save the collected data**
Start the Scan application

Step 1

Click the Windows Start button, point to Programs, Cary Eclipse and then select Scan. The Scan Application is displayed.

Note
If you are running a GLP system you may be prompted to enter a password before accessing the Scan Application.

Set up data collection parameters

Step 2 [Setup dialog]

Select the Setup button to display the Setup dialog and specify the method parameters for a new method.

Set up instrument parameters such as Data Mode and Scan Setup

Step 3 [Setup dialog] | Cary page]

a. Set the Data Mode to Bio-/Chemi-luminescence.

b. Select the Options button to display the Bio-/Chemi-luminescence Options dialog. Enter the Gate time to 200 ms. The Gate Time is the overall reading time of the emission signal and is typically set to between 100–500 ms.

c. Select OK to clear the Bio-/Chemi-luminescence Options dialog and return to the Cary page. Note that in the Scan setup group, the Excitation and Synchronous options are unavailable as the lamp does not flash for a Bio-/Chemi-luminescence scan. Therefore, it is set to Emission mode.

d. Set the X Mode to Wavelength (nm).

e. Enter a Start (nm) value of 300 nm and enter a Stop (nm) value of between 750 nm. The Start (nm) value should be set to the Excitation (nm) value plus the sum of the slits.

f. Enter an Emission slit (nm) value of 10. Slits determine the resolution of the spectrum and therefore are used in conjunction with the PMT Detector Voltage (set on the Options page) to determine concentration.

g. In the Scan control group, set the Averaging time to 0.20000 s and the Data interval to 1.0000 nm.

Set up visual system monitoring

Step 4 [Setup dialog | Cary page]

Select the Status Display check box so that you can view various instrument parameters during the scan.

Set up the display, signal-to-noise and filter options

Step 5 [Setup dialog | Options page]

a. Select the Options tab to display the Options page.

b. In the Display options group, select the Overlay traces check box to overlay the results of the traces on one graph.
c. Select the **CAT or S/N mode** option. If this option is not available, clear the **Cycle mode** and/or **Smoothing** check boxes.

d. Set the **Mode** to **S/N mode**.
   You will be informed that the **Scan control** on the Cary page will be set to **Manual**. Select **OK** to continue.

e. Set the **S/N value** to 600 and the **Time out (s)** value to 1.000. The Cary Eclipse instrument will control the **Averaging Time** by waiting at each data point and averaging the signal until the value set in the **S/N** field is obtained. This parameter is useful if you have areas of low transmission in your scan and wish to have the same level of 'noise' across the whole scan.

f. Set the **Emission filter** to **Open** in order to minimize steps in the spectra.

g. Set the **PMT Detector Voltage** to **High**. A **High** setting is recommended for low emitting samples. This setting can later be adjusted if results are over-range. If the signal is too high, decrease the **PMT Detector Voltage**.

**Make sure that no accessories are selected**

Step 6 [Setup dialog | Accessories page]

Make sure that no options are selected on any of the tabs in the **Accessories** page.

**Set up reporting and printing requirements**

Step 7 [Setup dialog | Reports page]

a. Select the **Reports** tab to display the **Reports** page where you can specify your reporting requirements for this method.

   i. Enter your name in the **Name** entry field.

   ii. If required, enter any comments relating to your experiment in the **Comment** field.

   iii. Set up your report style by selecting the appropriate check boxes in the **Options** group.

      For example, select the **Auto print** check box to obtain a printout of your report automatically. Select the **Parameters** check box to include your experimental parameters in the report. Select the **Graph** check box to include a graph in the generated report.

      **Note**

      If **Auto print** is selected, the system will send the report information to the specified printer as well as displaying it in the Report area. However if **Auto print** is not selected, the report will be displayed in the Report area only. The Report area can be viewed by selecting the **Report** command from the **View** menu.

b. Set up the **Peaks** reporting options.

   i. Select **Maximum peaks** to report the peak with the largest peak threshold that exceeds the peak **Threshold** value.

   ii. Select **All peaks** to report all peaks meeting the **Peak type** criterion and exceeding the **Threshold** value.

   iii. Select the **Peak type** and specify the peak **Threshold**.

c. If required, select the **X-Y Pairs table** check box. You can use the **Actual Data Interval** by which the data was collected or you can make the Cary Eclipse Interpolate the points to a new interval.
Set up storage of collected data before run

Step 8 [Setup dialog | Auto Store page]

a. Select the Auto Store tab to display the Auto store page. Use this page to set up whether the collected data is to be saved, and if so, when the Cary Eclipse should store the information.

b. Select Storage option of On; Prompt at end.

c. Select the Autoconvert option you require.
   If you select Select for ASCII (csv) or Select for ASCII (csv) with Log, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary Eclipse format as well as ASCII XY pairs format in the current folder.

Set up email options

Step 9

Refer to How to email your results for instructions on setting up the email options.

Finish Setup

Step 10

Once you are satisfied with your method setup select OK to confirm any changes you have made and close the Setup dialog.

Zero the instrument

Step 11

a. Place the blank solution in the sample compartment of the Cary Eclipse instrument.

b. Click the Zero button to zero the system. Alternatively, select Zero from the Commands menu to perform a zero. When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the Scan Application window.

Start the Scan run

Step 12

a. Select the Start button to commence a data collection. Alternatively, select Start from the Commands menu. The Sample Name dialog is displayed.

b. In the Sample Name dialog, enter the appropriate name for you sample and select OK. The Scan run will commence and the corrected trace will appear in the Graphics area.

Save the collected data

Step 13

a. Once the run is finished, select the Save As command from the File menu.

b. In the File name field, enter a file name for this scan run.
c. Select **Save**. The data will be stored as a Batch file.

See also:
[Scan How To](#)

### How to generate a report from previously stored data—Scan

**Step 1**
Select **Open…** from the **File** menu to display the Windows **Open** dialog.

**Step 2**
A list of previously stored files will appear. If this is not a list of batch files, select the down arrow at the right of the **Files of type** list box and select 'Batch' to list all the batch files.

**Step 3**
Select the Batch file you require then select **Open**.

The system will automatically retrieve the collected data into the Graphics area. The method that was used to collect the data will also be retrieved.

**Note**
You can also retrieve data from Methods (*.FMKN), Data (*.FDKN), Report (*.FRKN), Spreadsheet ASCII (*.CSV), GRAMS (*.SPC) and Rich Text Format (*.RTF) file types.

**Step 4**
Select the **Recalculate** button to display the **Setup** dialog.

**Step 5**
Select the **Reports** tab and on the **Reports** page specify the information you would like to include in the report. Select **Auto Print** if you require the new report to print to the Reports area and to a printer.

**Step 6**
Click **OK** to display the results in the Report area (and send to the printer if **Auto Print** is selected).

**Note**
If you require a printout of only the current graphics in the Graphics area of a previously stored data file:

a. Select the **Clear Report** option in the Commands menu to clear any text that may be displayed in the Report area.
   
b. Press the **Print** button to obtain a printout of the graphics.

Alternatively tick only the **Graph** check box in the **Reports** page to obtain a printout of graphics only.

See also:
[How to use advanced reporting options](#)

### How to import sample names into the Wellplate sample list—Scan

If you want to import sample names into the **Wellplate sample list** in the Scan application, then you simply need to enter your data in a *.txt file and then import this file into the **Wellplate sample list** dialog. The data should be
entered in the following manner:

Each sample name and Well position in the imported file must be on a separate line. Enter the Well position on the line immediately after its corresponding Sample Name. Note that any tabs in the file are ignored.

To illustrate this procedure, the following Sample name file (shown on the left) will be used. The picture on the right shows the wells that will be selected for inclusion in the Wellplate sample list. Note that you do not need to include the well positions, as they are entered automatically (in this case Sample_name1–Sample_name6 will correspond to wells A1–A6 and Sample_name7–Sample_name12 will correspond to wells B1–B6).

To import sample names:

Step 1

Enter your data into a text editor (e.g. Notepad) and save it as a text (*.TXT) file. Refer to Figure 1 for an example of the correct format for entering the text.

**Note:**
The first line of the text file you create is not included when you import the sample names list. Therefore you should leave this line blank.

Step 2

From the Scan Application window, select the **Setup** button to display the **Setup** dialog.

Step 3

Select the **Accessories** tab to display the **Accessories** page. Select the **Wellplate** tab and from the **Wellplate** page.

Step 5

From the Wellplate diagram, select the wells to be included in your analysis. (Refer to Figure 2.) These wells will be included in the **Wellplate sample list**.

Step 6

Select the **Sample names** button to display the **Wellplate sample list**.

Step 7
Select the **Import Names** button. The **Open** dialog is displayed.

**Step 8**

Browse for your text file and select **Open** to open the file. The Sample Names and their corresponding Well position will appear in the **Sample Name** table.

See also:
- [Wellplate sample list](#)
- [Wellplate Accessories page](#)
- [About the Wellplate Reader](#)

**How to resize graph boxes—Scan**

**Step 1**

Click on the graph that you want to resize so that the highlight box appears around it.

**Step 2**

Move the cursor to the edge of the highlight box and place it over one of the square marks (e.g. ■) in the box until it turns into a double headed arrow (↔).

**Step 3**

Click and hold the left mouse button and drag the box to the desired size.

**Step 4**

Release the left mouse button.

See also:
- [How to alter the look of your graphs](#)

**How to zoom a graph—Scan**

To zoom a graph, do the following:

**Step 1**

Click on the button on the toolbar, or select **Cursor Mode** from the **Graph** menu. The **Cursor Modes** dialog will appear.

**Step 2**

Select **Free** and select the **OK** button.

**Step 3**

Click and hold down left mouse button in the highlighted graph box.

**Step 4**

Drag the mouse sideways and up or down. You will see a zoom box made up of a dotted line appear on the screen. (See picture below.)

See also:
- [Wellplate sample list](#)
- [Wellplate Accessories page](#)
- [About the Wellplate Reader](#)
Step 5

Release the left mouse button. The area defined by the zoom box will be redrawn to fill the entire graph box, and the scans within the box will be enlarged accordingly. The abscissa (X) and ordinate (Y) coordinates displayed below the graph will also be changed to reflect the positions of the corners of the zoom box.

You can repeat this process if you need to zoom in even further.

Hot Tip
To move the box on the screen, hold down the left and right mouse buttons together. You can then position the box anywhere in the graph box. Releasing the right mouse button will still allow you to resize the zoom box.

Note
The system may limit the extent to which you can Zoom.

See also:
How to scale your graph manually
How to scale your graph automatically

How to take the derivative of a trace

Some uses of derivatives are:

- To resolve partly overlapping peaks
- To quantitatively determine trace compounds
- To characterize pure compounds
- Purity testing

To take the derivative of a collected trace and plot the result, follow these steps:

Step 1
Click on the trace to be smoothed. It will become red (highlighted).

Step 2
Click the Selected Trace or Focused Trace button. The name of the highlighted trace will appear in the calculation display field.

Step 3
In the Trace group, select your derivative order in the Operation field. Choose from derivatives 1 to 4 (Deriv1, Deriv2, Deriv3, Deriv4). You may have to scroll to find the derivative you require.

Step 4
Enter a Filter Size and an Interval in the corresponding fields.

Step 5
In the Display Options group, select New Graph to plot the result in a new graph box, or Selected Graph to plot the
Step 6

Click the Apply button in the Trace group. Your Deriv selection will be displayed before the trace name in the calculation display field.

Step 7

Click on the equals (=) button. The result will be displayed in the Graphics area.

**How to smooth a trace**

To smooth a collected trace and plot the result, follow these steps:

Step 1

Click on the trace to be smoothed. It will become red (highlighted).

Step 2

Click on the Maths button in the toolbar (it's on the far right) or select the Maths menu.

Step 3

Click the Selected Trace or Focused Trace button. The name of the highlighted trace will appear in the calculation display field.

Step 4

In the Trace group, select Smooth in the Operation field. (You may have to scroll to find it.)

Step 5

Enter a Filter Size and an Interval in the corresponding fields.

Step 6

In the Display Options group select New Graph to plot the result in a new graph box, or Selected Graph to plot the result in the current graph box.

Step 7

Click the Apply button in the Trace group. The word ‘Smooth’ will appear before the trace name in the calculation display field.

Step 8

Click on the equals (=) button. The result will be displayed in the Graphics area.

See also:

About derivatives
Introduction

Use the Simple Reads application to perform simple fluorescence, phosphorescence and bio/chemiluminescence readings of samples. In addition, simple mathematical operations can be performed (via the User Collect option) on readings performed at multiple wavelengths.

To access the Simple Reads Application, click the Windows Start button, point to Programs, Cary Eclipse and then select Simple Reads.

Software controls
The application comprises the following:

<table>
<thead>
<tr>
<th>Instrument Status Readings</th>
<th>The Instrument Status Readings, which display the Y and X values of the graph axes are displayed in the top left and right corners of the Simple Reads Application Window.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Y value</td>
<td>Displays the current Y value, usually intensity. This value is continually updated during a data collection.</td>
</tr>
<tr>
<td>X value</td>
<td>Displays the current X value, which is the current reading for the time. On the Cary page of the Setup dialog, choose whether to display the time value in minutes or seconds.</td>
</tr>
<tr>
<td>Read button</td>
<td>Select Read to perform a simple read using the current Setup parameters. The results of the read will be displayed in the report area.</td>
</tr>
<tr>
<td>Simple Reads Report</td>
<td>In the Simple Reads Report area you can view the current report. The report may list the results of your readings and the parameters (for example, ordinate mode and wavelengths) used to collect the results. You can copy the generated report onto the Windows clipboard. To do this, highlight the report (or the relevant section) and press CTRL+C. You can then paste the report into a word processor such as Microsoft® Word.</td>
</tr>
<tr>
<td>Setup</td>
<td>Select Setup to display the Setup dialog where you can enter the instrument and wavelength parameters and the filter and voltage options for a Simple Reads run. This command is also accessible from the Setup menu.</td>
</tr>
<tr>
<td>Zero</td>
<td>Select Zero to zero the current intensity value. To start your data at 0 intensity, select Zero prior to selecting the Read button. This command is also accessible from the Commands menu or by pressing the F5 function key.</td>
</tr>
<tr>
<td>Clear report</td>
<td>Select Clear report to clear all the text in the Report area. This command is also accessible from the Edit menu.</td>
</tr>
<tr>
<td>Print</td>
<td>Select Print to display the Windows Print dialog where you can select the printer that the report will be printed to. You can also choose which pages to print in the currently displayed report as well as the number of copies you wish to print. This command is also accessible from the File menu.</td>
</tr>
<tr>
<td>Read</td>
<td>Select Read to commence a Simple Reads measurement using the current Setup parameters. This command is also accessible from the Commands menu.</td>
</tr>
</tbody>
</table>

**Hot Tip**

By becoming familiar with these commands (Setup, Zero, Clear report, Print), you can remove
the **Command** buttons from the Simple Reads window. This will automatically enlarge the Report area, making viewing results easier. To do this, cancel the **Buttons** item in the **View** menu.

**Menus** The Simple Reads Application also contains the following menus:
- File, Edit, View, Commands, Setup, Help

---

**Getting ready to read—Simple Reads**

- **Setup dialog**
- **Setup menu**
- **Cary page**
  - User Collect Example
  - Bio-/Chemi-luminescence and Phosphorescence Options dialog
  - Gate Time for Bio-/Chemi-luminescence
  - Gate Time for Phosphorescence
  - How many flashes?
- **Options page**

**Setup—Simple Reads**

Use the **Setup** dialog to set up your instrument for a Simple Reads measurement.

To access the **Setup** dialog select the **Setup** button from the Simple Reads Application window or the **Setup** command from the **Setup** menu.

The dialog consists of the following tabbed pages:

- **Cary**
- **Options**

**Setup menu—Simple Reads**

Use the **Setup** menu to access the parameters for setting up a Simple Read measurement.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
</tr>
</tbody>
</table>
### Setup

| Setup | Choose Setup to display the Setup dialog, where you can set up your instrument for a Simple Reads measurement. The Setup dialog can also be accessed by selecting the Setup button from the Simple Reads Application window. |

---

See also: Setup

## Cary—Simple Reads

Use the Cary page to set up the Cary Eclipse instrument parameters associated with a Simple Reads measurement.

To access the Cary page, select the Setup command from the Setup menu. You can also access the page from the Setup dialog, by selecting the Cary tab. To access the Setup dialog, select the Setup button from the Simple Reads Application window or the Setup command from the Setup menu.

The page comprises the following:

### Instrument setup

<table>
<thead>
<tr>
<th>Data mode</th>
<th>Fluorescence</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fluorescence refers to the emission of light from an excited single state. The fluorescence signal is measured when the lamp flashes. Fluorescence mode has excitation, emission and synchronous scan modes.</td>
<td></td>
</tr>
</tbody>
</table>

**Bio-/Chemiluminescence**

Bio-/Chemiluminescence refers to bio- or chemi-luminescence, where the emission is generated via a chemical or biochemical reaction. The lamp does not flash in Bio-/Chemiluminescence mode. Emission is the only scan mode of bio-/chemi-luminescence.

**Phosphorescence**

Phosphorescence refers to the emission of light from an excited triplet state. Phosphorescence is measured after the lamp has stopped flashing. Phosphorescence mode has excitation, emission and synchronous scan modes.

**Options**

- Click Options to open the Options dialog where you can set parameters such as the Decay, Delay and Gate times for your sample.

**Note**

The Options button is only visible if you have selected Bio-/Chemiluminescence or Phosphorescence as the Data Mode.

### Wavelength setup

| Ex. Wavelength (nm) | Set the wavelength of the excitation monochromator, which is the wavelength value at which you want to excite your sample. To set the monochromators to zero order (white light), from the Commands menu, point to Zero Order and select Excitation. A tick next to this option indicates that it is selected. Clear this option to return to |

---
monochromator light.

**Hot Tip**
Click the arrow to the right of the field to quickly select the most recent wavelength entries.

**Em. Wavelength (nm)**
Set the wavelength of the emission monochromator, which is the wavelength value at which your sample emits. To set the monochromators to zero order (white light), from the **Commands** menu, point to **Zero Order** and select **Emission**. A tick next to this option indicates that it is selected. Clear this option to return to monochromator light.

**Hot Tip**
Click the arrow to the right of the field to quickly select the most recent wavelength entries.

**User Collect**
Select User Collect to change the Em. Wavelength field to an entry field in which you can enter an ADL command. The Syntax for ADL commands is:

```
Read (Ex. wavelength, Em. Wavelength)
```

Use this entry field to carry out multiple sample reads at various wavelengths and then apply simple mathematical equations to generate a result. You can pre-define the ADL User Collect commands present in the drop down list on the Edit tab in System Information.

This allows an ADL command to be entered which applies a mathematical expression to the intensity value collected. The most recent user collect entries will be saved in the drop down list to a maximum of 10.

**Hot Tip**
You can perform multi-ratio and multi-equation reads using the User Collect option. That is, you can calculate more than one ratio or equation in the one read, rather than performing separate reads for each ratio or equation. For multi-ratios, use this format:

```
Read(355, 405)/Read(355/480) etc.
```

An example of a multi-equation might be:

```
2.1*Read(340, 510)/Read(405, 600);  
7.5*Read(350,450)-4.5*Read(525, 610)+  
25.5*Read(600, 750)
```

The semi-colon (;) separates each ratio or equation. In addition to the results of each ratio or equation, the raw readings for each wavelength used will also be reported.

**Ex. Slit (nm)**
Set the slit width of the excitation monochromator

**Em. Slit (nm)**
Set the slit width of the emission monochromator
**Ave time (s)** Set the amount of time, in seconds, for which data is averaged. To do this, enter the amount in the Ave. time field, or use the up/down arrows to the right of the field to increase or decrease the time. The averaged result is displayed in the report.

When setting the Ave Time, consider the following:

- The longer the Ave Time, the longer the signal will be averaged
- A longer Ave Time will result in more precise results as the averaging of the signal reduces any contribution of background ‘noise’.

Averaging is calculated as follows:

*For fluorescence:*
the number of readings averaged are taken from consecutive flashes at 80 Hz where:
number of consecutive flashes = averaging time/0.0125.

*For phosphorescence:*
the intensity readings are taken from different decays, where:
number of decays = averaging time/gate time.

*For bio-/chemi-luminescence:*
a signal is collected every Gate time, and (averaging time/gate time) signals are averaged at each data interval

See also:
Simple Reads Application
Options dialog
User Collect examples

### User Collect examples—Simple Reads

You can extend the functionality of your Cary Eclipse instrument by using the built-in Read command. This command enables you to change the instrument to a different wavelength and take a reading. You can combine this command with the mathematical operators: ‘+’, ‘-’, ‘*’ and ‘/’. This is illustrated in the examples below.

**Read(500)**

This will cause the system to change the wavelength to 500 nm and then take a reading. The result will be printed in the Report area.

**260 nm/280 nm ratio**

Read(260)/Read(280)

In many Life Science settings such as medical institutions and university research departments, nucleic acids are extracted from living cells and are then purified. In these fields of science it is imperative that the samples collected are free from contamination by other molecules such as proteins and chemicals...
like phenol. To determine the purity and type of nucleic acid in a sample, it is necessary to calculate the ratio of intensity values, read at 260 and 280 nm. At 260 nm nucleic acid bases strongly absorb and at 280 nm proteins and other contaminating materials absorb light.

For pure DNA the 260/280 is 1.8 and for pure RNA the 260/280 is 2.0. Values significantly lower than these indicate contamination of the sample, which may require the customer to perform a further sample clean-up.

The above User Collect statement causes the system to first take a reading at 260 nm, then change to 280 nm and take a second reading. The system then divides the reading at 260 nm by the reading at 280 nm and prints the result in the Report area.

260 nm/280 nm ratio, with background correction at 320 nm

\[
\frac{\text{Read}(260) - \text{Read}(320)}{\text{Read}(280) - \text{Read}(320)}
\]

Neither nucleic acids or proteins absorb light at 320 nm, so this wavelength is used to correct the readings taken at both 260 nm and 280 nm. The same ratio values of 1.8 for pure DNA and 2.0 for pure RNA apply for 320 nm background corrected 260/280 data.

The above statement causes the instrument to perform a 260/280 ratio, with 320 nm background correction. The system first takes readings at 260 nm, 280 nm and 320 nm. It then subtracts the reading at 320 nm from the readings at 260 and 280 nm, and divides the result. The final answer is printed in the Report area.

Warburg-Christian calculations

Nucleic acid and protein concentrations can be determined, in mg/mL, using the factors and equations of Warburg and Christian. Warburg and Christian determined these equations and factors based on the relationship between the molar absorptivity values of a yeast RNA and protein called enolase, determined at 260 nm and 280 nm with background correction at 320 nm.

The User Collect statements for the Warburg-Christian calculations are:

Protein concentration
\[
\left( 1552 \times (\text{Read}(280) - \text{Read}(320)) \right) - \left( 757.3 \times (\text{Read}(260) - \text{Read}(320)) \right) \right) \] Protein Conc

This expression causes the instrument to take readings at 260 nm, 280 nm and 320 nm. The mathematical operations defined in the expression are then performed on each of these readings and the result is printed in the Report area.

Note
The single quote at the end of the equation indicates a comment that is ignored by the instrument.

Nucleic acid concentration
\[
\left( -36.0 \times (\text{Read}(280) - \text{Read}(320)) \right) + \left( 62.9 \times (\text{Read}(260) - \text{Read}(320)) \right) \right) \] 'Nucleic Acid Conc

This expression causes the instrument to take readings at 260 nm, 280 nm and 320 nm. The mathematical operations defined in the expression are then performed on each of these readings and the result is printed in the Report area.
References:

**Bio-/Chemi-luminescence/Phosphorescence Options—Simple Reads**

Use the **Bio-/Chemi-luminescence/Phosphorescence Options** dialog to set up the parameters associated with these two measurement modes.

To access the **Bio-/Chemi-luminescence/Phosphorescence Options** dialog, select the **Options** button from the Cary page.

**Note**
The **Options** button is only visible on the Cary page if you have selected **Bio-/Chemi-luminescence** or **Phosphorescence** as the **Data Mode**.

**Dialog controls**

The **Bio-/Chemi-luminescence/Phosphorescence Options** dialog comprises the following options:

- **Total decay time**
  - Enter, in seconds, the total decay time for your sample. The total decay time is the time that it takes for the phosphorescence signal to decay to dark levels (zero). The lamp will not flash again until the total decay time has elapsed. The range is 0.001–1800.000 s.
  - **Note**
    - This option is not available in Bio-/Chemi-luminescence mode.

- **No. of flashes**
  - Enter the number of lamp flashes required, which is how many times the lamp flashes consecutively before the delay time is counted.

  If you are measuring phosphorescence samples that have a relatively long decay time (that is, a number of seconds), you can setup the Cary Eclipse to pulse the lamp multiple times, as this will increase the intensity of the phosphorescent signal. For short lived phosphorescent samples, multiple flashes will not impact on the intensity of the signal, and therefore only one flash is required.

  **How many flashes should I perform?**

  **Note**
Example of the Bio-/Chemi-luminescence/Phosphorescence Options

![Phosphorescence options](image)

**Delay time**
Enter, in milliseconds, the delay time for your sample. The delay time is the time that elapses between the last flash and the beginning of data collection. The default value is 0.100 ms, which is the time it takes for the lamp intensity to decay to zero. Setting a delay time ensures that there is no lamp effects in the phosphorescence decay curve. The range is 0.000–5000.000 s.

**Gate time**
Enter, in milliseconds, the gate time for your sample. The gate time is the overall reading time of the emission signal. The range is from 10µs–10s.

[Understanding Average Time and Gate Time for Phosphorescence scans](#)
[Understanding Gate Time for Bio-/Chemi-luminescence scans](#)

See also:
[Cary page](#)

**Understanding Gate Time for Bio-/Chemi-luminescence scans**

**Simple Reads**
**Gate Time** is set on **Options** page, which is accessible from the **Cary** page.

The following diagram highlights the **Gate Time** setting.

The **Gate Time** is set to 10 and the **Intensity** is equal to $I_T$.

![Diagram of Gate Time and Intensity](image)

See also:
- Understanding Average Time and Gate Time for Phosphorescence scans
- Simple Reads Application
- Cary page
- Options dialog
- User Collect examples

### Understanding Average Time and Gate Time for Phosphorescence—Simple Reads

For Phosphorescence scans, there is a relationship between Average Time (the time for which data is averaged) and Gate Time (the overall reading time of the emission signal).

Averaging is calculated as follows:

The intensity readings are taken from different decays, where:

**Number of Decays** = **Average Time/Gate Time**

**Note:**

**Gate Time** is not used if you select **Fluorescence** as the **Data Mode**.

**Average Time** is set on the **Cary** page of the **Setup** dialog. **Gate Time** is set on **Options** page, which is accessible from the **Cary** page.

The following diagrams highlight the relationship between **Average Time** and **Gate Time**.
Example 1

When the Average Time and Gate Time are set to the same value:

<table>
<thead>
<tr>
<th>Average Time/Gate Time</th>
<th>1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intensity</td>
<td>1 (Area I₁)</td>
</tr>
</tbody>
</table>

where:

- Total Decay Time: 20 ms
- Delay Time: 10 ms
- Average Time: 5 ms
- Gate Time: 5 ms

Example 2

When the Average Time is set to five times the Gate Time:

<table>
<thead>
<tr>
<th>Average Time/Gate Time</th>
<th>5</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intensity</td>
<td>( A₁ + A₂ + A₃ + A₄ + A₅ )</td>
</tr>
<tr>
<td></td>
<td>5</td>
</tr>
</tbody>
</table>
where:

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total Decay Time</td>
<td>20 ms</td>
</tr>
<tr>
<td>Delay Time</td>
<td>10 ms</td>
</tr>
<tr>
<td>Average Time</td>
<td>5 ms</td>
</tr>
<tr>
<td>Gate Time</td>
<td>25 ms</td>
</tr>
</tbody>
</table>

See also:
How many flashes should I perform?—Simple Reads

The **Number of Flashes** option is set in the **Bio-/Chemi-luminescence/Phosphorescence Options** dialog. To access the dialog, select the **Options** button from the **Cary** page.

**Note**

The **Options** button is only visible on the **Cary** page if you have selected **Bio-/Chemi-luminescence** or **Phosphorescence** as the **Data mode**.

If you are measuring phosphorescence samples that have a relatively long decay time (that is, a number of seconds), you can setup the Cary Eclipse to pulse the lamp multiple times, as this will increase the intensity of the phosphorescent signal (refer to Example 1, below). For short lived phosphorescent samples, multiple flashes will not impact on the intensity of the signal, and therefore only one flash is required (refer to Example 2, below).

**Example 1**

The above example shows that the sample does not decay before the instrument lamp flashes again. Therefore setting multiple flashes will result in an increase in the intensity of the signal.
Example 2
The above example shows that the sample decays in less time than the time it takes the instrument to flash the lamp again. Therefore, setting more than 1 flash, in the **No. of Flashes** field will not increase the intensity of the signal.

See also:
- Bio-/Chemi-luminescence/Phosphorescence Options

### Options—Simple Reads

Use the **Options** page to set up the Cary Eclipse filters and the PMT Detector voltage.

The **Options** page is accessible from the **Setup** dialog by selecting the **Options** tab. To access the **Setup** dialog, select the **Setup** button from the Simple Reads Application window, or select the **Setup** command from the **Setup** menu.

<table>
<thead>
<tr>
<th><strong>Excitation Filter</strong></th>
<th>Set the excitation filter for the Simple Reads measurement. To do this, select the down arrow to the right of the field and select an option from the list.</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Emission Filter</strong></td>
<td>Set the emission filter for the Simple Reads measurement. To do this, select the down arrow to the right of the field and select an option from the list.</td>
</tr>
<tr>
<td><strong>PMT Detector voltage</strong></td>
<td>Set the EHT or sample detector PMT. To do this, select one of the following:</td>
</tr>
<tr>
<td><strong>Low</strong></td>
<td>Sets the PMT Detector voltage to a pre-defined value of 400 v.</td>
</tr>
<tr>
<td><strong>Medium</strong></td>
<td>Sets the PMT Detector voltage to a pre-defined value of 600 v.</td>
</tr>
<tr>
<td><strong>High</strong></td>
<td>Sets the PMT Detector voltage to a pre-defined value of 800 v.</td>
</tr>
<tr>
<td><strong>Manual</strong></td>
<td>Enables you to enter any PMT voltage in the adjacent Volts field.</td>
</tr>
<tr>
<td><strong>Volts</strong></td>
<td>Enter the PMT voltage in the adjacent field. Note that this option is only available if you have selected <strong>Manual</strong> as the PMT Detector voltage.</td>
</tr>
</tbody>
</table>

**Note**
The reference detector EHT is assigned according to the instrument settings, and will be determined when you select **OK** to exit the **Setup** dialog. The intensity may be doubled with every 10% increase in EHT on the sample detector.

See also:
- Setup
- Simple Reads Application

### Reading samples—Simple Reads
Go To Wavelength—Simple Reads

Use the **Go To Wavelength** dialog to temporarily change the instrument to a new excitation or emission wavelength. You would mainly use this if you wanted to quickly go to a wavelength to check the intensity reading of the sample. The lamp will flash continuously in this mode. This option is only available if the instrument is not currently collecting data.

To access the **Go To Wavelength** dialog, select the **Go To Wavelength** command from the **Commands menu**. You can also select **F4** to open the dialog.

**Note**
This is not the wavelength used in the application method. If your method uses a wavelength that is different from the **Go To Wavelength**, then when you select **Read** the system will change to the wavelength selected. (This is set on the **Cary** page of the **Setup** dialog.)

**Excitation**

**Wavelength**
Enter the new wavelength in this numeric entry field. Select **Go To** or press ENTER and the instrument will change to the new value.

**Zero order**
If this check box is selected, then the Cary Eclipse will be driven to 0 nm (white light). You can then use this light for aligning accessories.

**Emission**

**Wavelength**
Enter the new wavelength in this numeric entry field. Select **Go To** or press ENTER and the instrument will change to the new value.

**Zero order**
If this check box is selected, then the Cary Eclipse will be driven to 0 nm (white light). You can then use this light for aligning accessories.

**Go To**
Select **Go To** once you have entered a new wavelength, to change the instrument to the entered value.

See also:
**Function Keys**

Rapid Result—Simple Reads

Use the **Rapid Result** dialog to take a reading of a blank and sample/s, without having to change the current application method. The result is printed in the Report area.

To access the **Rapid Result** dialog, select the **Rapid Result** command from the **Commands menu**. You can also select **F6** to open the dialog.
**Zero—Simple Reads**

Use the **Zero** button to zero the current intensity value. The intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before commencing a data collection if you want your data to start at 0 intensity. The Cary Eclipse will prompt you to load a blank before performing the zero.

Before you select **Zero**, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.

When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the Simple Reads Application window.

Select the **Clear Zero** command from the **Commands** menu to remove zero correction and thus remove the subtraction effect.

---

**Excitation**

*Wavelength*

Enter the wavelength at which you want to take a reading.

**Zero order**

Select this option to set the excitation monochromator to zero order (white light).

**Emission**

*Wavelength*

Enter the wavelength at which you want to take a reading.

**Zero order**

Select this option to set the excitation monochromator to zero order (white light).

**Zero**

Click **Zero** to change the wavelength to the entered value and measure a blank.

**Result**

Click **Result** to change the wavelength to the entered value and measure a sample. The X,Y results are corrected for the blank.

---

**Commands menu—Simple Reads**

Use the **Commands** menu to access the commands for performing a Simple Read.

Commands in this application can be accessed both from the menu items at the top of the main window and from the application buttons. Alternatively you can use function key shortcuts for some of the commands. These appear next to the item in the **Commands** menu. Note that not all commands appear as both button and menu items.

**Hot Tip**

By becoming familiar with these commands, you can remove the **Command** buttons from the Simple Reads window. This will automatically enlarge the Report area, making viewing results more convenient.
How to remove or display the command buttons

The following commands are accessible from the **Commands** menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
</table>
| **Read**              | Select this to commence a Simple Reads measurement using the current Setup parameters. This command is also accessible by pressing F9.  
  **Note**             | If the **Connect** command is displayed in place of the **Read** command, this indicates that another Cary Eclipse application is in use and has control of the Cary Eclipse instrument. To make the Simple Reads application active so that you can perform a read, you will need to select **Connect**. However, you will not be able to select **Connect** if another application is collecting data. |
| **Clear Zero**        | Select this option to clear the **Zero** command. When you select the **Zero** command (from the **Commands** menu or by pressing F5), the intensity at that time will be reset to zero and all subsequent results will have the intensity value that was reset to zero subtracted from them. Selecting **Clear zero** cancels this zeroing (and thus subtraction) effect. |
| **Zero Order**        | **Excitation**  
  Select this command to set the excitation monochromator to zero order (white light).  
  **Emission**  
  Select this command to set the emission monochromator to zero order (white light). |
| **Reset**             |                                                                                                                                                                                                            |
| **Rapid Result**      | Select this to display the **Rapid Result** dialog where you can read a blank and multiple sample readings without changing the application method.  
  This command is also accessible by pressing F6. |
| **Zero**              | Select this to zero the current intensity value. The intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before selecting **Start** if you want your data to start at 0 intensity.  
  This command is also accessible by pressing F5.  
  **Note**             | Before you select **Zero**, ensure that you have placed the blank solution in the cell positions that are currently in the cell path. |
| **Go to**             | Select this to display the **Go To Wavelength** dialog and change the instrument to a new wavelength.  
  This command is also accessible by pressing F4. |
| **ADL Command**       | Select this to display the **ADL Command Line** window where you can execute an ADL command or ADL program. (You can also press Alt-Y to display this dialog.)  
  This command is also accessible by pressing CTRL+Y. |
| **Re-run ADL**        | Select this to run the ADL command or program in the ADL Command Line window again. (You can also press Alt-X to rerun programs.)  
  This command is also accessible by pressing CTRL+X. |
How to perform a measurement at a single wavelength—Simple Reads

This demonstrates how to read samples at a single wavelength.

Follow the steps through, adjusting them to meet the requirements of your particular data collection.

The procedure consists of the following steps:

- Start the application
- Set up instrument parameters
- Read the sample
- Print the results

Start the application

Step 1

On the Windows Taskbar click Start, then point to Programs, Cary Eclipse and select Simple Reads to open the Simple Reads application.

**Note**

If you are running a GLP system you may be prompted to enter a password before accessing the Simple Reads application.

Step 2

Make sure that the sample compartment is clear (or you have a blank in position) and zero the instrument by selecting the Zero button.

Set up the instrument parameters

Step 3

Select the Setup button or select the Setup command from the Setup menu to display the Cary page of the Setup dialog.

Step 4

In the Instrument Setup group, select the Data Mode you require. Choose from Fluorescence, Bio-/Chemi-luminescence or Phosphorescence.

Step 5
In the **Wavelength Setup** group, enter the required excitation and emission wavelengths at which you want to perform the read.

**Step 6**

Select the **OK** button. The instrument will change to the new wavelength.

**Read the sample**

**Step 7**

Insert the sample into the sample compartment.

**Step 8**

Wait while the instrument changes to the specified wavelength. The current wavelength is displayed in the Intensity status display at the top and right of the application window.

**Step 9**

Select the **Read** button to perform the read. The result will be displayed in the Report area.

**Print the results**

**Step 10**

To print the contents of the Report area, select the **Print** button.

See also:

[How to perform measurements at two wavelengths and subtract the result](#)

**How to perform measurements at two wavelengths and subtract the result—Simple Reads**

With the User Collect function you can extend the Simple Reads application to carry out simple mathematical functions on the results of readings performed on a sample at various wavelengths.

To do this:

- [Start the application](#)
- [Set up instrument parameters](#)
- [Read the sample](#)
- [Print the results](#)

**Start the application**

**Step 1**

On the Windows Taskbar click **Start**, then point to **Programs, Cary Eclipse** and select **Simple Reads**
to open the Simple Reads application.

**Note**
If you are running a GLP system you may be prompted to enter a password before accessing the Simple Reads application.

Step 2
Make sure that the sample compartment is clear (or you have a blank in position) and zero the instrument by selecting the **Zero** button.

**Set up instrument parameters**

Step 3
Select the **Setup** button or select the **Setup** command from the **Setup** menu to display the **Cary** page of the **Setup** dialog.

Step 4
In the **Instrument Setup** group, select the **Data Mode** you require. Choose from **Fluorescence**, **Bio-/Chemi-luminescence** or **Phosphorescence**.

Step 5
Select **User Collect**. The Em. Wavelength field will change to enable you to enter a simple Read command. You can also select a default command from the drop down list. To do this, select the arrow on the right side of this field.

**User collect example:**

```
Read(340, 400)-Read(340, 450)
This reads the intensity at Excitation monochromator = 340, Emission monochromator = 400 and subtracts the intensity read at Excitation monochromator = 340 and Emission monochromator = 450. This reading is performed at each data point.
```

Step 7
Set the slit widths of the excitation and emission monochromators in the **Ex. Slit** and **Em. Slit** fields.

Step 8
Enter the averaging time, which is the time for which data is averaged.

Step 9
Select the **OK** button. The User Collect statement will appear in the Report area.

**Read the sample**
Step 10

Insert the sample into the sample compartment.

Step 11

Select the Read button to perform photometric measurements on the sample at the specified wavelength.

The value for each sample read will appear in the Report area as well as the result of the User Collect equation.

Print the results

Step 12

To print the contents of the Report area, select the Print button.

See also:
How to perform a measurement at a single wavelength

Viewing run progress and results—Simple Reads

- View menu
- Edit menu
- Print Preview dialog
- How to open a previously stored report
- How to use advanced reporting options

View menu—Simple Reads

Use the View menu to show or hide toolbars, buttons, reports and other items in the Simple Reads Application window.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Toolbars</strong></td>
<td>Show or hide the following toolbars or toolbar items:</td>
</tr>
<tr>
<td><strong>Report</strong></td>
<td>View the Report toolbar which contains shortcuts for common report editing functions such as cutting and pasting text, changing text fonts and preview the report.</td>
</tr>
<tr>
<td><strong>Text Labels</strong></td>
<td>View the text labels associated with the Report toolbar. Note that the Report toolbar must be visible for these labels to be displayed.</td>
</tr>
<tr>
<td><strong>Buttons</strong></td>
<td>Select this command to toggle the left-hand button display (button area) on and off.</td>
</tr>
<tr>
<td><strong>Report</strong></td>
<td>Select this command to view the Simple Read report.</td>
</tr>
<tr>
<td><strong>ADL</strong></td>
<td>Select this command to toggle the ADL Editor on and off.</td>
</tr>
</tbody>
</table>
**Edit menu—Simple Reads**

Use the **Edit** menu to perform simple text functions such as copying, pasting and formatting text.

**Hot Tip**

The buttons shown below can be made visible in the Simple Reads Application window by selecting the **View** menu, pointing to **Toolbars** and then selecting **Report**.

<table>
<thead>
<tr>
<th>Command</th>
<th>Button</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Undo</td>
<td><img src="#" alt="Undo" /></td>
<td>If available, this undoes the last action. This command is also accessible by pressing CTRL+Z.</td>
</tr>
<tr>
<td>Cut</td>
<td><img src="#" alt="Cut" /></td>
<td>Select this command to remove the highlighted text and place it on the Windows Clipboard. This command is also accessible by pressing CTRL+X.</td>
</tr>
<tr>
<td>Copy</td>
<td><img src="#" alt="Copy" /></td>
<td>Select this command to copy the highlighted text onto the Windows Clipboard. This command is also accessible by pressing CTRL+C.</td>
</tr>
<tr>
<td>Paste</td>
<td><img src="#" alt="Paste" /></td>
<td>Select this command to insert the contents of the Windows Clipboard (text only) at the cursor. Any highlighted text will be replaced. This command is also accessible by pressing CTRL+V.</td>
</tr>
<tr>
<td>Delete</td>
<td><img src="#" alt="Delete" /></td>
<td>Select this command to remove selected text in the Report area. This command is also accessible by pressing DEL. <strong>Note</strong> This option is only available once you select <strong>Edit Report</strong> and there is text highlighted in the Report area.</td>
</tr>
<tr>
<td>Select All</td>
<td></td>
<td>Select this option to highlight all text in the Report area. This command is also accessible by pressing CTRL+A.</td>
</tr>
</tbody>
</table>
Print Preview—Simple Reads

Use the Print Preview dialog to view the contents and layout of current report before you print it.

To access the Print Preview dialog, select the Print preview command from the File menu.

You can reduce or enlarge the print preview on screen. Click the left mouse button in the preview area to toggle through the five available views. Click the right mouse button in the preview area to return to the full page view.

Use the scroll bars to the right and bottom of the report to move around the preview.

**Note**
The scroll bars are not available for the full page view.

<table>
<thead>
<tr>
<th>Command</th>
<th>Button</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td>First arrow</td>
<td><img src="arrow.png" alt="First Arrow" /></td>
<td>Use this arrow to view the first page of the report.</td>
</tr>
<tr>
<td>Previous arrow</td>
<td><img src="arrow.png" alt="Previous Arrow" /></td>
<td>Use this arrow to view the previous page of the report.</td>
</tr>
<tr>
<td>Next arrow</td>
<td><img src="arrow.png" alt="Next Arrow" /></td>
<td>Use this arrow to view the next page of the report.</td>
</tr>
<tr>
<td>Last arrow</td>
<td><img src="arrow.png" alt="Last Arrow" /></td>
<td>Use this arrow to view the last page of the report.</td>
</tr>
<tr>
<td>Print</td>
<td><img src="print.png" alt="Print" /></td>
<td>Use this to display the Windows Print dialog where you can choose the printer and pages to print as well as the number of copies you wish to print.</td>
</tr>
</tbody>
</table>

Q. Why are all of the arrow buttons grayed out?
A. All of the buttons will be grayed out if the report has only one page of data.
How to open a previously stored report—Simple Reads

Step 1

Select **Open**... from the **File** menu to display the Windows **Open** dialog.

Step 2

Click the down arrow at the right of the **Files of type** list box and select **Report (*.FRSR)** from the list. A list of previously stored report files will be displayed.

Step 3

Select the report file you require and select **Open**. The system will display the report information in the Report area.

See also:

How to use advanced reporting options

How to use advanced reporting options—Simple Reads

This outlines ways to fully customize your reports. Options available include:

- Including header and footer information
- Including a logo
- Editing the text in the report
- Viewing the report on screen before printing
- Cutting and pasting to a word processor

Including header and footer information

You can have text appearing at the top and bottom of every page in your report. To specify what the text should be, do the following:

Step 1

On the Windows Taskbar, click the Windows **Start** button, point to **Programs, Cary Eclipse** and then select **System Information**. The System Information application appears with the Company page displayed.

Step 2

Enter the header and footer information in the **Header and Footer** fields.

Step 3

Click **OK** to implement your changes.

Including a logo
You can include a logo on the first page of your report. To specify what the logo should be, do the following:

Step 1

On the Windows Taskbar, click the Windows **Start** button, point to **Programs, Cary Eclipse** and then select **System Information**. The System Information application appears with the Company page displayed.

Step 2

Enter a logo using the **Add Logo** button. This logo will always be included on the front page of reports.

Step 3

Click **OK** to implement your changes.

**Q.** Why isn't the logo I selected included in my report?

**A.** If you are running the Cary Eclipse software under Windows NT you need to be logged on as an Administrator in order to specify the logo in the System Information application.

**Editing the text in the report**

You can edit any of the text in the Report area. Just select **Edit Report** from the **Edit** menu. You can then add, delete and change text in the Report area.

**Q.** The **Edit Report** option is not available, why not?

**A.** You do not have reporting editing privileges set in the GLP Administration application. The Administrator of the system will have to change your privileges before you can edit reports.

**Viewing the report on the screen before printing**

You can view a report before printing it. To do this, from the **File** menu select **Print Preview**. You can then ensure that the report format is correct before you print it.

**Hot Tip**

Use the arrow keys at the bottom of the **Print Preview** screen to change pages.

**Cutting and pasting to a word processor**

To have complete control over how your report looks you may wish to copy it to a word processor program, such as Microsoft Word. To do this:

Step 1

Open a report.

Step 2
From the **Edit** menu, choose **Edit Report**.

**Step 3**

Click in the Report area and highlight the text you want to export to your word processor.

**Step 4**

Right-click on the Report area and select **Copy** from the menu.

**Step 5**

Start up your word processor and use the Paste function (usually under the **Edit** menu) to place the text into a document.

**File Management—Simple Reads**

- [File menu](#)
- [Open dialog](#)
- [Save As dialog](#)
- [Filename extensions](#)

**File menu—Simple Reads**

Use the **File** menu to open, save or print a report, or exit the Simple Reads application.

[Hot Tip](#)

Highlight a report file (*.FRSR) in Windows® Explorer and simply drag and drop it onto the application to open it. The data will be displayed in the Report area.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td>New…</td>
<td>Select this command to clear the current report and create a new one. This command is also accessible by pressing CTRL+N.</td>
</tr>
<tr>
<td>Open method…</td>
<td>Select this command to display the <strong>Open</strong> dialog and load a stored method file. Method files have the file extension ‘.FMSR’. This command is also accessible by pressing CTRL+M.</td>
</tr>
<tr>
<td>Open data…</td>
<td>Select this command to display the <strong>Open</strong> dialog and load a stored data file. Data files have the file extension ‘.FMSR’. This command is also accessible by pressing CTRL+O.</td>
</tr>
<tr>
<td>Save method as…</td>
<td>Select this command to display the <strong>Save As</strong> dialog and store the current method. Methods are saved with the extension ‘.FMSR’. This command is also accessible by pressing CTRL+S.</td>
</tr>
<tr>
<td>Save data as…</td>
<td>Select this command to display the <strong>Save As</strong> dialog and store the current report. Data is saved with the extension ‘.FMSR’.</td>
</tr>
<tr>
<td>Print setup…</td>
<td>Select this command to display the Windows Print Setup dialog where you can select the printer, printer page size and orientation.</td>
</tr>
</tbody>
</table>
Use the Open dialog to open a method or report file previously saved by the application.

To access the Open dialog, select the Open method or Open data command from the File menu. You can also select CTRL+M (for a method) or CTRL+O (for data) open the dialog.

| Print preview… | Select this command to display the Print Preview dialog to view the current report before you print it. |
| Print… | Select this command to display the Windows Print dialog where you can choose the printer and pages to print as well as the number of copies you wish to print. This command is also accessible by pressing CTRL+P. |
| Explorer… | Select this command to run Windows Explorer. This may help you locate report files. |
| History list | This displays a list of the four most recently opened files. |
| Exit | Select this command to close the Simple Reads application. You will be prompted if there is any unsaved data in the report. |

See also:
Open
Save As
Print Preview

Open—Simple Reads

Use the Open dialog to open a method or report file previously saved by the application.

To access the Open dialog, select the Open method or Open data command from the File menu. You can also select CTRL+M (for a method) or CTRL+O (for data) open the dialog.

**Look in**
This field lists the available folders and files. Click the down arrow on the right of this field to see how the current folder fits in the hierarchy on your computer.

To view the contents of a folder, select it. The folders and files in the selected folder are displayed the list box below.

**Note**
To open the folder one level higher, select on the toolbar.

**Look in list box**
This lists the folders and files in the selected location. To view the contents of a folder, double-click it. The following buttons of the Look in toolbar can assist you in locating the appropriate file.

- Click to create a new folder. Type the name of the new folder in the highlighted text entry field that appears and press Enter. Then double-click on the new folder to open it.
- Click to display the directory structure of the drive.
- Click to display the files as a list.
- Click to display files as a detailed list including the file size, file type and the date the file was last modified.
- Click to preview the selected trace before you load it into the application.

**File name**
Enter the name of the file you want to open in this field. The easiest way to do this is to find the file in the Look in list box and then select the file. However, you can also type the full path of a file. Then select the Open button to open the file.
Files of type  Click on the arrow on the right of this field to view a list of all application files that can be opened in the list box.

This is useful for reducing the list of files displayed to only those files in which you are interested.

*Methods (*.FMSR), Report (*.FRSR)*
In the Simple Reads application, you may open Method or Report files only.

Open  This button opens the highlighted file/s or the files listed in the File name field.

See also:  
Filename extensions

**Save As—Simple Reads**

Use the Save As dialog to save the method or data.

To access the Save As dialog, select the **Save method as** or **Save data as** command from the File menu. You can also select CTRL+S (for a method) to open the dialog.

**Save in**  This field lists the available folders and files. Click the down arrow on the right of this field to see how the current folder fits in the hierarchy on your computer.

To view the contents of the folder, select it. The folders and files in the selected folder are displayed the list box below.

**Note**

To open the folder one level higher, select  on the toolbar.

**Save in list box**  This lists the folders and files in the selected location. To view the contents of a folder, double-click it. The following buttons of the Look in toolbar can assist you in locating the appropriate file.

- Click to create a new folder. Type the name of the new folder in the highlighted text entry field that appears and press ENTER. Then double-click on the new folder to open it.
- Click to display the directory structure of the drive.
- Click to display the files as a list.
- Click to display files as a detailed list including the file size, file type and the date the file was last modified.
- Click to preview the selected trace before you load it into the application.

**File name**  Enter the name of the file you want to save in this field. You can also type the full path of a file then select the Save button to save the file.

**Files of type**  Use this field to specify the type of file you are saving. Click on the arrow on the right of this field to display the list of all the available file types that this application can recognize.

*Methods (*.FMSR), Report (*.FRSR)*
In the Simple Reads application, you may save Method or Report files only.
Save

Select **Save** to save the file currently present in the **File name** field in the directory specified in the **Look in** field.

![See also: File menu Filename extensions](image)

### Filename extensions—Simple Reads

The Cary Eclipse system uses the filename extension to distinguish between data types (e.g. method or data) and also to differentiate applications. (e.g. Concentration or Kinetics).

The Syntax for file extensions is:

<table>
<thead>
<tr>
<th>Letter</th>
<th>Represents</th>
<th>Available Options</th>
</tr>
</thead>
<tbody>
<tr>
<td>First</td>
<td>Type of analysis</td>
<td>F Fluorescence</td>
</tr>
<tr>
<td>Second</td>
<td>Type of file</td>
<td>B Batch, D Data, G Graph Template, M Method, R Report</td>
</tr>
<tr>
<td>Third and fourth</td>
<td>Application identifier</td>
<td>AB Advanced Reads, CN Concentration, KN Kinetics, LT Lifetimes, RT Ratio, SR Simple Reads, SW Scan, TM Thermal, VO Validate</td>
</tr>
</tbody>
</table>

The following files may be opened and saved in the Simple Reads Application:

<table>
<thead>
<tr>
<th>Extension</th>
<th>Available action</th>
<th>File type</th>
</tr>
</thead>
<tbody>
<tr>
<td>*.FMSR</td>
<td>Open/Save</td>
<td>A <em>method</em> file (*.FMSR) consists of the software setup by which data is collected.</td>
</tr>
<tr>
<td>*.FRSR</td>
<td>Open/Save</td>
<td>A <em>report</em> file (*.FRSR) contains the report generated by the Cary Eclipse about the data.</td>
</tr>
</tbody>
</table>

**Note**

Selecting **All Files** in the Windows Open dialog will list all available file types.

![See also: Open Save As](image)
Software and Screen features—Simple Reads

- Report toolbar
- Status line
- Function keys
- How to remove or display the command buttons

Report Toolbar—Simple Reads

The Report toolbar is part of the Simple Reads Application. To display the Report toolbar, click the View menu, point to Toolbars and then select Report.

<table>
<thead>
<tr>
<th>Button</th>
<th>Command</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td>![Edit]</td>
<td>Edit</td>
<td>Select this option to edit the report.</td>
</tr>
<tr>
<td>![Undo]</td>
<td>Undo</td>
<td>If available, this undoes the last action.</td>
</tr>
<tr>
<td>![Cut]</td>
<td>Cut</td>
<td>Select this command to remove the highlighted text and place it on the Windows Clipboard.</td>
</tr>
<tr>
<td>![Copy]</td>
<td>Copy</td>
<td>Select this command to copy the highlighted text onto the Windows Clipboard.</td>
</tr>
<tr>
<td>![Paste]</td>
<td>Paste</td>
<td>Select this command to insert the contents of the Windows Clipboard (text only) at the cursor. Any highlighted text will be replaced.</td>
</tr>
<tr>
<td>![Delete]</td>
<td>Delete</td>
<td>Select this command to remove selected text in the Report area.</td>
</tr>
<tr>
<td>![Font]</td>
<td>Font</td>
<td>This enables you to change the font of new or selected text in the report. For existing text, simply highlight the text in the report, select Font and make your changes in the Font dialog. When you select OK, the appearance of the highlighted text in the report will alter to reflect your font changes.</td>
</tr>
<tr>
<td>![Preview]</td>
<td>Preview</td>
<td>Click this button to open the Print Preview dialog where you can view the report as it will be printed.</td>
</tr>
</tbody>
</table>

Status line—Simple Reads

As displayed on the screen from left to right:

- **Hardware Status**
  This field displays status messages indicating the current instrument activity.
- **Sequence Information**
  This field displays status messages indicating the current system activity.
- **Current Results**
  This field displays information on the last reading.
**Function key—Simple Reads**

The function keys at the top of your keyboard perform basic Cary Eclipse operations simply and quickly.

<table>
<thead>
<tr>
<th>Function key</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td>F4</td>
<td>Press this key to display the Go To Wavelength dialog. This command is also accessible by selecting the Commands menu and then selecting Go to.</td>
</tr>
<tr>
<td>F5</td>
<td>Press F5 to perform an instrument zero. The current intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before selecting Start if you want your data to start at 0 intensity. This command is also accessible from the Commands menu or by selecting the Zero button.</td>
</tr>
<tr>
<td>Note</td>
<td>Before you select Zero, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.</td>
</tr>
<tr>
<td></td>
<td>Select the Clear Zero command from the Commands menu to remove zero correction.</td>
</tr>
<tr>
<td>F6</td>
<td>Press this key to display the Rapid Result dialog where you can read a blank and multiple sample readings without changing the application method. This command is also accessible by selecting the Commands menu and then selecting Rapid Result.</td>
</tr>
<tr>
<td>F9</td>
<td>If the application is online, press this key to perform a read. This is equivalent to selecting the Start button. If the application is not online, press F9 to first connect to the Cary Eclipse before you can press F9 to start the data collection. This command is also accessible by selecting the Start button.</td>
</tr>
<tr>
<td>Note</td>
<td>You will not be able to perform a Start if another application is collecting data.</td>
</tr>
</tbody>
</table>

*See also: [Rapid Result](#)*

**How to remove or display the command buttons—Simple Reads**

The command buttons (Setup, Zero, Clear Report and Print) appear on the left side of the Simple Reads Application window.

**To remove the command buttons**

**Step 1**

From the Simple Reads Application window, select the View menu.

**Step 2**
Clear the **Buttons** menu command.

The buttons will be removed from the application screen, giving you more room to display reports.

**To show the command buttons**

Step 1

From the Simple Reads Application window, select the **View** menu.

Step 2

Select the **Buttons** menu command.

The buttons will re-appear in the application screen.

**ADL—Simple Reads**

- [ADL Command Line](#)
- [ADL Hooks](#)
- [How to insert ADL commands](#)

**ADL Command Line—Simple Reads**

**Introduction to ADL**

Use the ADL Command Line to run Single-line or Multi-line Applications Development Language (ADL) commands that enable you to perform functions over and above that of the existing program functionality.

ADL is not a general purpose programming language. It has been developed specifically by Varian to meet the needs of spectroscopists. It can be used to handle all the data collected from the Cary Eclipse—both photometric values and entire data collections. ADL programs that you develop should be stored with the extension 'ADL'.

To access the ADL Command Line, select the ADL Command item from the **Commands menu**. You can also press ALT+Y to open the dialog.

**Note**

Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.

**Software controls**

**Single Line** Selecting this lets you enter ADL commands on a single line into the Command Line.
The ADL Hooks command is accessible from the View menu.

Instead of writing a full ADL program you can use ADL hooks to link into an existing application. This is particularly useful if you want to make small changes to the way an application works, e.g. you want to pop up a dialog for the user to enter some specific data before the run starts. ADL hooks are links at specific points in an application at which you can run an ADL program. For example, ADL hooks have been supplied to run the following ADL programs:

Q. Can I enter more than one ADL command on a single line?
A. Yes, provided that the commands are separated by a semicolon ‘;’.

Multi-line
The Multi-line capability enlarges the Command Line entry box. Use the Multi-line capability to create a more complex ADL program by adding a series of commands over multiple lines. When Multi-line mode is selected, the Quick Reference option becomes available.

Quick Reference
Select Quick Reference to open the Quick Reference dialog where you can easily add a command in the correct syntax to your ADL program. This option is only available for Multi-line commands.

Show me How to insert ADL commands using the Quick Reference.

A detailed description of all the commands in the Quick Reference is available by selecting the Help button on the Quick Reference dialog.

Browse
Click Browse to browse your release CD for ADL programming examples that you can load and run. Alternatively, you can select and load ADL programs that you may have created.

Command Line (entry box)
This entry box enables you to use ADL commands. You can enter single ADL commands into this field, or you can enter a series of ADL commands. If you have the entry type set to Single Line, you can review previous ADL entries by selecting the down arrow to the right of the list box.

The ADL commands in the Command Line are executed when you select OK.

Note
On entering this window, the Cary Eclipse will recall the last command or program executed from the Command Line entry box. The ADL commands entered in the last executed program are stored in the Adl folder (accessed from the Varian/Cary Eclipse softwarefolder) as CaryEquation.ADL.

Hot Tip
You can rerun the ADL commands in the Command Line entry box (CaryEquation.ADL) from any screen in the Simple Reads application by pressing Alt-X or select Rerun ADL from the Commands menu. The

See also:
ADL Hooks

ADL Hooks—Simple Reads Application

The ADL Hooks command is accessible from the View menu.

Instead of writing a full ADL program you can use ADL hooks to link into an existing application. This is particularly useful if you want to make small changes to the way an application works, e.g. you want to pop up a dialog for the user to enter some specific data before the run starts. ADL hooks are links at specific points in an application at which you can run an ADL program. For example, ADL hooks have been supplied to run the following ADL programs:
Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.

<table>
<thead>
<tr>
<th>Hook</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>SR_Startup.ADL</td>
<td>This is run when the Simple Reads application starts. For this to to work the application must be started from a batch or method file that has ADL Hooks on.</td>
</tr>
<tr>
<td>SR_PreSeq.ADL</td>
<td>This runs when the Start button is selected.</td>
</tr>
<tr>
<td>SR_PostSeq.ADL</td>
<td>This runs at the end of the Start button sequence.</td>
</tr>
<tr>
<td>SR_Online.ADL</td>
<td>This runs when Cary Eclipse is detected online.</td>
</tr>
<tr>
<td>SR_OffLine.ADL</td>
<td>This runs when Cary Eclipse goes offline.</td>
</tr>
<tr>
<td>SR_Zero.ADL</td>
<td>This runs when the Zero button is selected.</td>
</tr>
<tr>
<td>SR_Shutdown.ADL</td>
<td>This runs when the application is shutting down.</td>
</tr>
<tr>
<td>SR_PreRead.ADL</td>
<td>This runs just before a read is taken.</td>
</tr>
<tr>
<td>SR_PostRead.ADL</td>
<td>This runs just after a read is taken (It has the reading result stored in the UserResult variable.)</td>
</tr>
<tr>
<td>SR_PreSetup.ADL</td>
<td>This runs when the Setup button is selected.</td>
</tr>
<tr>
<td>SR_PostSetup.ADL</td>
<td>This runs when the OK button on the Setup dialog is selected.</td>
</tr>
<tr>
<td>SR_CellChange.ADL</td>
<td>This runs instead of the Cary Eclipse driving the Multicell Holder. It enables you to add your own instructions for the Cary Eclipse Multicell Holder or to use your own cell changing accessory.</td>
</tr>
</tbody>
</table>

**How to insert ADL commands using the Quick Reference—Simple Reads**

The Quick Reference window is accessed from the ADL Command window. It enables you to easily add a command in the correct syntax to your ADL program. To do this:

**Step 1**
From the Commands menu, choose ADL Command or press Alt-Y to open the ADL Command Line window.

**Step 2**
Click the Multi-line option. The Command Line entry box will expand to take multiple lines of ADL commands and the Quick Reference check box will appear.

**Step 3**
Select the Quick Reference check box. The Quick Reference window will appear.

**Step 4**
Highlight the command you require in the Quick Reference. If necessary, use the Go To options to jump
Step 5
Click the Insert into ADL button. The selected ADL command will appear in the Command Line entry box.

Step 6
Where necessary, alter the defaults supplied by the inserted Quick Reference command in the Command Line entry box to suit your program.

Step 7
Repeat steps 4–7 to add more commands as required.

Help and Safety information—Simple Reads

- Help menu
- Safety home page

Help menu—Simple Reads

Use the Help menu to view the online Help and software information for this application.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td>Simple Reads</td>
<td>Select this to display the Simple Reads Home Page. This contains a list of online Help topics specific to this application.</td>
</tr>
<tr>
<td>Help Help</td>
<td></td>
</tr>
<tr>
<td>Help Topics</td>
<td>Select this to display the Cary Eclipse Windows Online Help Home Page.</td>
</tr>
<tr>
<td>Tip of the day</td>
<td>Select this option to view the tips of the day.</td>
</tr>
<tr>
<td>Eclipse Tour</td>
<td>Select this to display the Cary Eclipse Multimedia Online Help. This will give you an animated introduction to the Cary Eclipse software and hardware.</td>
</tr>
<tr>
<td>About…</td>
<td>Select About to access a dialog that displays information about the application as well as Varian email and Internet contact information.</td>
</tr>
</tbody>
</table>

Safety Home Page

Click on the desired index entry to view the Help.

- About Safety
- Lamp module
- Ultraviolet radiation
- Electrical hazards — Cary Eclipse
- Panels, covers and modules
- Other precautions
- Warnings and cautions
- Warning symbols
Simple Reads How To

Select the procedure you require below for a step-by-step informative guide on running the application software.

Complete Procedures

- How to perform a measurement at a single wavelength
- How to perform measurements at two wavelengths and subtract the result

Other

- How to remove or display the command buttons
- How to insert ADL commands using the Quick Reference
- How to open a previously stored report
- How to use advanced reporting options

See also:
Simple Reads application
System Information Home Page

The Cary Eclipse System Information application enables you to control information that is used by all applications such as company details, email host options and the way in which hints are displayed in the various applications. The information stored here is then made available to the reporting functions of all the other applications. For example, if you store your company's logo here it can be printed on a report issued from any application.

To access the System Information Application, from the Windows Taskbar click the Windows Start button, point to Programs, Cary Eclipse and then select System Information. The System Information application appears.

The System Information Application comprises the following tabbed pages:

- Company
- Hints
- Email Options

The System Information Application also contains the following menus:

- File
- View
- Help

See also: System Information How To's

Company—System Information

Use the Company page to enter company data for use throughout the system. To access the Company page, select the Company tab from the System Information application window.

Note

The details entered on this tab are stored when you select OK.

The page comprises the following:

Company Information

- **Header**
  Enter the text you wish to appear at the top of any reports printed from Cary Eclipse applications. Information regarding the date the report was generated and the number of pages in the report are automatically included in the header of any report produced.

- **Comments**
  Enter any additional information you wish to be included in your reports, for example, company details such as name and address. This information will be added to the first page of the report only.

- **Footer**
  Enter the text you wish to appear at the bottom of any reports printed from Cary
Eclipse applications.

**Hot Tip**
To alter the appearance of your text, change the settings in the Font dialog which can be accessed by right-clicking in the above fields.

**Logo Information**

**Add Logo**  
The **Add Logo** option enables you to include your company logo (or other graphic) in your reports. The logo will be added to the first page of the report between the Header and Comments information.

Click this option to open the **Open** dialog. Use the **Open** dialog to locate and select your company logo file (or other graphic) that will be included in Cary Eclipse reports. The file must be in one of the following graphics formats:

- bitmap (BMP)
- icon (ICO)
- enhanced Metafile (EMF)
- metafile (WMF)

Images up to 200 x 50 (width x height) pixels in size are recommended.

**Hot Tip**
As an alternative to selecting the Add Logo button, you can drag and drop images straight into the gray area from Windows Explorer.

Use the **Print Preview** command (accessible from the **File** menu) to ensure that the logo is displayed correctly.

**Clear Logo**  
This deletes the selected logo.

**See also:**
- [System Information home page](#)
- [How to enter a company logo](#)

**Hints—System Information**

Use this page to change the properties of **Hints**. To access the Hints page, select the Hints tab from the System Information application window.

The page comprises the following:

**Hint Properties**

Use the Hint properties to alter the timing and background color of the displayed Hint text. To do this, increase or decrease the pause times by selecting the slider bar and moving it to the left to decrease the pause or to the right to increase the pause:

**Hint Hide Pause**  
Sets the amount of time before the Hint will be hidden, (i.e. sets how long the Hint will remain visible).
In order for the Cary Eclipse's email facility to work correctly, you must set up the options on this page. The *Email Options* page is accessible from the System Application window by selecting the *Email Options* tab.

The page comprises the following:

- **Hint Pause**: Sets the period of time between when the mouse pointer is placed over an item and when the Hint appears.
- **Hint Short Pause**: Selects the amount of time to wait before displaying a Hint if a Hint is already showing in the screen.
- **Hint Color**: Click Hint Color to open the Windows Color dialog where you can change the color of the Hint background. You can select from the basic colors displayed or create your own color by selecting the Design Custom Colors button. Click OK to confirm your selection.
- **Test Area**: Place the mouse cursor in this area to preview the Hint properties.
- **Reset Hint Defaults**: Click this button to reset the Hints to the default system values.

**See also:**
- [View menu System Information](#)

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**Email Options—System Information**

In order for the Cary Eclipse's email facility to work correctly, you must set up the options on this page. The *Email Options* page is accessible from the System Application window by selecting the *Email Options* tab.

The page comprises the following:

- **Hot tip:**
  It is recommended that you contact your System Administrator to assist you with setting up the email options.

- **Mail host**: Enter the *Mail Host* for the computer on which the Cary Eclipse software is installed. The Mail Host will be listed in your email program, under the same or a similar title such as ‘Mail Server’ or ‘SMTP Server’.

- **Port**: Enter the *Port* number. This is usually set to 25.

**See also:**
- [System Information home page](#)

---

**Menu line—System Information**

- [File menu](#)
- [View menu](#)
- [Help menu](#)

**File menu—System Information**

The *File* menu comprises the following menu commands:
### View menu—System Information

The View menu comprises the following menu commands:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td>Print Setup</td>
<td>Select <strong>Print Setup</strong> to display the Windows <strong>Print Setup</strong> dialog where you can select the printer, printer page size and orientation.</td>
</tr>
<tr>
<td>Print Preview</td>
<td>Select <strong>Print Preview</strong> to display the <strong>Print Preview</strong> dialog where you can view the current report.</td>
</tr>
<tr>
<td>Print</td>
<td>Select <strong>Print</strong> to display the Windows <strong>Print</strong> dialog where you can select the printer and pages to print as well as the number of copies you wish to print. This command is also accessible by pressing CTRL+P.</td>
</tr>
<tr>
<td>Explorer</td>
<td>Select <strong>Explorer</strong> to open Windows Explorer. This may help you locate method and data files.</td>
</tr>
<tr>
<td>Exit</td>
<td>Select <strong>Exit</strong> to close the System Information application.</td>
</tr>
</tbody>
</table>

See also: [Print Preview](#), [System Information Home Page](#)

### Help menu—System Information

The Help menu enables you to view the online help and software information for this application. The menu comprises the following commands:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hints</td>
<td>Select this if you want the software to display Hints. A ‘Hint’ is a dialog containing a brief explanation of an item (e.g. a tab option, a data entry field, etc.), that appears when the mouse pointer is held over that item.</td>
</tr>
<tr>
<td>System Information Help</td>
<td>Select this to display the System Information Home Page. This contains a list of online Help topics specific to this application.</td>
</tr>
<tr>
<td>Help Topics</td>
<td>Select this to display the Cary Eclipse Windows Online Help Home Page.</td>
</tr>
<tr>
<td>About</td>
<td>Select this option to access a dialog that displays information about the application as well as Varian email and Internet contact information.</td>
</tr>
</tbody>
</table>

See also: [Hints](#), [System Information home page](#)
Print Preview—System Information

Use the Print Preview dialog to view the contents and layout of current report before you print it.

To access the Print Preview dialog, select the Print Preview command from the File menu.

You can reduce or enlarge the print preview on screen. Click the left mouse button in the preview area to toggle through the five available views. Click the right mouse button in the preview area to return to the full page view.

Use the scroll bars to the right and bottom of the report to move around the preview.

Note
The scroll bars are not available for the full page view.

<table>
<thead>
<tr>
<th>Command</th>
<th>Button</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td>First arrow</td>
<td><img src="image" alt="First arrow" /></td>
<td>Use this arrow to view the first page of the report.</td>
</tr>
<tr>
<td>Previous arrow</td>
<td><img src="image" alt="Previous arrow" /></td>
<td>Use this arrow to view the previous page of the report.</td>
</tr>
<tr>
<td>Next arrow</td>
<td><img src="image" alt="Next arrow" /></td>
<td>Use this arrow to view the next page of the report.</td>
</tr>
<tr>
<td>Last arrow</td>
<td><img src="image" alt="Last arrow" /></td>
<td>Use this arrow to view the last page of the report.</td>
</tr>
<tr>
<td>Print</td>
<td><img src="image" alt="Print" /></td>
<td>Use this to display the Windows Print dialog where you can choose the printer and pages to print as well as the number of copies you wish to print.</td>
</tr>
</tbody>
</table>

Q. Why are all of the arrow buttons grayed out?
A. All of the buttons will be grayed out if the report has only one page of data.

See also:
File menu

System Information How To

Select this procedure for a step-by-step informative guide on importing company logos.

- How to enter a company logo

See also:
System Information home page

How to enter a company logo—System Information

Step 1
On the Windows Taskbar, click the Windows Start button, point to Programs, Cary Eclipse and then select System Information. The System Information application appears.

**Note**
If you are running the Cary Eclipse software under Windows NT you need to be logged on as an Administrator in order to specify the logo in the System Information application.

**Step 2**

On the Company page, select your logo by doing the following:

a. Click the Add Logo button to display the Open dialog.

b. Select the directory containing your logo in the Look in field.

c. Enter the name of your logo in the File name field. You can use Bitmap, Icon, Enhanced Metafile and Metafile file types. Images should be no larger than 200 x 50 (width x height) pixels.

d. Click Open to clear Open dialog. Your logo will appear in the gray area just below the Company Information fields.

If you are not satisfied with the current logo, select Clear Logo to delete it and repeat Step 2 to select a different logo.

**Hot Tip**
As an alternative to steps a–d above, you can drag and drop images straight into the gray area from Windows Explorer.

**Step 3**

When you have made your logo selection, select OK to exit the System Information application. Your logo will be present in any application reports when Company Logo is selected on the Reports page. The logo will be displayed on the first page of your reports, underneath the header information.

**Note**
In the Simple Reads application, the Logo will be automatically selected, as the Report page is not available.

See also:
Company page
## Thermal Home Page

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<th>To…</th>
<th>Use the…</th>
<th>Show me…</th>
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<td>• Setup dialog</td>
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<td>• Commands menu</td>
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</tr>
</tbody>
</table>

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<tr>
<th>To find out about…</th>
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<td>Accessories</td>
<td>• Multicell Holder</td>
<td></td>
</tr>
</tbody>
</table>
Introduction

The Thermal application allows you to perform thermal analyses on DNA using one of the Cary accessories with temperature control. Once the data is collected, you can choose to calculate the melting temperature, Tm, by either the derivative or hyperchromicity methods. The hyperchromicity methods also allow you to determine thermodynamic parameters such as ΔS, ΔH and ΔG.

Use the Thermal application to measure the increase or decrease in emission intensity as a function of time. From this you can obtain an intensity versus time plot. In addition, you can also perform polarization measurements as a function of temperature.

To access the Thermal Application, click the Windows Start button, point to Programs, Cary Eclipse and then select Thermal.

Software controls

The application comprises the following:

**Instrument Status Readings**

- The Instrument Status Readings, which display the Y and X values of the graph axes are displayed in the top left and right corners of the Thermal Application Window.
  - **Y value**
    - Displays the current Y value, usually intensity. This value is continually updated during a data collection.
  - **X value**
    - Displays the current X value, which is the current reading for the Temperature.

**Start and Stop buttons**

- The Start and Stop buttons at the top of the Thermal Application window are used to start and stop instrument analyses. These buttons can also be accessed from the Commands menu.

**Graphics area**

- The Graphics area refers to the plotting area of the application. It can contain traces of collected data, labels and pictures. In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area defined by the axes is referred to as a graph box.

  - To display or hide the Graphics area, select the Graphics command on the View menu. Double-clicking on the Graphics area enables you to toggle between half or full size display sizes. To manually size the Graphics area, place the mouse cursor over the border between the Graph and Report areas until the cursor changes to a double arrow (}
Validation of your Cary Eclipse for the Thermal application

Two Validation files are shipped with the Thermal application to enable you to validate the software. The files are stored on your CD in a Validation subdirectory.

To validate your software, you should Open each file in turn and select the Recalculate button. The results calculated by the Cary Eclipse should match those given in the validation files listed below. These

Tell me more about the Graphics area

Report area
Use the Thermal Report area to view the current report. The report may include the results of your data collection, the method parameters used to collect the results and any graphics you have included. You can select the information to include in your report in the Reports tab of the Setup dialog.

To display or hide the Report area, select the Report command from the View menu. Double-clicking on the Report area enables you to toggle between half or full size display sizes. To manually size the Report area, place the mouse cursor over the border between the Graph and Report areas until the cursor changes to a double arrow ( mogul). Then hold down the mouse and drag it up or down to resize the areas.

You can copy the generated report onto the Windows clipboard. To do this, highlight the report (or a relevant section of it) and press CTRL+C. You can then paste the report into a word processor such as Microsoft® Word.

Hot Tip
You can edit the Thermal Report by pressing CTRL+R or selecting the Edit Report command from the Edit menu.

Commands
The Thermal Application window contains the following commands, all of which can also be accessed from the various menus in the application.

Click on the buttons to view information about them:

<table>
<thead>
<tr>
<th>Setup ...</th>
<th>Zero</th>
<th>Set temp ...</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clear report</td>
<td>Recalculate</td>
<td>Print ...</td>
</tr>
</tbody>
</table>

Hot Tip
By becoming familiar with these commands (Setup, Zero, etc.), you can remove these buttons from the Thermal window. This will enlarge the Report area and make it easier to view results. To do this, clear the Buttons item in the View menu.

Menus
The Thermal Application contains the following menus:
File, Edit, View, Commands, Setup, Graph, Help

See also:
Status Display
Status Line
Keyboard function keys
Thermal home page

Validation of your Cary Eclipse for the Thermal application
results were determined by exporting the data to a spreadsheet program and performing the same calculation as that used in the Thermal application.

- Thermal hyperchromicity calculation validation.FBTM
- Thermal derivative calculation validation.FBTM

**Getting ready for a run—Thermal**

- Setup dialog
- Setup menu
- Cary page
- User collect example
- Bio-/Chemi-luminescence and Phosphorescence Options dialog
- Gate Time for Bio-/Chemi-luminescence
- Gate Time for Phosphorescence
- How many flashes?
- Options page
- About smoothing
- Accessories page
- Multicell/Temperature accessory
- Multicell Changer
- Automated and Manual Polarization Accessories
- Analyze page
- Information on van't Hoff calculations
- Reports page
- Report toolbar
- Clear Report
- How to use the advanced reporting options
- Auto Store page
- How to email the results

**Setup—Thermal**

Use the Setup dialog to set up your instrument and accessories for a Thermal run. You can also use the dialog to set up the Analysis, Report and Storage parameters associated with the run.

To access the Setup dialog select the Setup button from the Thermal Application window or the Setup command from the Setup menu.

The Setup dialog comprises the following tabbed pages:

- Cary
- Options
- Accessories
- Analyze
- Reports
- Auto Store
Setup menu—Thermal

Use the Setup menu to access the parameters for setting up a Thermal read.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Setup</td>
<td>Choose Setup to display the Setup dialog where you can set up your instrument for a Thermal measurement. The Setup dialog can also be accessed from the Setup command button.</td>
</tr>
<tr>
<td>Cary</td>
<td>Choose Cary to display the Cary page. You can use this page to set up the instrument parameters associated with the data collection.</td>
</tr>
<tr>
<td>Options</td>
<td>Choose Options to display the Options page where you can set up your filters and set the detector voltage. You can also use this page to select the way the collected data is displayed in the Graphics area, and the type of smoothing applied (if any).</td>
</tr>
<tr>
<td>Accessories</td>
<td>Choose Accessories to display the Accessories page. You can use this page to set up the accessory parameters associated with the data collection.</td>
</tr>
<tr>
<td>Analyze</td>
<td>Choose Analyze to display the Analyze page where you can select from various mathematical equations and set the parameters used to generate the calculation of reaction rates.</td>
</tr>
<tr>
<td>Reports</td>
<td>Choose Reports to display the Reports page. You can use this page to set up the report parameters associated with data collection.</td>
</tr>
<tr>
<td>Auto Store</td>
<td>Choose Auto Store to display the Auto Store page where you can specify if and when data will be stored, and in what format.</td>
</tr>
</tbody>
</table>

See also:
- Setup
- Cary
- Options
- Accessories
- Analyze
- Reports
- Auto Store

Example of a User Collect—Thermal

You can extend the functionality of your Cary Eclipse instrument by using the built-in Read command. This command enables you to change the instrument to a different wavelength and take a reading. You can combine this command with the mathematical operators: ‘+’, ‘–’, ‘*’ and ‘/’. This is illustrated in the example below.

**Read(340, 400)-Read(340, 450)**

This reads the intensity at Excitation monochromator = 340 nm, Emission monochromator = 400 nm and subtracts the intensity read at Excitation monochromator = 340 nm and Emission monochromator = 450 nm. This reading and/or calculation is performed at each temperature.

The User collect command can be selected on the Cary page of the Setup dialog. Enter the Read
command in the **User collect result** field.

**Note:**
User collect can not be used with the Auto Polarizer Accessory.

**See also:**
Cary

**Bio-/Chemi-luminescence/Phosphorescence Options—Thermal**

Use the **Bio-/Chemi-luminescence/Phosphorescence Options** dialog to set up the parameters associated with these two measurement modes.

To access the **Bio-/Chemi-luminescence/Phosphorescence** dialog, select the **Options** button from the Cary page.

**Note**
The **Options** button is only visible on the Cary page if you have selected **Bio-/Chemi-luminescence** or **Phosphorescence** as the **Data Mode**.

**Dialog controls**

The **Bio-/Chemi-luminescence/Phosphorescence Options** dialog comprises the following options:

- **Total decay time**
  Enter, in seconds, the total decay time for your sample. The total decay time is the time that it takes for the phosphorescence signal to decay to dark levels (zero). The lamp will not flash again until the total decay time has elapsed. The range is 0.001–1800.000 s.

  **Note**
  This option does not apply to Bio-/Chemi-luminescence mode.

- **No. of flashes**
  Enter the number of lamp flashes required, which is how many times the lamp flashes consecutively before the delay time is counted.

  If you are measuring phosphorescence samples that have a relatively long decay time (that is, a number of seconds), you can setup the Cary Eclipse to pulse the lamp multiple times, as this will increase the intensity of the phosphorescent signal. For short lived phosphorescent samples, multiple flashes will not impact on the intensity of the signal, and therefore only one flash is required.

  **How many flashes should I perform?**

  **Note**
  This option does not apply to Bio-/Chemi-luminescence mode.
Delay time  Enter, in milliseconds, the delay time for your sample. The delay time is the time that elapses between the last flash and the beginning of data collection. The default value is 0.100 ms, which is the time it takes for the lamp intensity to decay to zero. Setting a delay time ensures that there is no lamp effects in the phosphorescence decay curve.

The range is 0.000–5000.000 s.

Note  This option does not apply to Bio-/Chemi-luminescence mode.

Gate time  Enter, in milliseconds, the gate time for your sample. The gate time is the overall reading time of the emission signal.

The range is from 10µs–10s

Example of the Bio-/Chemi-luminescence/Phosphorescence Options

See also:  Cary page

Understanding Gate Time for Bio-/Chemi-luminescence scans—Thermal

Gate Time is set on Options page, which is accessible from the Cary page.
The following diagram highlights the Gate Time setting.

The Gate Time is set to 10 and the Intensity is equal to $I_T$.

![Diagram showing Gate Time and Intensity relationship]

Understanding Average Time and Gate Time for Phosphorescence Scans—Thermal

For Phosphorescence scans, there is a relationship between Average Time (the time for which data is averaged) and Gate Time (the overall reading time of the emission signal).

Averaging is calculated as follows:

The data point at each wavelength will be the average of the signals from a number of gate time measurements that occur during this averaging time, that is:

**Averaging Time/Gate Time**

These signals are accumulated from successive decay curves so that the actual time taken to generate a point at a single wavelength will be approximately:

**(Averaging Time/Gate Time x Total Decay Time)**
In phosphorescence mode, the Averaging Time ranges from the Gate Time to a maximum of 1000 s.

**Note:**
Gate Time is not used if you select Fluorescence as the Data Mode.

**Average Time** is set on the Cary page of the Setup dialog. Gate Time is set on Options page, which is accessible from the Cary page.

The following diagrams highlight the relationship between Average Time and Gate Time.

**Example 1**

When the Average Time and Gate Time are set to the same value:

<table>
<thead>
<tr>
<th>Average Time/Gate Time</th>
<th>1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intensity</td>
<td>I (Area I_A, I_B)</td>
</tr>
</tbody>
</table>

where:

- Total Decay Time: 20 ms
- Delay Time: 10 ms
- Cycle Time: 1 sec
- Stop Time: 3 sec
Average Time 5 ms
Gate Time 5 ms

Example 2

When the Average Time is set to five times the Gate Time:

<table>
<thead>
<tr>
<th>Average Time/Gate Time</th>
<th>5</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intensity (Area I₂)</td>
<td>(A₁ + A₂ + A₃ + A₄ + A₅)</td>
</tr>
<tr>
<td></td>
<td>5</td>
</tr>
</tbody>
</table>
where:

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total Decay Time</td>
<td>20 ms</td>
</tr>
<tr>
<td>Delay Time</td>
<td>10 ms</td>
</tr>
<tr>
<td>Cycle Time</td>
<td>1 sec</td>
</tr>
<tr>
<td>Stop Time</td>
<td>3 sec</td>
</tr>
<tr>
<td>Average Time</td>
<td>25 ms</td>
</tr>
</tbody>
</table>
How many flashes should I perform?—Thermal

The **Number of Flashes** option is set in the **Bio-/Chemi-luminescence/Phosphorescence Options** dialog. To access the dialog, select the **Options** button from the **Cary tab** page.

**Note**
- The **Options** button is only visible on the **Cary page** if you have selected **Bio-/Chemi-luminescence** or **Phosphorescence** as the **Data mode**.

If you are measuring phosphorescence samples that have a relatively long decay time you can setup the Cary Eclipse to pulse the lamp multiple times, as this will increase the intensity of the phosphorescent signal (refer to Example 1, below).

**Example 1**
The above example shows that the sample does not decay before the instrument lamp flashes again. Therefore setting multiple flashes will result in an increase in the intensity of the signal.

For short lived phosphorescent samples, multiple flashes will not impact on the intensity of the signal, and therefore only one flash is required (refer to Example 2, below).
Example 2
The above example shows that the sample decays in less time than the time it takes the instrument to flash the lamp again. Therefore, setting more than 1 flash, in the No. of Flashes field will not increase the intensity of the signal.

See also:
Bio-/Chemi-luminescence/Phosphorescence Options

Options—Thermal

Display Options | Smoothing | Excitation Filter | Emission Filter | PMT Detector Voltage | Status Display

Use the Options page to set up your filters and set the detector voltage. You can also use this page to select the way the collected data is displayed in the Graphics area, and the type of smoothing applied (if any).

From the Setup menu, select Options to access the Options page. You can also access the page from the Setup dialog by selecting the Options tab. To access the Setup dialog, select the Setup button from the Thermal Application window.

Display Options

Overlay traces
Select this option to superimpose the collected data of each sample in the run in one graph box. If you select this option, a graph box is created at the start of the collect in the Graphics area and each sample’s data is overlaid in that box as the data is collected. Clear this option to display all of the traces in individual graphs.

Y minimum/Y maximum
Enter the minimum and maximum Y values to be displayed on the graph axis. The software will update the Y axes to reflect these settings during the thermal run.

Smoothing

Select Smoothing to reduce the noise of your collected data. When you select this option, smoothing will be applied to the intensity signal using the algorithms defined below. You can select one of the three types of smoothing that can be performed during a run. Only the smoothed trace is displayed during data collection, however the unsmoothed data is also saved with the experiment.

Type
Savitzky-Golay
This algorithm will take the number of Filter points, apply the Savitzky and Golay smoothing algorithm and display the smoothed value at the middle X-point of the filter.
The naming of the smoothed trace will be 'Sample name_SG'.

Tell me more about Savitzky-Golay smoothing

**Boxcar average**
This algorithm will take the number of points as defined by the Filter size, and average these, displaying the average at the middle X-point of the filter. The naming of the smoothed trace will be 'Sample name_Box'.

Tell me more about Boxcar averaging

**Moving average**
This algorithm will take the number of points as defined by the Filter size, and average these, displaying the average at the middle X-point of the filter. The naming of the smoothed trace will be 'Sample name_Mov'.

Tell me more about Moving average

**Filter size**
Enter the Filter size, which determines the number of points that will be used in the smoothing procedures. The higher the filter size (and thus the number of points used), the greater the amount of smoothing that will be performed.

Filter sizes must be odd values. For Boxcar and Moving averages, the range is 3–101. For Savitzky-Golay smoothing, the range is 5–101.

**Excitation filter**
Set the excitation filter for the Thermal measurement. To do this, select the down arrow to the right of the field and select an option from the list.

A range of filter settings are available:

**Auto**
If you select the Auto option, the filter wheel will be automatically moved to the appropriate position for the selected excitation wavelength. It is recommended that you set the filter to Auto (which is the default setting).

**Open**
If you select Open no filter is used.

**Closed**
If you select Closed, a blank filter is used to prevent light from getting through.

**Emission filter**
Set the emission filter for the Thermal measurement. To do this, select the down arrow to the right of the field and select an option from the list.

A range of filter settings are available:

**Auto**
If you select the **Auto** option, the filter wheel will be automatically moved to the appropriate position for the selected emission wavelength. It is recommended that you set the filter to **Auto** (which is the default setting).

**Open**  
If you select **Open** no filter is used.

**Closed**  
If you select **Closed**, a blank filter is used to prevent light from getting through.

**250–395, 295–1110, 360–1110, 430–1110, 550–1110**  
If you are looking at a wavelength range that is within one of these filter ranges, you can select your filter manually. Otherwise it is recommended that you set the filter to **Auto**.

**3%T Attenuator**  
Select **3%T Attenuator** to decrease the fluorescence intensity and thus the amount of light going to the emission PMT. This setting is useful for highly fluorescent samples when you are using wide slits.

**PMT Detector Voltage**  
Set the sample PMT high voltage. To do this, select one of the following:

- **Low**  
Sets the PMT Detector voltage to a pre-defined value of 400 v. Typically you would select this setting if you are reading highly emitting samples.

- **Medium**  
Sets the PMT Detector voltage to a pre-defined value of 600 v. Typically you would select this setting if you are reading moderately emitting samples.

- **High**  
Sets the PMT Detector voltage to a pre-defined value of 800 v. Typically you would select this setting if you are reading low emitting samples.

- **Manual**  
Enables you to enter any PMT voltage in the adjacent Volts field.

**Volts**  
Enter the PMT voltage in the adjacent field. Note that this option is only available if you have selected **Manual** as the PMT Detector voltage.

**Tip**  
Increasing or decreasing the **PMT Detector Voltage** by 10% will approximately double or halve the fluorescence intensity observed.

**Status Display** Select this check box to display the **Status Display** window. This window allows you to view the status of various parameters during the data collection.

See also:  
- **Setup**  
- **Status Display window**

### About Smoothing—Thermal
Savitzky-Golay

Savitzky-Golay smoothing is used to obtain good noise reduction with sharp peaks. The equation constants are specific to Filter Size. The constants for each filter size are published in the Savitzky-Golay paper (A. Savitzky, M.J.E. Golay, Anal. Chem., 36 (1964) 1627). The equation also requires that Filter Sizes entered must be odd values, between 5 and 101.

Example

Data \((x_1, y_1), (x_2, y_2), (x_3, y_3) \ldots (x_n, y_n)\)

Filter size 5

Result

The first smoothed data point will be:
\((x_3, (Y_1a_1+Y_2a_2+Y_3a_3+Y_4a_4+y_5a_5)/b),\)

the next:
\((x_4, (y_2a_1+y_3a_2+y_4a_3+y_5a_4+y_6a_5)/b) \ldots\)
\((x_{n-2}, \text{sum(y}_{n-4}a_1+y_{n-3}a_2+y_{n-2}a_3+y_{n-1}a_4+y_n a_5)/b),\)

where \(a_1..a_5, b\) are constants defined by Savitzky and Golay.

Each smoothed point is calculated using the previous and next data points. For this reason, calculations cannot be performed on the very first and very last data points in a data set. Hence, smoothed traces will lack the first and last X values of the original trace.

Boxcar average

Boxcar averaging can be used when the change in intensity is minimal.
Example

Data \((x_1, y_1), (x_2, y_2), (x_3, y_3) \ldots (x_n, y_n)\)
Filter size 5

Result

The first smoothed data point will be:
\((x_3, \text{sum}(Y_1:y_5)/5),\)

the next:
\((x_8, \text{sum}(y_6:y_{10})/5) \ldots (x_{n-2}, \text{sum}(y_{n-4}:y_n)/5).\)

If there are not enough points at the end of the data set to complete the filter size, then these points will be ignored, hence, the gap at the end may be larger than the gap in the beginning.

Moving average
Example

Data \((x_1, y_1), (x_2, y_2), (x_3, y_3) \ldots (x_n, y_n)\)

Filter size 5

Result

The first smoothed data point will be:
\((x_3, \text{sum}(Y_1:y_5)/5),\)

the next:
\((x_4, \text{sum}(y_2:y_6)/5) \ldots (x_{n-2}, \text{sum}(y_{n-4}:y_n)/5).\)

Accessories—Thermal

Use the Accessories page to set up the accessory parameters associated with the data collection.

From the Setup menu, select Accessories to access the Accessories page. You can also access the page from the Setup dialog by selecting the Accessories tab. To access the Setup dialog, select the Setup button from the Thermal Application window.

The Accessories page will vary, depending upon two things. Firstly which radio button is selected in the Collect Temperatures group box, eg. Ramps, Go to or Thermal Cycles and will also vary depending on which accessory is selected on the Accessory tab.
The Multicell and Temperature accessory page is displayed when you select the Multicell/Temperature tab on the Accessories page.

**Note**

If you select the Ramps or Thermal Cycles radio button on the Cary tab you can only use the Peltier 4 Position Multicell Holder Accessory. If you select the Go To radio button you can use any of the thermostatted Eclipse accessories.

The page comprises the following:

**Multicell Holder**

Select this option to use the Multicell Holder and cells with the Cary Eclipse.

- **Multi-zero**
  - If you select Multi-zero, a Multi-zero will be performed when the Zero button is selected in the Thermal Application window. Because cuvettes can have different intensities, and samples vary, this field is used when you require all blank readings to consistently start at zero intensity.
  
  A reading will be taken at the set **Averaging Time** at the set Ex. and Em. wavelengths and at every selected cell position. These readings will be offset to zero and subsequent readings will have the corresponding offset value subtracted from each cell.

- **Reset**
  - Select Reset to drive the Multicell Holder to cell position 1, which is its fully retracted or zero position.

- **Go to cell...**
  - Select Go to cell to open the Multicell Changer dialog. You can use this dialog to manually move the Multicell Holder accessory to a new cell. To do this, simply select the corresponding button of the cell you require in the Multi Cell Changer dialog. The Multicell Holder will move the selected cell to the measurement position in the sample compartment.

**Note**

The Multicell Holder checkbox must be unchecked to use the Go to cell option.

The Multicell Changer dialog is ‘modeless’ which means that it can remain open even
once you have cleared the Setup dialog. This enables you to use the **Rapid Read** command to perform a reading at the cell specified in the Cell Changer.

If you move to a new cell after you have performed a Multi-zero thermal, the Cary Eclipse will display the intensity reading corrected by the determined zero-offset amount for that cell.

**Note**

You can use this option to check the intensity of a cell or start a thermal run at a particular cell.

## Temperature Control

Select this option to use the Temperature Controller with the Cary Eclipse.

**Note**

When the Thermal application is opened the Multicell peltier and Temperature controller are selected by default.

**Note**

The **Temperature Control** option does not control temperature accessories that are not listed in the **Device** field. For example, if you have selected the **Temperature Control** option and set the **Device** to **None** because you have a different temperature control accessory, the Cary Eclipse software will not control that accessory. However, it will monitor the temperature and prompt the software to start when the accessory reaches the temperature listed in the **Temperature** field.

**Device**

Select the device that will be used to control the solution temperature. If the device options are not available ensure that **Go To** is selected on the Cary tab.

The options available will depend upon whether or not you have selected the **Multicell holder** option or the **Single Cell holder** option.

If you *have* selected the Multicell holder, the following **Device** options will be available for selection:

- Temperature controller
- PCB 150 water bath
- None

If you *have* selected the Single Cell holder, the following **Device** options will be available for selection:

- Single cell peltier
- PCB 150 Water bath
- None

**None**

Select this option if you are using a Temperature Control device not listed in this field (for example, a third party water bath). Note that the Cary Eclipse software will not control the accessory. The software will simply monitor the temperature and prompt the software to start when the accessory reaches the temperature listed in the **Temperature** field.

**PCB 150 Water Bath**
Select this option if you are using the PCB 150 water bath.

**Single cell peltier**
Select this option if you are using the Single cell peltier.

**Temperature controller**
Select this option if you are using the Temperature controller.

**Port**
Select from COM1, COM2, COM3 or COM4.
This option is not available if you have selected None as the Device.

**Monitor**
Select which device will be used to monitor the measurement temperature. Select from Block, Probe 1, Probe 2, Probe 3 or Probe 4.
Note that the the options in this field may change depending on the Device selected.

**Temperature Display**

Select this option to view Temperature Display information in the Status Display window.
The Temperature Display option is only available if you have selected the Temperature Control option.

**Block/**
PCB 150
Select this option to view the temperature of the Block (if you have selected the Single Cell Peltier device) or the PCB 150 (if you have selected the PCB 150 Water Bath) in the Status Display window.

**Probes**
Select one or more of these options to view the temperature of the Probes in the Status Display window.

**Status Display**

Select this check box to display the Status Display window. Use this window to view the status of various parameters during the data collection.

See also:
Accessories
Multi Cell Changer
Status Display

**Multi Cell Changer—Thermal**

Use the Multi Cell Changer dialog to manually move the Multicell Holder accessory to a new cell position. To access the Multi Cell Changer dialog, select the Go to cell button from the Multicell/Temperature Accessories page.

**Note**
The Go to cell button is only available when the Multicell Holder checkbox is unchecked.

The dialog comprises the following:

**Multicell changer**
To manually move the Multicell Holder accessory to a new cell, simply select the corresponding button of the cell you require in the Multi Cell Changer dialog. The Multicell Holder will move the selected cell to the measurement position in the sample...
Two types of polarizer accessories are available for the Cary Eclipse: (1) the Manual Polarizer Accessory and (2) the Automated Polarizer Accessory, either can be used with the Multicell Accessory. Click on the links above for installation instructions for these accessories.

The Polarizer accessory page is displayed when you select the Polarizer tab on the Accessories page after you have selected "Go To" on the Cary tab. If you are using the manual or automatic polarizer accessories, this page enables you to setup the polarizers.

The Manual and Automatic Polarizer accessories enable you to monitor the change in polarization of the fluorescence from a compound over a range of temperatures.

Polarizer

Select this option to use the Polarizer with the Cary Eclipse.

Q. Why is the Polarization option unavailable?

A. The User Collect option has been selected on the Cary page. The Polarizer accessory can be used for both single and multtispectral measurements but is not available for User collect measurements.

A. The Ramps or Thermal Cycles option has been selected on the Cary page. You cannot use the Polarizer accessory if you are using either of these temperature ramping options. You must select "Go To" on the Cary Page.

Measurement Select from the following measurement types: Polarization or Anisotropy.

G Factor The G Factor is a value that reflects the polarization bias of the emission monochromator. It is dependant upon the emission wavelength. If you do not select the
G Factor option, then a G-factor value must be collected. If you do select the G Factor option then the value entered is used at the emission wavelength in subsequent polarization or anisotropy calculations. If Multiwavelength is selected on the Cary tab then G Factors must be measured at all emission wavelengths; the manual entry G Factor field is not available.

Determining the G Factor

On exiting the Setup dialog a G Factor validity test is performed. If the G Factor is not valid, the following prompt is displayed:

‘The current G Factor is not valid at the emission wavelength. Press the G Factor button to determine a new grating factor.’

Select OK to clear this dialog and select the G Factor button from the Thermal Application window.

Status Display

Select this check box to display the Status Display window. Use this window to view the status of various parameters during the data collection.

See also:
Manual Polarizer accessory
Automated Polarizer accessory
Accessories page

Analyze—Thermal

Derivative | Hyperchromicity | Post-Run Smoothing | Show Status Display

The Analyze tab enables you to select the calculation used to estimate the thermal melting temperature (ie, the temperature at which the strands in the DNA separate (Tm)).

Two methods are available to calculate Tm:

- The Derivative Method
- The Hyperchromicity Method

None

When this is selected, no Tm calculations will be performed.

Derivative

The Derivative method calculates and plots the first derivative of the selected thermal.

The point at which the derivative plot peaks, (i.e. maximum absolute gradient on the original plot), is reported as the Tm value, and the original plot will be marked with an arrow to indicate this point.

Note

There are situations, where there are multiple transitions within a single run, resulting in more than
When you select Derivative the following options are available:

**Autocalculate**  Select this option to enable the system to collect data and automatically perform a derivative calculation at the end of each run. Otherwise you can perform the calculation manually, once the run is completed.

**Interval**  A derivative calculation involves the selection of points from a plot, from which a derivative calculation is performed. Selection of these points is dependent upon the Interval chosen. Also, the Savitzky-Golay method can only be used if the data points are equidistant, i.e. the same X distance from each other. In cases where data is not equidistant the Interval is used to create an equidistant data set prior to performing the Savitzky-Golay calculation.

**Example:**
If a data interval of ‘1’ is chosen, then data will be taken every 1 °C over the entire range of the continuum.

The selected data points are then used to generate a separate continuum on which a derivative calculation is performed.

The selection of the Derivative Interval is dependent on the rate of data collection. If data is collected every 0.1 °C then a Derivative Interval of 0.1 °C will give the most accurate result. A Derivative Interval of a less value than 0.1 °C will be no more accurate and will produce the same result.

**Filter Size**  This sets the derivative Filter Size.

The derivative calculation uses a Savitzky-Golay smoothing technique where a number of points surrounding an individual point are averaged to produce a new, derivative point. The Filter Size will be automatically adjusted so that it is a multiple of the total number of data points.

**Note**
The larger the Filter Size, the more data points that are included in the filter for the derivative calculation.

**Low/High Calculation Limits**  Low and High Calculation Limits are used to define the range over which a derivative calculation is performed. The selection of these limits is only applicable when performing a derivative calculation using Autocalculate.

Q. Why are the Derivative group options grayed out?
A. Select the Derivative radio button to activate these.

**Hyperchromicity**

The Hyperchromicity option enables you to calculate multiple parameters using hyperchromicity calculations.

When you select Hyperchromicity the following options are available:
Post-Run Smoothing

Signal noise from biological material will cause interference and noise within the system. Smoothing, applied to the absorbance value, gives the ability to smooth a data curve. This is achieved with the use of a Savitzky-Golay smoothing filter.

When you select **Smoothing** the following options are available:

- **Interval**
  Selection of a smoothing interval is independent of the data collection interval. Data can be collected at one rate and smoothed at another. For example, data collection can be performed every half of a degree, however smoothing can be applied every two degrees.

  Type the number in this numeric field to select the smoothing interval. The larger the number the smoother the data will become, but you will start to lose detail as you increase the interval.

- **Filter Size**
  Set the filter size by entering a value in the numeric field.

  The smoothing algorithm uses a Savitzky-Golay smoothing technique where a number
of points surrounding an individual point are averaged to produce a new, smoothed point.

**Note**
The larger the filter size, the more data points that are included in the filter for smoothing.

**Show Status Display**

Select this checkbox to display the Status Display window. This window allows you to view the status of your Thermal run during the data collection.

See also:
- Savitzky-Golay
- Example of Thermal trace with derivative
- More Information on van't Hoff calculations
- How to perform Derivative calculations from previously stored data
- How to perform Hyperchromicity calculations from previously stored data

**More information on van't Hoff Calculations**

- **Tm** value from alpha curve °C and **K**
- **K** at various temperatures from alpha curve
- Delta S (ΔS) and Delta H (ΔH) from van't Hoff plot
- Delta G (ΔG) and **K**
- **K** (Tm) calculations

**Calculating the Tm value from alpha curve °C and K**

The Cary firstly converts the intensity versus temperature data to a form that expresses the fraction of hybridized strands remaining as a function of temperature.

By specifying the lower and upper limits of the linear region of the Associated DNA part of the curve, a linear least squares line is calculated and is extrapolated to both ends of the melting curve.

Similarly by specifying the lower and upper limits of the linear region of the Dissociated DNA part of the curve, a linear least squares line is calculated and is extrapolated to both ends of the melting curve.

If x is the distance from the curve to the upper line and y is the distance from the lower line to the curve, then:

\[ \alpha(T) = \frac{x}{x+y} \]

where \( \alpha \) is the fraction of strands remaining hybridized at a given temperature.

From the displayed alpha curve you are able to select the region which will be used to calculate the Tm value.

A linear least squares line is drawn between the selected points and the value of temperature, where \( a = 0.5 \), is calculated to give the Tm value.
The Tm value is automatically reported in °C and K.

**K at various temperatures from alpha curve**

To calculate the equilibrium constants as a function of temperature, K(t), you must know the values of α (alpha). This can be calculated according to either of the following equations:

- **Self complementary sequence**
- **Non-self complementary sequence**

You will be required to specify:

- Molecularity (i.e. the number of strands) of the hybrid
- Total concentration of strands.

**Self complementary sequence**

The general equilibrium equation shown below indicates how self complementary sequences form n-mer structures:

\[ nA \rightleftharpoons A(n) \]

In this type of situation the following formula is used to calculate the equilibrium constant:

\[
K(T) = \frac{a(T)}{(n*C_t^{(n-1)} * (1-a(T))^n}
\]

where:

- \(a(T)\) = the fraction of strands remaining hybridized at a given temperature
- \(n\) = the molecularity
- \(C_t\) = total concentration of strands

**Non-self complementary sequence**

The general equilibrium equation shown below indicates how non-self complementary sequences form n-mer structures:

\[ A(1) + A(2) + A(3) + \ldots + A(n) \rightleftharpoons A(1)A(2)A(3)\ldots A(n) \]

In this type of situation the following formula is used to calculate the equilibrium constant:

\[
K(T) = \frac{a(T)}{(C_t/n)^{(n-1)} * (1-a(T))^n}
\]

where:

- \(a(T)\) = the fraction of strands remaining hybridized at a given temperature
\( n \) = the molecularity
\( C_t \) = total concentration of strands

**Delta S (\( \Delta S \)) and Delta H (\( \Delta H \)) from van't Hoff plot**

The parameters chosen are used to calculate a van't Hoff plot (i.e. \( \ln(K(T)) \) vs \( 1000/T(K) \)).

By selecting a lower and upper region of the calculated plot, a linear least squares line is calculated and extrapolated to both ends of the van't Hoff plot.

The slope and intercept of the calculated line gives the enthalpy and entropy of the hybrid formation (\( \Delta H^\circ \) and \( \Delta S^\circ \), respectively) according to the following equations:

\[
\text{slope} = -\frac{\Delta H^\circ}{R}
\]

\[
\text{intercept} = \frac{\Delta S^\circ}{R}
\]

where \( R \) is the gas constant = 1.986 calories/mole.K

---

**Note**

\( \Delta H^\circ \) is reported in Kcal/mol and KJ/mol units. \( \Delta S^\circ \) is reported in cal/mol/K and as J/mol/K units.

**Delta G (\( \Delta G \)) and K at various temperatures**

If selected, this enables you to choose the various temperatures (in °C) at which \( \Delta G \) (free energy) and \( K \) (equilibrium constant) are calculated.

\[
\Delta G = \Delta H - T (\Delta S)
\]

\[
K(T) = e^{(\Delta G / -RT)}
\]

where:

\( \Delta H \) = the enthalpy of the hybrid formation
\( \Delta S \) = the entropy of the hybrid formation
\( T \) = temperature
\( R \) = the gas constant (1.986 calories/mole K)

Enter the required Number of Temperatures at which you want to calculate \( \Delta G \) and \( K \).

Fill in the required temperatures in the numeric fields.

**K (Tm) calculations**

The calculated alpha curve is used to report the following parameters:

\( K(Tm) \) experimental
\( K(Tm) \) from first principles
\( K(Tm) \) experimental/\( K(Tm) \) from first principles.
where the calculation of $K(Tm)$ from first principles, is dependent on the type of sequence being analyzed.

$K(Tm)$, from first principles, is calculated using one of the following equations.

**Self complementary sequences**

\[
K(T) = \frac{1}{n^*(Ct/2n)^{n-1}}
\]

**Non-self complementary sequences**

\[
K(T) = \frac{1}{(Ct/2n)^{(n-1)}}
\]

**Reports—Thermal**

Use the **Reports** page to set up the report parameters associated with data collection.

From the **Setup** menu, select **Reports** to access the **Reports** page. You can also access the page from the **Setup** dialog by selecting the **Reports** tab. To access the **Setup** dialog, select the **Setup** button from the Thermal Application window.

**Hot Tip**

You can view your generated report before you print it out using the **Print Preview** command in the **File** menu.

If you decide to make changes to the report layout, select **(Edit Report)** from the **Report** toolbar. This enables you to directly edit the report.

The page comprises the following:

**Operator**

**Name**  Enter the instrument operator name in this text entry field. Use the arrow to the right of this field to view and select one of the last ten entries in this field.

**Comment**  Enter operator details and comments in this text entry field.

**Options**
This group enables you to select the information to include in your report.

**Auto Print**  
Select **Auto Print** to print a report automatically during the data collection.  
If **Auto Print** is selected, then the system will send the report information to the specified printer(s) as well as the Report area. However if **Auto Print** is not selected, the report will only be sent to the Report area and can be viewed by selecting **Report** in the View menu.

To automatically obtain a copy of the graphics, you will need to select both the **Auto Print** and **Graph** options (see below). In this case, all graphical information displayed on screen will be printed with the report. Therefore it is advisable to arrange the graphical information as you require on screen, prior to printing.  
For example, if you require a single graph print out, then the **Single/Multi Graph** option in the **Graph** menu should be changed to the correct setting.

**Parameters**  
Select this option to include the method setup parameters within your report.

**User data form**  
Select this option to include in the report any information entered in the **User Data Form**.

**Graph**  
Select **Graph** to include any graphs or graphic information associated with your report.  
If you require a printout of the displayed graphics only, you must ensure that only this option is selected. Select the **Clear Report** button to clear any information in the Report area before you print otherwise this will be included in your printout.

**% Page Height**  
Enter the size at which you want the graph to be printed, as a percentage of total page height. For example, if you want graphical information to take up half of the page, enter 50% in this field.

**Company Logo**  
Check this option to include your company logo in the report.  
A company logo will only be included if you have added one on the Company page of the System Information Application.  

**Hot Tip**  
If you have not included a Company logo, select the Header/Footer button to open the System Information Application and use the Add Logo option to add a logo.

**Header/Footer**  
Select the **Header/Footer** button to open the System Information application where you can enter the Header and Footer information to be included in your report. You can also enter any additional comments, and add a logo to your report. Once you have made all your changes, select OK to exit the System Information application and return to the Reports page of the Thermal application.

**Focused trace**  
Select this option to apply the **Report** selections to the focused trace only.

**All traces**  
Select this option to apply the **Report** selections to all visible traces.

**X-Y Pairs Table**  
Select this if you want to print a table showing the X-Y pairs of the collected data in your report.
**Data Interval**  
*Actual*  
Select **Actual** to report the X-Y pairs table for the trace at the actual **Data Interval** at which the data was collected.

*Interpolated*  
Select **Interpolated** to report the X-Y pairs table for the trace at an interpolated data interval. Enter this data interval in the adjacent field. If no data has been collected at the interval you enter, the Cary Eclipse software will interpolate the data to produce an X-Y result.

**Status Display**

Select this check box to display the **Status Display** window, where you can view the status of your thermal run during the data collection.

**Hot Tip**  
You can copy a picture, spreadsheet file or document from another application (for example, Microsoft Word or Microsoft Excel) into the report area of the of the Thermal application. To edit these objects, simply double click on them to open the associated application in which they were created.

See also:  
*User Data Form*  
*View menu*  
*Status Display window*

**Report Toolbar—Thermal**

Use the Report toolbar buttons to gain quick access to many of the **Edit** menu options. You can also use the toolbar to cut, copy and paste information and to change the look of your report.

To display or hide the toolbar, point to the **Toolbars** option in the **View** menu and select **Reports**. To display or hide the toolbar's text labels, point to the **Toolbar** option in the **View** menu and select **Text Labels**.

**Hot Tip**  
To see the name of a button, point to it with the mouse.

The following commands can be accessed from the Reports Toolbar:

<table>
<thead>
<tr>
<th>Button</th>
<th>Menu command</th>
<th>Enables you to…</th>
</tr>
</thead>
</table>
|       | **Edit Report** | Click this button to edit the report.  
This command is also accessible by pressing CTRL+R. |
|       | **Undo** | If available, this undoes the last action.  
This command is also accessible by pressing CTRL+Z. |
|       | **Cut** | Click this button to remove the highlighted text and place it on the Windows Clipboard. |
Clear Report—Thermal

Select the Clear Report command to remove the contents of the Report Area in the Thermal Application window. If you have saved the report, selecting this option does not delete the contents of the report, the information is simply removed from the Report Area.

To access the Clear Report option, select the Clear Report command from the Commands menu, or select the Clear Report button from the Thermal Application window.

See also:
Thermal Application Commands menu

How to use advanced reporting options—Thermal

This outlines ways to fully customize your reports. Options available include:

- Including header and footer information
- Including a logo
- Editing the text in the report
- Viewing the report on screen before printing
- Cutting and pasting to a word processor

Including header and footer information

You can have text appearing at the top and bottom of every page in your report. To specify what the text should be, do the following:
Step 1

On the Windows Taskbar, click the Windows Start button, point to Programs, Cary Eclipse and then select System Information. The System Information application appears with the Company page displayed.

Step 2

Enter the header and footer information in the Header and Footer fields.

Step 3

Click OK to implement your changes.

Including a logo

You can include a logo on the first page of your report. To specify what the logo should be, do the following:

Step 1

On the Windows Taskbar, click the Windows Start button, point to Programs, Cary Eclipse and then select System Information. The System Information application appears with the Company page displayed.

Step 2

Enter a logo using the Add Logo button. This logo will always be included on the front page of reports.

Step 3

Click OK to implement your changes.

Q. Why isn't the logo I selected included in my report?
A. If you are running the Cary Eclipse software under Windows NT you need to be logged on as an Administrator in order to specify the logo in the System Information application so as to comply with GLP regulations.

Editing the text in the report

You can edit any of the text in the Report area. Just select Edit Report from the Edit menu. You can then add, delete and change text in the Report area.

Q. The Edit Report option is not available, why not?
A. You do not have reporting editing privileges set in the GLP Administration application. The Administrator of the system will have to change your privileges before you can edit reports.

Viewing the report on the screen before printing
You can view a report before printing it. To do this, from the **File** menu select **Print Preview**. You can then ensure that the report format is correct before you print it.

**Hot Tip**

Use the arrow keys at the bottom of the Print Preview screen to change pages.

**Cutting and pasting to a word processor**

To have complete control over how your report looks you may wish to copy it to a word processor program, such as Microsoft Word. To do this:

**Step 1**

Open a report.

**Step 2**

From the **Edit** menu, choose **Edit Report**.

**Step 3**

Click in the Report area and highlight the text you want to export to your word processor.

**Step 4**

Right-click on the Report area and select **Copy** from the menu.

**Step 5**

Start up your word processor and use the Paste function (usually under the **Edit** menu) to place the text into a document.

**Auto Store—Thermal**

Use the **Auto Store** page to specify if and when data will be stored, and in what format. You can also use this page to set up the system to automatically send an email at the end of a read.

From the **Setup** menu, select **Auto store** to access the **Auto store** page. You can also access the page from the **Setup** dialog by selecting the **Auto store** tab. To access the **Setup** dialog, select the **Setup** button from the Thermal Application window.

The page comprises the following:

**Storage**

**Off**

Select this if you do not wish to be automatically prompted to save collected data.
Note
If you wish to store your unsaved data manually, choose Save Data As… from the
File Menu to save your data after the run is complete. You can save your data as a
Batch or Data file.

On: Prompt
At Start  Select this to display the Windows Save As dialog at the start of the collect where you
can enter the Batch filename for your data.

On: Prompt
At End  Select this to display the Windows Save As dialog at the end of a collect where you can
enter the Batch filename for your data.

Auto convert

Use the Auto convert options to convert your data into an ASCII file format that can be opened in
spreadsheet programs such as Microsoft Excel.

None  Select None if you do not need to generate an ASCII file.

ASCII (csv)  Select this option to save the data as a comma delimited ASCII Spreadsheet file (*.CSV)
file. Only the currently collected data is converted.

ASCII files are stored in the same folder as the data file from which they were generated
and designated the same filename as the method used to collect the data plus the
extension ‘CSV’.

ASCII (csv)
with log  Select this option to save the data as a comma delimited ASCII Spreadsheet file (*.CSV)
file. Only the currently collected data is converted. The key method parameters used to
collect the data are also saved in this file.

ASCII files are stored in the same folder as the data file from which they were generated
and designated the same filename as the method used to collect the data plus the
extension ‘CSV’.

Note
If you choose to auto-convert the data to an ASCII file but you have set the
Storage option to Off, then the ASCII file will be saved with the following format:
DATE TIME. CSV

The date and time will appear in the same form that is set in the Windows
Regional Settings Properties dialog that is accessed from the Windows Control
Panel.

For example, 22_4_97 1;32;08.CSV

Email

Select the Email check box and enter the Email details to send an email to yourself or a colleague once
the read is completed. This useful facility can be used to alert you to the completion of the read,
particularly if you are working away from the laboratory where the Cary Eclipse instrument is located.
You can also attach the newly collected data to the email.
Status display

Select this check box to display the **Status Display** window, where you can view the status of your thermal run during the data collection.

**How to email your results—Thermal**

You may wish to send an email to a person once the read is completed. This useful facility can be used to alert you to the completion of the read, particularly if you are working away from the laboratory where the Cary Eclipse instrument is.

To do this:

**Step 1**

Ensure that you have set up the **Host** and **Port** options on the **Email options** page of the System Information application in order for the email facility to work correctly.

**Step 2**

From the **Setup** menu, select **Auto Store**. The **Auto Store** page is displayed.

**Step 3**

Select the **Email** check box to enable the email options.
Step 4

In the **To** field, enter the recipient's email address (for example, *rchandler@varianinc.com*). To enter multiple recipients, simply type a comma ',' between each email address.

Step 5

In the **From** field, enter the email address of the computer on which the Cary Eclipse software is installed. This may be your email address, or a communal email address, if the Cary Eclipse software is installed on a computer that has multiple users.

Step 6

Enter information in the **Subject** and **Message** fields as required.

Step 7

Select **Attach current data**. The data that has been collected during the run will be attached to the email.

**Hot Tip**

You can attach other documents to your email. To do this, select the **Attached** button and browse your computer to select a file, using the Windows® **Open** dialog that is displayed.

Step 8

Select **OK** to clear the **Auto Store** dialog. Your email will be sent once the run has been completed.

See also:

[Auto Store](#)

**Performing runs—Thermal**

- [Starting a run](#)
- [Accessory selection](#)
- [Sample Name dialog](#)
- [Cell Loading Guide](#)
- [Sync Start dialog](#)
- [Start/Stop buttons](#)
- [Go To dialog](#)
- [Go To Temperature dialog](#)
- [Rapid Result dialog](#)
- [Commands menu](#)
- [How to perform a run and determine Tm using Derivative calculations](#)
- [How to perform a run and determine Tm using Hyperchromicity calculations](#)

**Information about starting a run—Thermal**

The sequence that occurs when you select **Start** to commence the thermal run depends upon your storage
and accessory selections.

**Storage selections**

<table>
<thead>
<tr>
<th>If you have selected … as the Storage option on the Auto Store page</th>
<th>The following occurs</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>On, prompt at start</strong></td>
<td>The Windows Save As dialog will appear as soon as you select Start. You can save the current method, collected data, report and graph template as a Batch file.</td>
</tr>
<tr>
<td><strong>On, prompt at end</strong></td>
<td>The Windows Save As dialog will appear at the end of the data collection. You can save the current method, collected data, report and graph template as a Batch file.</td>
</tr>
<tr>
<td><strong>Off</strong></td>
<td>The Windows Save As dialog will not appear.</td>
</tr>
</tbody>
</table>

**Accessory selections**

<table>
<thead>
<tr>
<th>If you have selected … on the Accessories page</th>
<th>The following dialog is displayed</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>No accessories</strong></td>
<td>The Sample Name dialog will appear when you select Start, prompting you to load the sample and enter your sample name. Select OK to display the Sync Start dialog and commence the thermal run.</td>
</tr>
<tr>
<td><strong>Multicell Holder</strong></td>
<td>The Cell Loading Guide dialog will appear. Enter the sample name of each activated cell then select OK to display the Sync Start dialog. When you select OK to clear the Cell Loading Guide dialog, the cell changer will move to cell position 4, enabling you to easily access all of the cell positions and load your samples. <strong>If you need to start data collection immediately:</strong> Load the cells into the cell changer and select Reset Slide from the Sync Start dialog. The cell changer will move to cell position 1. You should then add the reactants to the cells as quickly as possible, close the lid and select OK on the Sync Start dialog. The collect will start immediately without performing another reset command. <strong>If you do not need to start the data collection immediately:</strong> Add the reactants to all the cuvettes and select OK on the Sync Start dialog. The system will automatically reset the cell changer to cell position 1 and start measurements on the selected cuvettes.</td>
</tr>
<tr>
<td><strong>Manual and Automated Polarizers</strong></td>
<td>The system will drive to the set temperature and then check for a valid G Factor. If the G Factor is not valid, the following prompt is displayed: ‘The current G Factor is not valid at the emission wavelength.</td>
</tr>
</tbody>
</table>
Sample Name—Thermal

The software selects the multicell accessory by default on opening the Thermal application.

If you are using a single cell peltier accessory, select the Go to radio button on the Cary tab and uncheck the multicell holder option on the Accessories tab in the Setup dialog. The Sample Name dialog appears once you select the Start button to commence an analysis.

The page comprises the following:

- **Sample Name** Enter the name of the sample to be used in the thermal run. Click on the arrow to the right of the field to view and select the most recently entered sample names. Once you have entered the Sample Name, select OK to display the Sync Start dialog and commence the thermal run.

See also:
- Cell Loading Guide
- Sync Start
- Start/Stop buttons
- Setup
- Accessories
- Auto Store

Cell Loading Guide—Thermal

If you are using a Multicell Holder (selected by default on opening the Thermal application), the Cell Loading Guide dialog appears once you select the Start button to commence an analysis.

The Cell Loading Guide dialog prompts you to insert blank samples in the Multicell Holder to collect the Thermal melts. You can also use the dialog to name the samples in each selected cell in the Multicell Holder.

The dialog comprises the following:

- **Loading Guide/Sample Name entry** Enter the names of the sample to be used in the thermal run in each available field. This sample name will be used as the Name of Trace in the Trace Preferences dialog.

**Note** The Cell Loading Guide dialog appears only if you have selected the Multicell Holder option on the Accessories page of the Setup dialog. Only cells selected in the Multicell Holder diagram on the
Accessories page will be available in the Loading Guide.

In the adjacent example, cells 1—3 will be available in the Loading Guide dialog, as they have been selected in the Multicell Holder diagram.

Once you have entered the Sample Name, select OK to display the Sync Start dialog and commence the Thermal run.

See also:
Accessories
Sample Name

Sync Start—Thermal

Use the Sync Start dialog to synchronize the start of a reaction and the start of the thermal data collection. Before displaying the Sync Start dialog, the Cary Eclipse will set up all the instrument and graphing parameters so that data collection can start as soon as the countdown finishes or you select OK.

The sequence for a Sync Start is as follows:

- Set up your data collection parameters
- Click the Start button (and load the samples if necessary)
- Once the Sync Start dialog is displayed, add your active reagent
- Commence the data collection by selecting the OK button

The dialog comprises the following:

**Count Down**  This field displays a two minute countdown in seconds. If you want to start your data collection before the two minute countdown ends select the OK button. Alternatively, you can wait for the countdown to end and the data collection will start automatically.

**Note**  If you are using a Multicell Holder and you select the OK button, the system will automatically Reset this accessory and immediately start the thermal run.

**Reset Slide**  Click this button to drive the Multicell Holder to its zero point (Cell position 1).

**Start/Stop Instrument buttons—Thermal**

The following buttons may be displayed at the top of the Thermal Application window. The **Start**, **Pause**, **Continue** and **Connect** buttons will change depending upon the status of the run. For example, if a run has not been started, the **Start** button will be visible to indicate that the system is ready to commence a run.

Select **Start** to commence a thermal run using the current Setup parameters. If any unsaved data exists in the Graphics area you will be prompted to save the data. If you choose not to save, any unsaved data, pictures and labels will be deleted when you start the thermal run.
When a thermal run is started, the **Start** button will change to **Pause**.

The **Pause** button is displayed in place of the **Start** button once you have selected **Start** to commence a run. Click **Pause** to temporarily suspend the data collection, so that you can modify your solutions during the thermal run. While the system is in the paused state, it will continue to keep track of the elapsed time, however data collection will stop until the **Continue** button is selected.

The benefit of using **Pause** instead of **Stop** is that you are able to add a secondary substrate to your thermal solution(s) and then continue to monitor a secondary reaction, without having to Stop and Start the collect again. It also means that you will not get a spike in your data when you open the sample compartment to add the reagent.

When a thermal run is paused, the **Pause** button will change to **Continue**.

The **Continue** button is displayed in place of the **Pause** button when you have paused the run. Click **Continue** to proceed with the thermal run after it has been paused.

Once you select **Continue**, is selected the button changes back to **Pause**.

If another application is already online with the Cary Eclipse, the **Start** button will change to **Connect**, and you will need to select **Connect** to bring this application online before the **Start** button will re-appear. (You will not be able to Connect if the other application is collecting data.)

Select this to end a thermal run.

If you have selected **Storage On** in the **Auto Store** page of the **Setup** dialog, all data collected is automatically stored in the file specified in the Batch name field of the Windows **Save As** dialog before or after the run commenced.

See also:
- [Graphics area](#)
- [Commands menu](#)

### Zero—Thermal

Use the **Zero** button to zero the current intensity value. The intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before commencing a data collection if you want your data to start at 0 intensity. The Cary Eclipse will prompt you to load a blank before performing the zero.

Before you select **Zero**, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.

When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the Thermal Application window.

Select the **Clear Zero** command from the **Commands** menu to remove zero correction and thus remove the subtraction effect.
To access the **Zero** option, select the **Zero** command from the **Commands** menu, or select the **Zero** button from the Thermal Application window.

**Go to Wavelength—Thermal**

Use the **Go to Wavelength** dialog to temporarily change the instrument to a new excitation or emission wavelength. You would mainly use this dialog if you want to quickly go to a wavelength and check the intensity reading of the sample. The lamp will flash continuously in this mode. This option is only available if the instrument is not currently collecting data.

To access the **Go to Wavelength** dialog, select the **Go To Wavelength** command from the **Commands** menu. You can also select **F4** to open the dialog.

**Note**

This is not the wavelength used in the application method. If your method uses a wavelength that is different from the ‘Go to wavelength’ setting you select, then when you select Start the system will change to the wavelength selected. (In most applications, this is set in the Cary page of the Setup dialog.)

The Go to Wavelength page comprises the following:

**Excitation**

**Wavelength**

Enter the new wavelength in this numeric entry field. When you select **Go to** the instrument will change to the new value.

**Zero Order**

Select **Zero Order** to drive the Cary Eclipse fluorescence spectrophotometer to 0 nm (white light). You can then use this light for aligning accessories.

**Emission**

**Wavelength**

Enter the new wavelength in this numeric entry field. When you select **Go to** the instrument will change to the new value.

**Zero Order**

Select **Zero Order** to drive the Cary Eclipse fluorescence spectrophotometer to 0 nm (white light). You can then use this light for aligning accessories.

**Go To**

Click **Go to** once you have entered a new wavelength, to change the instrument to the entered value.

See also:

**Commands menu**
**Function Keys**

**Go to Temperature—Thermal**

Use the **Go to Temperature** dialog to change the current temperature to a new value.
To access the **Go to Temperature** dialog, select the **Set temp** button from the Thermal Application window or the **Set temp** command from the **Commands** menu.

The **Set Temp** button is only accessible if you are using a thermostatted accessory. (On the **Accessories** page, the **Temperature control** option must be selected.)

You can access the **Go to Temperature** dialog prior to starting a run or once a run is in progress.

This dialog enables you to drive the Multicell Holder or Single Cell Holder block to the temperature entered in the Go to Temperature field.

**Hot Tip**
By using the Temperature probe accessory, you can determine the exact temperature of the solution within the cell. The start will be delayed until the solution has reached the required temperature.

**Hot Tip**
You can use this dialog to equilibrate your samples at the Start temperature before performing a Temperature run. Normally, the Multicell Holder or Single Cell Peltier will drive to the Start temperature when you press OK on the Setup dialog. If you then press Start immediately, the Cary will start collecting data soon as it determines the block temperature has reached the set value. However, this may not give the sample time to equilibrate.

**To change the current temperature of a run in progress:**

1. Access the **Set Temp** button by selecting the **Pause** button to pause the run. You can then select the **Set Temp** button.

2. In the **Set temp** field, enter the temperature you want the instrument to change to. Select **OK** to return to the Thermal Application window.

3. Select **Continue** to proceed with the run.

**See also:**
Thermal Application

**Rapid Result—Thermal**

Use the **Rapid Result** dialog to take a reading of a blank and sample/s, without having to change the current application method. You can use the **Multi Cell Changer** dialog to specify the cell you want to read. This means that between runs, while new samples are being prepared and loaded, other samples can quickly be measured. The result is printed in the Report area.

To access the **Rapid Result** dialog, select the **Rapid Result** command from the **Commands** menu or press F6.

**Excitation**

**Wavelength**
Enter the wavelength at which you want to take a reading.

**Zero order**
Select this option to set the excitation monochromator to zero order (white light).
**Use the Commands menu to access major operational commands in the Thermal application.** The menu is accessible from the menu line at the top of the Thermal Application window.

Alternatively you can use function key shortcuts for some of the commands. These appear next to the item in the Commands menu. Note that not all commands appear as both button and menu items.

**Emission**  
*Wavelength*  
Enter the wavelength at which you want to take a reading.

**Zero order**  
Select this option to set the excitation monochromator to zero order (white light).

**Zero**  
Click **Zero** to change the wavelength to the entered value and measure a blank.

**Result**  
Click **Result** to change the wavelength to the entered value and measure a sample. The X,Y results are corrected for the blank and the results are displayed in the Report window.

See also:  
*Multi Cell Changer*

**Commands menu—Thermal**

Use the Commands menu to access major operational commands in the Thermal application. The menu is accessible from the menu line at the top of the Thermal Application window.

Alternatively you can use function key shortcuts for some of the commands. These appear next to the item in the Commands menu. Note that not all commands appear as both button and menu items.

**Hot Tip**  
If you access these commands using the Commands menu, rather than from the application buttons, you can remove these buttons from your application window. To do this, clear the Buttons command in the View menu. This will enlarge the Graphics area, making viewing and selection of thermals easier.

Tell me how to remove or display the command buttons

The Commands menu comprises the following commands:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Start</strong></td>
<td>Select this to start a data collection using the currently set up parameters. If the Connect command is displayed in place of the Start command, this indicates that another Cary Eclipse application is in use and has control of the Cary Eclipse instrument. To make the Thermal application active so that you can perform a read, you will need to select Connect. However, you will not be able to select Connect if another application is collecting data.</td>
</tr>
<tr>
<td><strong>Stop</strong></td>
<td>Select this to stop the data collection. If you have selected Storage On in the Auto Store page of the Setup dialog, all data collected is automatically stored in the file specified in the Batch name field of the Windows Save As dialog before or after the run commenced.</td>
</tr>
<tr>
<td><strong>Clear Zero</strong></td>
<td>Select this option to clear the Zero command. When you select the Zero command (from the Commands menu or by pressing F5), the intensity at that time will be reset to zero and all subsequent results will have the intensity value that was reset to zero subtracted from them. Selecting Clear zero cancels this zeroing (and thus subtraction) effect.</td>
</tr>
</tbody>
</table>
Zero order… Select the Zero order command to set one of the monochromators to zero order. If you use this command to zero either the excitation or emission monochromators, this will override the Ex. Wavelength or Em. Wavelength set on the Cary page. The Cary Eclipse software will use the zero setting when collecting data.

The Zero order command is generally used during alignment procedures.

**Excitation**
Select this command to set the excitation monochromator to zero order (white light).

**Emission**
Select this command to set the emission monochromator to zero order (white light).

<table>
<thead>
<tr>
<th>Reset</th>
<th>Select this command to re-initialize the instrument. This will perform the same initializing procedure that is performed when the instrument is powered on.</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Rapid Result</strong></td>
<td>Select this to display the Rapid Result dialog. Use this dialog to take a reading of a blank and multiple sample readings without changing the application method. This means that between runs, while new samples are being prepared and loaded, other samples can quickly be measured. This command can also be accessed by pressing F6.</td>
</tr>
<tr>
<td><strong>Zero</strong></td>
<td>Select this to zero the current intensity value. The intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before selecting Start if you want your data to start at 0 intensity. This command is also accessible by pressing F5.</td>
</tr>
</tbody>
</table>

**Note**
Before you select Zero, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.

| **Go to…**             | Select Go To… to display the Go To Wavelength dialog where you can change the instrument to a new wavelength. Use this command if you want to quickly go to a wavelength to check the intensity reading. This command is only available if the instrument is not currently collecting data. It can also be accessed by pressing F4. |

**Note**
This is not the wavelength used in the application method. If your method uses a wavelength that is different from the 'Go To wavelength', when you select Start, the system will change to the wavelength selected in the current method. (In most applications, this is set in the Cary page of the Setup dialog.)

| **Set temp**            | Select Set temp to open the Set Temperature dialog where you can change the temperature during a thermal run. |

The Set temp command button is only accessible if you are using a thermostatted accessory. (On the Accessories page, the Temperature control option must be selected.) Once you have started a run, select the Pause button to pause the run and then select the Set temp command.

| **ADL Command**         | Select this to display the ADL Command Line window where you can execute an ADL command or ADL program. (You can also press Alt-Y to display this dialog.) |

| **Rerun ADL**           | Select this to run the ADL command or program in the ADL Command Line window again. (You can also press Alt-X to rerun programs.) |
How to perform a run and determine Tm using Derivative calculations—Thermal

This demonstrates how to perform a Thermal run using a simple collect (single stage or single ramp rate) and automatically calculate Tm from the first derivative.

Follow the steps through adjusting them to meet the requirements of your particular Thermal run.

- Start the Thermal application
- Set up data collection parameters
- Set up instrument parameters such as wavelengths, slit width, etc.
- Set up accessories
- Set up a derivative method of analysis
- Set up your reporting and printing requirements
- Set up storage of collected data
- Set up visual system monitoring
- Finish Setup
- Equilibrate the sample
- Start the Thermal run
- Cary actions after the run
- Save your data

Start the Thermal application

Step 1

On the Windows Taskbar click Start to display the Start Menu.

Step 2

Move the cursor over the Programs menu item to display the Programs menu.

Step 3

Select Thermal in the Cary Eclipse menu.

Step 4

Select your instrument type (if necessary) and press OK to open the Thermal application.
(Alternatively, you can double click on the Thermal icon in the Cary Eclipse folder on the desktop.)

**Note**
If you are running a GLP system, you may be prompted to enter a password before accessing the Thermal application.

**Set up data collection parameters**

**Step 5 [Setup dialog]**

Select the Setup menu or click the Setup… button to display the Setup dialog and specify the method parameters for a new method.

**Set up instrument parameters such as wavelengths, slit width, etc.**

**Step 6 [Setup dialog | Cary tab]**

a. Select the Cary tab and specify the instrument set-up for your analysis. For example selecting Fluorescence from the drop down menu.

b. In the Wavelength setup enter the excitation and emission wavelengths that you want monitored.

c. In the Wavelength setup enter the required excitation and emission slit widths.

**Step 7 [Setup dialog | Cary tab] [Collect Temperatures] Field**

a. Select the Ramps radio button.

b. Select Simple Collect to set a single rate of temperature change for the entire Thermal run. Select Advanced Collect for more than one stage of temperature ramping.

**Note**
For slow changes in temperature (i.e. slow ramp rates) it is recommended that a long Ave Time (e.g. 2 seconds) is set.

b. Set up the rate parameters such as the start and stop temperatures, the rate of temperature ramping, and the hold times at the start and end temperatures.

d. In the Ave Time field enter the time(s) you require the instrument to excite the sample and record an emission for each data point.

e. In the Cary tab specify the Collect Temperatures parameters.

**Note**
Once you press OK to exit the Setup dialog, the Cary will drive the temperature to the specified Start Temperature. The Start button becomes enabled once the monitoring device (Temperature probe or block probe) indicates that the temperature is within 0.5°C of the specified start temperature.

**Step 8 [Setup dialog | Options tab]**
a. In the Display Options group, select the way in which you want the data displayed as it is collected. Choose the Overlay Data radio button to superimpose the collected data of each sample in the Thermal run in one graph box.

b. Deselect the Overlay Data radio button to plot individual graphs for each thermal melt sample.

c. Adjust the PMT voltage if necessary to increase or decrease the fluorescence intensity recorded.

**Note**

If you are unsure of the expected change in fluorescence intensity during your thermal ramp, you can use the Set Temperature button to quickly ramp the temperature to a value at which you expect the intensity to be largest. Then adjust the PMT HV so that the intensity at this temperature is approximately 750 a.u..

**Set up accessories**

**Step 9 [Setup dialog | Analyze tab] Lost-run smoothing**

a. Select the Analyze tab.

b. Select Smoothing and nominate the smoothing Interval and Filter Size to control the number of measurement points to be averaged if required.

**Set up a derivative method of analysis**

**Step 10 [Setup dialog | Analyze tab] Lost-run smoothing**

a. Select the Derivative radio button to choose the derivative method to calculate the temperature at which 50% of the DNA strands have separated (Tm).

b. Select the Autocalculate checkbox to automatically perform a derivative calculation on collected data at the end of each run.

**Note**

You can also perform the derivative calculation manually, once the run is completed.

c. Now set the parameters to define the range over which to calculate and plot the first derivative of the selected thermal. To do this, first enter the Interval at which data is sampled in order to perform a derivative calculation.

d. A derivative calculation involves the collection of points from a plot. Selection of these points is dependent upon the data interval chosen. The data points are then used to generate a separate trace from which a derivative calculation is performed. The selection of the Derivative Interval is dependent on the rate of data collection. If data is collected every 0.1 °C then a derivative interval of 0.1 °C will give the most accurate result. Enter the Filter Size you require for the derivative calculation.

The derivative uses a Savitzky Golay technique where a number of points surrounding an individual point are averaged to produce a new, smoothed point. For example, a five point derivation uses the two data points before and after each data point.
Note
The larger the filter size, the more data points that are included in the filter for smoothing.

e. In the Low Calculation Limit entry field, enter the temperature where you want to start the calculation of the derivative for the plot.

f. In the High Calculation Limit entry field, enter the temperature where you want to end the calculation of the derivative for the plot.

Set up your reporting and printing requirements

Step 11 [Setup dialog | Report tab]
Select the Reports tab and specify your reporting requirements.

a. Enter your name in the Name entry field.

b. Enter a comment relating to your experiment in the Comment entry field.

c. Set up your report style by selecting the appropriate radio buttons in the Options group.

   For example, select the Auto Print checkbox to obtain a printout of your report automatically. Select the Parameters checkbox to include your method parameters in the report. Select the Graph checkbox to include the graphics in the generated report.

Note
If Auto Print is selected, then the system will send the report information to the specified printer as well as the Report area. However if Auto Print is not selected, the report will only be sent to the Report area and can be viewed by selecting the Report option in the View menu.

d. Select the Autoconvert option you require.

   If you choose Select for ASCII (csv) or Select for ASCII (csv) with Log, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary format as well as ASCII XY pairs format in the current folder.

Set up storage of collected data

Step 12 [Setup dialog | Auto Store tab]

a. Select the Auto Store tab.

b. Select Storage Off.

Set up visual system monitoring

Step 13 [Setup dialog]
Select the Show Status Display checkbox to display various information fields on your current reaction.
**Finish Setup**

**Step 14**

Once you are satisfied with your method setup click OK to save any changes made and close the Setup dialog. The Cary will start to dive the temperature to the Start temperature.

**Equilibrate the sample**

**Step 15**

Make sure that the Cary has reached the Start temperature and has been at this temperature for 2–3 minutes.

**Start the Thermal run**

**Step 16**

a. Push the Start button to start a data collection. Alternatively you can select Start in the Commands menu. The system will display a Loading Guide dialog box.

b. Load your sample into the appropriate cell holder.

c. Enter the name of your sample in the text entry field then press OK.

The Cary will wait for the specified Hold time and then perform the Thermal run.

**Cary actions after the run**

**Step 17**

If Autocalculate (Derivative group) was selected on the Analyze tab of the Setup dialog box, a derivative calculation will be performed at the end of the data collection.

The sample under investigation, with corresponding Tm values, will appear in the Report area as well as parameters used to generate the selected data file, Temperature profile, and the method used to contain the Tm value.

A plot will appear in the Graphics area indicating the original temperature data as well as a plot of the first derivative of the selected thermal. The point at which the derivative plot peaks (i.e. maximum or minimum gradient on the original plot) is reported as the Tm value and the original plot is marked with an arrow to indicate this point.

To view a plot of the collected data, from the View menu select the Graph option.

You may use the Recalculate button to reanalyze your data using different parameters if you wish.

**Save your data**

**Step 18**
a. Click on the File menu item and select Save Data As…

b. Click in the Files of Type field and select Batch.

c. Enter the file name for this Thermal run in the File name field.

d. Click Save.

The data will be stored as a Batch file.

See also:
- Example of Thermal trace with derivative
- How to perform Derivative calculations from previously stored data

**How to perform a run and determine Tm using Hyperchromicity calculations— Thermal**

This demonstrates how to perform a Thermal run with multiple temperature ramps and determine Tm using a Hyperchromicity calculation.

Follow the steps through, adjusting them to meet the requirements of your particular Thermal run.

- Start the Thermal application
- Set up the data collection parameters
- Set up instrument parameters such as wavelengths, slit width, etc.
- Set up accessories
- Set up your hyperchromicity method of analysis
- Set up your reporting and printing requirements
- Set up storage of collected data
- Set up visual system monitoring
- Finish Setup
- Equilibrate the Samples
- Start the Thermal run
- Cary prompts after the run
- Save your data
- Hyperchromicity calculations

**Start the Thermal application**

**Step 1**

On the Windows Taskbar click Start to display the Start Menu.

**Step 2**

Move the cursor over the Programs menu item to display the Programs menu.

**Step 3**

Select Thermal in the Cary Eclipse menu.
Step 4

Select your instrument type (if necessary) and press OK to open the Thermal application. (Alternatively, you can double click on the Thermal icon in the Cary Eclipse folder on the desktop.)

Note
If you are running a GLP system, you may be prompted to enter a password before accessing the Thermal application.

Set up data collection parameters

Step 5 [Setup dialog]

Select the Setup menu or click the Setup… button to display the Setup dialog and specify the method parameters for a new method.

Set up instrument parameters such as wavelengths, slit width, etc.

Step 6 [Setup dialog | Cary tab]

a. Select the Cary tab and specify the instrument set-up for your analysis. For example selecting Fluorescence from the drop down menu.

b. In the Wavelength setup enter the excitation and emission wavelengths that you want monitored.

c. In the Wavelength setup enter the required excitation and emission slit widths.

Step 7 [Setup dialog | Cary tab] [Collect Temperatures] Field

a. Select the Ramps radio button.

b. Select Simple Collect to set a single rate of temperature change for the entire Thermal run. Select Advanced Collect for more than one stage of temperature ramping.

Note
For slow changes in temperature (i.e. slow ramp rates) it is recommended that a long Ave Time (e.g. 2 seconds) is set.

c. Set up the rate parameters such as the start and stop temperatures, the rate of temperature ramping, and the hold times at the start and end temperatures.

d. In the Ave Time field enter the time(s) you require the instrument to excite the sample and record an emission for each data point.

e. In the Cary tab specify the Collect Temperatures parameters.

Note
Once you press OK to exit the Setup dialog, the Cary will drive the temperature to the specified Start Temperature. The Start button becomes enabled once the monitoring device (Temperature probe or block probe) indicates that the temperature is within 0.5°C of the specified start temperature.
Step 8 [Setup dialog | Options tab]

a. In the Display Options group, select the way in which you want the data displayed as it is collected. Choose the Overlay Data radio button to superimpose the collected data of each sample in the Thermal run in one graph box.

b. Deselect the Overlay Data radio button to plot individual graphs for each thermal melt sample.

c. Adjust the PMT voltage if necessary to increase or decrease the fluorescence intensity recorded.

**Note**
If you are unsure of the expected change in fluorescence intensity during your thermal ramp, you can use the Set Temperature button to quickly ramp the temperature to a value at which you expect the intensity to be largest. Then adjust the PMT HV so that the intensity at this temperature is approximately 750 a.u..

Set up accessories

Step 9 [Setup dialog | Analyze tab] Lost-run smoothing

a. Select the Analyze tab.

b. Select Smoothing and nominate the smoothing Interval and Filter Size to control the number of measurement points to be averaged if required.

Set up your hyperchromicity method of analysis

Step 10 [Setup dialog | Analyze tab]

a. Check the Hyperchromicity option to calculate the temperature at which 50% of the DNA strands have separated (Tm).

**Note**
The hyperchromicity calculation is performed once the run is completed as the user needs to select their region of interest from the graph.

b. Check Vant Hoff Calculation to calculate a van't Hoff plot (Ln(K(T)) vs 1000/T(deg K)) and activate the Hyperchromicity group.

c. Select the Self Complementary radio button if the DNA molecule being analyzed consists of identical strands. Otherwise, select Non Self Complementary.

d. Enter the concentration in the Total Conc of Strands entry field.

e. Enter the Molecularity (i.e. the number of strands) of the hybrid. For example, enter '2' in this field if the DNA molecule being analyzed consists of two strands.

f. Check the Delta G checkbox to perform Delta G (ΔG or free energy) and K (equilibrium constant) calculations.
g. In the Number of Temperatures field that is now accessible, specify the number of temperatures at which you would like to perform the $\Delta G$ and $K$ calculations. The number set here is reflected in the Temperature table below this field.

h. Enter the temperature at which you would like to calculate $\Delta G$ or $K$ in each cell in the Temperature table.

**Set up your reporting and printing requirements**

**Step 11 [Setup dialog | Report tab]**

Select the Reports tab and specify your reporting requirements.

a. Enter your name in the Name entry field.

b. Enter a comment relating to your experiment in the Comment entry field.

c. Set up your report style by selecting the appropriate radio buttons in the Options group.

For example, select the Auto Print checkbox to obtain a printout of your report automatically. Select the Parameters checkbox to include your method parameters in the report. Select the Graph checkbox to include the graphics in the generated report.

**Note**

If Auto Print is selected, then the system will send the report information to the specified printer as well as the Report area. However if Auto Print is not selected, the report will only be sent to the Report area and can be viewed by selecting the Report option in the View menu.

d. Select the Autoconvert option you require.

If you choose Select for ASCII (csv) or Select for ASCII (csv) with Log, then at the end of the data collection the system will automatically generate a report and store the data both in the Cary format as well as ASCII XY pairs format in the current folder.

**Set up storage of collected data**

**Step 12 [Setup dialog | Auto Store tab]**

a. Select the Auto Store tab.

b. Select Storage Off.

**Set up visual system monitoring**

**Step 13 [Setup dialog]**

Select the Show Status Display checkbox to display various information fields on your current reaction.

**Finish Setup**
Step 14

Once you are satisfied with your method setup click OK to save any changes made and close the Setup dialog. The Cary will start to dive the temperature to the Start temperature.

Equilibrate the sample

Step 15

Make sure that the Cary has reached the Start temperature and has been at this temperature for 2–3 minutes. If you enter a hold time in the Stage 1 Hold field, the system will wait and equilibrate at this temperature for the hold time before proceeding with the ramp. It is not necessary to wait to press the Start button.

Start the Thermal run

Step 16

a. Push the Start button to start a data collection. Alternatively you can select Start in the Commands menu. The system will display a Loading Guide dialog box.

b. Load your sample into the appropriate cell holder.

c. Enter the name of your sample in the text entry field then press OK.

The Cary will wait for the specified Hold time and then perform the Thermal run.

Cary prompts after the run

Step 17

a. At the end of the run, depending on what you have set in Step 14, the User Data Form may appear. If it does, then fill-in the appropriate entries in the columns provided for each sample trace and press OK. The calculation equation will appear in the report.

b. Next the Cary will prompt you to use the Thermal ruler to define various limits for the hyperchromicity calculations. (See the following sections).

Save your data

Step 18

a. Click on the File menu item and select Save Data As…

b. Click in the Files of Type field and select Batch.

c. Enter the file name for this Thermal run in the File name field.

d. Click Save.
The data will be stored as a Batch file.

**Hyperchromicity calculations**

**Step 19**

a. An Attention dialog will appear asking you to select the Associated DNA limits using the Ruler. Press the OK button on this dialog. The cursor changes to ruler mode.

b. Place your mouse on the Thermal curve at one end of the linear region of the Associated DNA part of the curve (prior to transition). Click and hold down the left mouse button, then drag the mouse along the curve and release the left mouse button at the other end of the linear region.

   The software will automatically calculate a linear least squares line between the selected points and extrapolate to both ends of the melting curve.

c. An Associated DNA Limits dialog will appear displaying the upper and lower limits which you selected with the Thermal Ruler. Press the OK button to accept the calculated line.

d. An Attention dialog will then appear asking you to select the Deassociated DNA limits using the Ruler, press the OK button on this dialog. The cursor changes to ruler mode.

e. Place your mouse on the Thermal curve at one end of the linear region of the Deassociated DNA part of the curve (after the transition). Click and hold down the left mouse button, then drag the mouse along the curve and release the left mouse button at the other end of the linear region.

   The software will automatically calculate a linear least squares line between the selected points and extrapolate to both ends of the melting curve.

f. A Dissociated DNA Limits dialog will appear displaying the upper and lower limits which you selected with the ruler. Press the OK button to accept the calculated line. The Linear Region Associated and Dissociated correction points are displayed in the Report area.

**Note**

The Associated and Dissociated DNA Limits dialogs allow you to re-select your limits if you are not satisfied with the linear least squares lines drawn in the graph box. To do this, click the Retry button on either dialog and select another region. If you wish to exit from the calculation mode, click Cancel. The ruler trace is removed from the graph.

**Note**

If the two extrapolated lines cross at any point, a warning will appear. Click Cancel and repeat steps (a) to (f) above and re-select the linear regions. Click Ignore to proceed with the calculation using the regions you have selected.

Using the lines calculated in the previous steps the software will now automatically create a second graph box and display the calculated Alpha curve.

g. An Attention dialog will appear asking you to select the range on the Alpha curve which will be used for the Tm calculation. Press the OK button to make your selection.

h. Place your mouse on the alpha curve at one end of the linear region. Click and hold down the left
mouse button, then drag the mouse along the curve and release the left mouse button at the other end of the linear region.

i. An Alpha Tm Limits dialog box will appear displaying the selected limits. Click the Retry button to re-select the limits if required. Press the OK button to accept the selected ranges.

A linear least squares line is drawn between the selected points and the value of temperature where \( a = 0.5 \) is calculated to give the Tm value. (The Tm value is automatically reported in °C and K.)

If you have chosen the van't Hoff Calculation method to calculate Delta G and K:

j. Using the data calculated in step (j) above, the system automatically creates a third graph box displaying a van't Hoff plot. An Attention dialog will appear asking you to select the required range on the van't Hoff plot to calculate Delta G and K. Press the OK button on this dialog.

k. Place your mouse on the van't Hoff curve at one end of the linear region. Click and hold down the left mouse button, then drag the mouse along the curve and release the left mouse button at the other end of the linear region.

l. A van't Hoff Limits dialog box will also appear displaying the upper and lower limits that you selected with the ruler. Press the OK button to accept the calculated line. Alternatively you can select the Retry button to re-select the limits using the ruler.

**Note**

If there is an error in the van't Hoff calculation you will be prompted to re-select the linear region.

A linear least squares line is calculated between these points and is extrapolated to both ends of the van't Hoff plot. The slope and intercept of the calculated line gives the enthalpy and entropy of the hybrid formation (\( \Delta H^\circ \) and \( \Delta S^\circ \) respectively).

The software will automatically send the results, using the calculated alpha curve, to the Report area including:

- \( K(T_m) \) experimental
- \( K(T_m) \) from first principles
- \( K(T_m) \) experimental/\( K(T_m) \) from first principles

**Step 20**

Step 20 will be repeated for each sample trace measured.

See also:
- Hyperchromiciry ruler steps
- How to use the ruler during Hyperchromicity calculations
- How to perform Hyperchromicity calculations on previously stored data

**How to use the ruler during Hyperchromicity calculations—Thermal**

You are required to use the ruler during Hyperchromicity calculations after Thermal data is collected to
To use the ruler:

**Step 1**
Select a region of the trace that is relatively straight and then place the cursor on the first point on the trace where you wish to start drawing the ruler.

**Step 2**
Press and hold down the left mouse button.

**Step 3**
Move the cursor to the second point on the trace. You will see the ruler line drawing as you move the cursor.

**Step 4**
Release the mouse button.

The Limits dialog box for the currently displayed curve appears enabling you to view the X value of the start and end positions of the region that you selected.

If you are not satisfied with the selected range, click the Retry button to repeat Steps 3 to 6 and select another range.

**Note**
If you wish to exit from the Ruler mode, click Cancel. The ruler trace is removed from the graph.

**Step 5**
Click the OK button.

See also:
- Hyperchromicity ruler steps
- How to perform a run and determine Tm using Hyperchromicity calculations
- How to perform Hyperchromicity calculations on previously stored data
Viewing run progress and results—Thermal

- Reports page
- Reports toolbar
- View menu
- Edit menu
- Status display
- Print Preview dialog
- Print

View menu—Thermal

The View menu is accessible from the menu line at the top of the Thermal Application window.

Hot Tip
Highlight a method or data file in Explorer and simply drag and drop it onto the application to open it. Any data will be displayed in the Graphics area.

The following commands can be accessed from the View menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Toolbar</td>
<td>Select this option to toggle the Graphics and Reports toolbar on and off. You can also select to display or hide the toolbar Text Labels.</td>
</tr>
<tr>
<td>Buttons</td>
<td>Select this option to toggle the left-hand button display (button area) on and off.</td>
</tr>
<tr>
<td>Graphics</td>
<td>Select this option to toggle the Graphics area on and off.</td>
</tr>
<tr>
<td>Report</td>
<td>Select this option to toggle the Report area on and off.</td>
</tr>
<tr>
<td>Report Vertical</td>
<td>If the Graphics option is selected, select Report Vertical to display the Graphics area, Report area and/or ADL Editor side by side vertically.</td>
</tr>
<tr>
<td>ADL</td>
<td>Select this option to toggle the ADL Editor on and off.</td>
</tr>
<tr>
<td>ADL Vertical</td>
<td>If the ADL and Report options are selected, select ADL Vertical to display the ADL Editor vertically on the right hand side of the Report area.</td>
</tr>
<tr>
<td>ADL Quick Reference</td>
<td>This displays the Quick Reference window where you can easily add a command in the correct syntax to your ADL program. A detailed description of all the commands in the Quick Reference is available by selecting the Help button.</td>
</tr>
<tr>
<td>ADL Hooks</td>
<td>Select this to activate a link (or hook) to an ADL program at a specific point in the application, and then run that ADL program. For example, ADL hooks have been supplied to run specific programs:</td>
</tr>
<tr>
<td></td>
<td>- on start-up of the Cary Eclipse software;</td>
</tr>
<tr>
<td></td>
<td>- when the system goes online or offline;</td>
</tr>
<tr>
<td></td>
<td>- when the Start button is selected;</td>
</tr>
<tr>
<td></td>
<td>- when a run finishes;</td>
</tr>
<tr>
<td></td>
<td>- when a zero is performed etc.</td>
</tr>
</tbody>
</table>
For the hook to work, **ADL Hooks** must be selected, and the appropriate ADL file for the application must be present in the ADL folder in the directory.

**Note**
Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.

### Hints
Select this option to toggle the Hints bubble text on and off. The bubble text appears when you place the arrow cursor over a field, button or text entry field for a short period. It offers a quick explanation of the item’s function.

### Status Display
Select this option to display the **Status Display** window, where you can view the status of various parameters during the data collection.

---

**See also:**
- **Thermal Application**
- **Graphics area**
- **Status Display**
- **ADL Hooks**
- **How to insert ADL commands using the Quick Reference**

## Edit menu—Thermal

Use the **Edit** menu to copy, cut, paste and delete information. You can also use the menu to access the Report editing commands. The menu is accessible from the menu line at the top of the Thermal Application window.

The following commands can be accessed from the **Edit** menu:

**Note**
Some options are only available once you select **Edit Report**.

<table>
<thead>
<tr>
<th>Command</th>
<th>Button</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Undo</td>
<td>🔄</td>
<td>If available, this undoes the last action. This command is also accessible by pressing CTRL+Z.</td>
</tr>
<tr>
<td>Cut</td>
<td>⌇</td>
<td>Select this command to remove the highlighted text and place it on the Windows Clipboard. This command is also accessible by pressing CTRL+X.</td>
</tr>
<tr>
<td>Copy</td>
<td>📄</td>
<td>Select this command to copy the highlighted text onto the Windows Clipboard. This command is also accessible by pressing CTRL+C.</td>
</tr>
<tr>
<td>Paste</td>
<td>📑</td>
<td>Select this command to insert the contents of the Windows Clipboard (text only) at the cursor. Any highlighted text will be replaced. This command is also accessible by pressing CTRL+V.</td>
</tr>
<tr>
<td>Delete</td>
<td>🗑️</td>
<td>Select this command to remove selected text in the Report area. This option is only available once you select <strong>Edit Report</strong> and there is text highlighted in the Report area. This command is also accessible by pressing DEL.</td>
</tr>
</tbody>
</table>
Use the Status Display window to view the status of your thermal run during the data collection. Depending upon settings you have for the Thermal run, the following parameters may be displayed:

- Excitation Wavelength (nm): 0.02
- Emission Wavelength (nm): 0.02
- Ex. Slit Width (nm): 5
- Em. Slit Width (nm): 5
- Excitation Filter: Auto
- Emission Filter: Open
- Emission PMT (V): 600
- Monitoring device: Block
- Block Temperature: 0.00
- Current Cell: 1

To access the Status Display window, select Status Display from the View menu, or from any page of the Setup dialog. You can choose to view the Status Display at any time during a thermal run.
Print Preview—Thermal

Use the Print Preview dialog to view the current report before you print it. From the File menu, select Print Preview to access the dialog.

The contents of the Graphics area appear first in the preview area followed by the contents of the Report area. You can reduce or enlarge the print preview on screen. To do this, click the left mouse button in the preview area to toggle through the five available views. Click the right mouse button in the preview area to return to the full page view.

The page comprises the following commands:

- **First arrow** Use this arrow to view the first page of the report.
- **Previous arrow** Use this arrow to view the previous page of the report.
- **Next arrow** Use this arrow to view the next page of the report.
- **Last arrow** Use this arrow to view the last page of the report.
- **Print** Use this to display the Windows Print dialog where you can choose the printer and pages to print as well as the number of copies you wish to print.

**Q.** Why are the arrow buttons grayed out?

**A.** The buttons will be grayed out if the report has only one page of data.

See also:

- [File menu](#)

Print—Thermal

Use the Print dialog to select the printer and pages to print of the currently displayed report as well as the number of copies you wish to print.

To access the Print option, select the Print command from the File menu, or select the Print button from the Thermal Application window.

All graphical information displayed on screen will be printed with the report, therefore it is advisable to edit the report on the screen prior to printing. You can also preview the report prior to printing by using the Print Preview command.

If you require a printout of the graphics in the Graphics area only, select the Clear Report button to erase the contents of the Report area before you print.

**Note**

The options on the Report page are used to generate the contents of the Report area during a run or on a recalculation. If Auto Print has been selected the Cary Eclipse will automatically print a report at the end of a data collection.
Performing Calculations—Thermal

- Recalculate
- How to perform post run calculations on previously stored data
- How to generate a report from previously stored data
- How to use advanced reporting options
- Analyze
- Maths Calculator
- Maths Operations
- Maths How To
- User Data Form

How to perform Derivative calculations on previously stored data—Thermal

- Retrieve the data
- Set up recalculation
- Set up for Derivative recalculation

Retrieve the data

Step 1

Select Open Data… from the File menu to display the Windows Open dialog box.

Step 2

A list of previously stored Batch files will appear. Select the Batch file you require then click the Open button.

The system will automatically retrieve the collected data into the Graphics area. The method that was used to collect the data will also be retrieved.

Step 3

Select the sample you want to investigate.

Note
Recalculations are only performed on 'Visible' traces.

Set up recalculation
Step 4

Click the Recalculate button to display the Setup dialog box. The Analyze, Calculations and Reports tabs will appear to allow you to set up the analysis, calculation and report parameters.

**Note**

If the selected sample contains results from a previous calculation a confirmation dialog will appear warning you that these will be deleted. Click OK to delete the calculations.

Step 5

Select the Reports tab to ensure that the method will report exactly what you require. For example, Parameters, Results etc.

Step 6

Select the Analyze tab to enter the recalculation parameters.

**Set up for Derivative recalculation**

Step 7

Select the Derivative radio button to choose the derivative method to calculate the temperature at which 50% of the DNA strands have separated (Tm). In the Interval entry field, specify the interval at which data is collected in order to perform a derivative calculation.

A derivative calculation involves the collection of points from a plot. Selection of these points is dependent upon the data interval chosen. The data points are then used to generate a separate trace from which a derivative calculation is performed.

The selection of the Derivative interval is dependent on the rate of data collection. If data is collected every 0.1 °C then a derivative interval of 0.1 °C will give the most accurate result.

Step 8 [Setup dialog | Calculations tab]

Select the Calculations tab to enter the calculation parameters.

Step 9

Click OK to exit the Setup dialog.

Step 10

Select the Derivative button and Derivative calculation fields as required.

Step 11

Click OK to exit the Set up dialog.

The software will calculate and plot the first derivative of the selected sample. The point at which the
derivative plot peaks (i.e. maximum or minimum gradient on the original plot) is reported as the Tm value and the original plot is marked with Tm to indicate this point. The calculation results will also be displayed in the Report area.

**Note**
The first derivative is also calculated and plotted for all other 'Visible' traces in the Graphics area. Select Single/Multi Graph in the Graph menu to toggle to the Multi-graph mode and view other samples in the current Batch file.

See also:
- [Example of Thermal trace with derivative](#)
- [How to perform Hyperchromicity calculations on previously stored data](#)

### How to perform Hyperchromicity calculations on previously stored data—Thermal

- [Retrieve the data](#)
- [Set up recalculation](#)
- [Set up other recalculations and normalizations](#)
- [Set up for Hyperchromicity recalculations](#)
- [Hyperchromicity recalculation procedure](#)

#### Retrieve the data

**Step 1**

Select Open Data… from the File menu to display the Windows Open dialog box.

**Step 2**

A list of previously stored Batch files will appear. Select the Batch file you require then click the Open button.

The system will automatically retrieve the collected data into the Graphics area. The method that was used to collect the data will also be retrieved.

**Note**
You can also retrieve data from other file types.

**Step 3**

Select the sample you want to investigate.

**Note**
Recalculations are only performed on 'Visible' traces

#### Set up recalculation
Step 4

**Hot Tip**
Before commencing, enlarge the Graphics area. To do this, double click on the Graphics area with the left mouse button.

Click the Recalculate button to display the Setup dialog box. The Analyze, Calculations and Reports tabs will appear to allow you to set up the calculation, analysis and report parameters.

**Note**
If the selected sample contains results from a previous calculation a confirmation dialog will appear warning you that these will be deleted. Click OK to delete the calculations.

Step 5

Select the Reports tab to ensure that the method will report exactly what you require. For example, Parameters, Results etc.

**Set up for Hyperchromicity recalculation**

Step 6

Select the Analyze tab to enter the recalculation parameters.

Step 7

Select the Hyperchromicity radio button to choose the Hyperchromicity method to calculate the temperature at which 50% of the DNA strands have separated (Tm).

a. Check the Vant Hoff Calculation to obtain a van't Hoff plot (Ln(K(T)) vs 1000/T(K)) and activate the Hyperchromicity group.

b. Select the Self Complementary radio button if the DNA molecule being analyzed consists of identical strands. Otherwise select Non Self Complementary.

c. Enter the concentration in the Total Conc of Strands entry field.

d. Enter the Molecularity (i.e. the number of strands) of the hybrid. For example, enter '2' in this field if the DNA molecule being analyzed consists of two strands.

e. Check the Delta G checkbox to perform Delta G (ΔG or free energy) and K (equilibrium constant) calculations.

f. In the Number of Temperatures field that is now accessible, specify the number of temperatures at which you would like to perform the Delta (Δ) G and K calculations. The number set here is reflected in the Temperature table below this field.

g. Enter the required temperature in each cell in the Temperature table and click OK.

**Hyperchromicity recalculation procedure**
Step 8

a. An Attention dialog will appear asking you to select the Associated DNA limits using the Ruler. Press the OK button on this dialog. The cursor changes to ruler mode.

b. Place your mouse on the Thermal curve at one end of the linear region of the Associated DNA part of the curve (prior to transition). Click and hold down the left mouse button, then drag the mouse along the curve and release the left mouse button at the other end of the linear region.

The software will automatically calculate a linear least squares line between the selected points and extrapolate to both ends of the melting curve.

c. An Associated DNA Limits dialog will appear displaying the upper and lower limits which you selected with the Thermal Ruler. Press the OK button to accept the calculated line.

d. An Attention dialog will then appear asking you to select the Deassociated DNA limits using the Ruler, press the OK button on this dialog. The cursor changes to ruler mode.

e. Place your mouse on the Thermal curve at one end of the linear region of the Deassociated DNA part of the curve (after the transition). Click and hold down the left mouse button, then drag the mouse along the curve and release the left mouse button at the other end of the linear region.

The software will automatically calculate a linear least squares line between the selected points and extrapolate to both ends of the melting curve.

f. A Dissociated DNA Limits dialog will appear displaying the upper and lower limits which you selected with the ruler. Press the OK button to accept the calculated line. If you are not happy with the linearity, reselect a linear region using the Retry button. The Linear Region Associated and Dissociated correction points are displayed in the Report area.

Note
The Associated and Dissociated DNA Limits dialogs allow you to re-select your limits if you are not satisfied with the linear least squares lines drawn in the graph box. To do this, click the Retry button on either dialog and select another region. If you wish to exit from the calculation mode, click Cancel. The ruler trace is removed from the graph.

Note
If the two extrapolated lines cross at any point, a warning will appear. Click Cancel and repeat steps (a) to (f) above and re-select the linear regions. Click Ignore to proceed with the calculation using the regions you have selected.

Using the lines calculated in the previous steps the software will now automatically create a second graph box and display the calculated Alpha curve.

g. An Attention dialog will appear asking you to select the range on the Alpha curve which will be used for the Tm calculation. Press the OK button to make your selection.

h. Place your mouse on the alpha curve at one end of the linear region. Click and hold down the left mouse button, then drag the mouse along the curve and release the left mouse button at the other end of the linear region.
i. An Alpha Tm Limits dialog box will appear displaying the selected limits. Click the Retry button to re-select the limits if required. Press the OK button to accept the selected ranges.

A linear least squares line is drawn between the selected points and the value of temperature where \( a = 0.5 \) is calculated to give the Tm value. (The Tm value is automatically reported in °C and K.)

If you have chosen the van't Hoff Calculation method to calculate Delta G and K:

j. Using the data calculated in step (i) above, the system automatically creates a third graph box displaying a van't Hoff plot. An Attention dialog will appear asking you to select the upper and lower limits on the van't Hoff plot to calculate Delta G and K. Press the OK button on this dialog.

k. Place your mouse on the van't Hoff curve at one end of the linear region. Click and hold down the left mouse button, then drag the mouse along the curve and release the left mouse button at the other end of the linear region.

l. A van't Hoff Limits dialog box will also appear displaying the upper and lower limits that you selected with the ruler. Press the OK button to accept the calculated line. Alternatively you can select the Retry button to re-select the limits using the ruler.

Note
If there is an error in the van't Hoff calculation you will be prompted to re-select the linear region.

A linear least squares line is calculated between these points and is extrapolated to both ends of the van't Hoff plot. The slope and intercept of the calculated line gives the enthalpy and entropy of the hybrid formation (\( \Delta H^\circ \) and \( \Delta S^\circ \) respectively).

The software will automatically send the results, using the calculated alpha curve, to the Report area including:

- K(Tm) experimental
- K(Tm) from first principles
- K(Tm) experimental/K(Tm) from first principles

See also:
Hyperchromicity ruler steps
How to use the ruler during Hyperchromicity calculations
How to perform Derivative calculations on previously stored data

Recalculate—Thermal

Use the pages accessible from the Recalculate option to perform re-calculations on data that has been analyzed. These re-calculations can be made using new Setup parameters. For example you may wish to change the order of a reaction. You can also use the Recalculate button to access Report options.

To access the Recalculate option, select the Recalculate button from the Thermal Application window.

When you select this option, the Setup dialog will appear, containing the following tabbed pages:
How to perform post run calculations on previously stored data—Thermal

- Retrieve the data
- Set up the recalculation
- Print the report

Retrieve the data

Step 1

From the **File** menu, select **Open** to display the Windows **Open** dialog.

Step 2

A list of previously stored Batch files will appear. Select the Batch file you require then select **Open**.

The system will automatically retrieve the collected data into the Graphics area. The method that was used to collect the data will also be retrieved.

**Note**

You can also retrieve data from Data (*.FDTM), Method (*.FMTM) and Spreadsheet ASCII (*.CSV) file types.

Set up the recalculation

Step 3

Select the **Recalculate** button to display the **Setup** dialog. The **Analyze** and **Reports** pages will be displayed to enable you to set up the recalculation parameters.

Step 4

Select the **Analyze** tab to access the **Analyze** page where you can set the analysis parameters you require.

For example, select the **Auto Calculate** option to automatically perform a rate calculation on collected data at the end of each run.

Step 5

a. Select the **Reports** tab to display the **Reports** page where you can specify your reporting requirements for this method.

b. Enter your name in the **Name** field.
c. Enter a comment relating to your experiment in the **Comment** field *(optional)*.

d. Set up your report style by selecting the appropriate check boxes in the **Options** group.

   For example, select the **Auto Print** check box to obtain a printout of your report automatically. Select the **Parameters** check box to include your experimental parameters in the report. Select the **Graph** check box to include a calibration graph in the generated report.

**Note**

If **Auto Print** is selected, then the system will send the report information to the specified printer as well as the Report area. However if **Auto Print** is not selected, the report will only be sent to the Report area and can be viewed by selecting the **Report** command from the **View** menu.

**Step 6**

Select **OK** to clear the **Setup** dialog and start the recalculations. The results will be displayed in the Report area.

**Print the report**

**Step 7**

a. From the **File** menu, select **Print**. The Windows **Print** dialog is displayed.

b. Set up your printing parameters and select **OK** to print your report.

**See also:**

*How to generate a report from previously collected data*

### How to generate a report from previously stored data—Thermal

**Step 1**

Select **Open**… from the **File** menu to display the Windows **Open** dialog.

**Step 2**

A list of previously stored files will appear. If this is not a list of batch files, select the down arrow at the right of the **Files of type** list box and select 'Batch' to list all the batch files.

**Step 3**

Select the Batch file you require then select **Open**.

The system will automatically retrieve the collected data into the Graphics area. The method that was used to collect the data will also be retrieved.

**Note**

You can also retrieve data from Methods (*.FMTM), Data (*.FDTM), Report (*.FRTM),
Spreadsheet ASCII (*.CSV), GRAMS (*.SPC) and Rich Text Format (*.RTF) file types.

Step 4

Select the **Recalculate** button to display the **Setup** dialog.

Step 5

Select the **Reports** tab and on the **Reports** page specify the information you would like to include in the report. Select **Auto Print** if you require the new report to print to the Reports area and to a printer.

Step 6

Click **OK** to display the results in the Report area (and send to the printer if **Auto Print** is selected).

**Note**

If you require a printout of only the current graphics in the Graphics area of a previously stored data file:

a. Select the **Clear Report** option in the Commands menu to clear any text that may be displayed in the Report area.

b. Press the **Print** button to obtain a printout of the graphics.

Alternatively tick only the **Graph** check box in the **Reports** page to obtain a printout of graphics only.

**See also:**

How to use advanced reporting options

**Maths window**

Use the **Maths** window to perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator.

To access the **Maths** window, select the **Calculator** command from the **Graph** menu, or select the **Calculator** button on the **Graphics** toolbar.

The **Maths** window is only available in the Kinetics, Lifetimes, Scan and Validate applications.

The **Maths** window comprises the following:

<table>
<thead>
<tr>
<th><strong>Calculation display field</strong></th>
<th>View the current calculation, including the trace/s being used, the trace operation being carried out and any functions that have been applied to the trace.</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Selected</strong></td>
<td>Choosing either of these options selects the highlighted (red) trace in the current Graph</td>
</tr>
</tbody>
</table>
**Trace/Focused** box for manipulation. When choosing between these options, use this simple rule:

- Use the **Selected Trace** option when the selected calculation is only to be applied to one trace.
- Use the **Focused Trace** option when the same calculation is to be applied to a number of traces.

The following example illustrates the difference between these options.

To perform the same manipulation on several traces:

a. Highlight a trace by clicking on it with the mouse. The trace will become red.
b. Select **Selected Trace, Apply** and then = to plot the result of the calculation
c. Highlight the next trace and repeat step b.

OR

a. Highlight the first trace.
b. Select **Focused Trace, Apply** and then = to plot the result of the calculation;
c. Highlight the next trace and select the = button to perform the same calculation on the newly selected trace.

**Trace**

Use the Trace group options to select the operation to be performed on the traces, the filter size and the interval (where applicable).

Click **Apply** to insert the selected operation into the equation

Click **=** (equals) to display the result of the calculations.

**Operation**

Click on an operation to find out more about it

- **Anisotropy**
- **Polarization**
- **Sum Function**
- **Mean**
- **Boxcar Averaging**
- **Moving Average**
- **Savitzky-Golay**
- **Spline**
- **Deriv 1**
- **Deriv 2**
- **Deriv 3**
- **Deriv 4**

**Display Options**

Select whether the results of the operation performed on the trace will be viewed in a New Graph box or in the Selected Graph box. If you choose the Selected Graph option, you will be able to directly compare the results with existing traces.

**Functions**

This group contains number and function buttons that can be used as a standard calculator in addition to being applied to traces.

To use the functions:
1. select a trace or value
2. select either a second trace or number
3. select the equals (=) button to display the result

All the basic mathematical functions are available (addition, subtraction, multiplication and division), as well as logarithm (Log) and square root (Sqrt) functions. When used as a calculator, the results of equations appear in the Report area.

**Additional buttons**

- **C**
  Click C to clear the entire operation in the calculation display field.

- **CE**
  Click CE to clear the last entry entered in the calculation display field.

- **=**
  Click = (equals) to start the operation shown in the calculation display field.

See also:
- Maths Home Page
- Maths How To

**Maths Operations**

The following operations can be selected on the Maths window. To access the Maths window, select the Calculator command from the Graph menu, or select the Calculator button on the Graphics toolbar.

**Anisotropy, Polarization and Sum Function**

The term Anisotropy refers to an unequal angular distribution about a point in space. Anisotropy also refers specifically to a simple expression for measuring the degree of polarization of the fluorescence. It is almost identical to the expression for polarization apart from having a factor of two in the denominator. Anisotropy is usually represented by the lowercase letter "r", while polarization by an uppercase "P".

The two expressions are listed below:

*Anisotropy*

\[ r = \frac{(I_{vv} - G I_{vh})}{(I_{vv} + 2G I_{vh})} \]

*Polarisation*

\[ P = \frac{(I_{vv} - G I_{vh})}{(I_{vv} + G I_{vh})} \]

where

- **I_{vv}** equals the intensity of fluorescence with excitation polarizer = vertical, emission polarizer = vertical
- **I_{vh}** equals the intensity of fluorescence with excitation polarizer = vertical, emission polarizer = horizontal

G is a factor that accounts for the polarization bias of the instrument. In particular it accounts for the wavelength-dependent polarization bias of the emission side of the instrument (dominated by the polarization bias of the emission grating).

The G factor is given by:
\[ G = \frac{I_{hv}}{I_{hh}} \]

where

\textbf{I}_{hv} \text{ equals the intensity of fluorescence with excitation polarizer } = \text{ horizontal, emission polarizer } = \text{ vertical} \]

\textbf{I}_{hh} \text{ equals the intensity of fluorescence with excitation polarizer } = \text{ horizontal, emission polarizer } = \text{ horizontal} \]

The expression for the anisotropy is a quotient. The denominator of the expression, as given by \( I_{vv} + 2GI_{vh} \) is the ‘sum’ function, and actually represents the total emission unbiased by any sample polarization effects.

**Mean**

The Mean operation allows the average of two or more traces to be taken and the resulting trace to be plotted. The mean of the traces can also be used in conjunction with other functions.

The equation used in mean calculations is:

\[
\frac{(scan 1 + scan 2 + \ldots + scan x)}{x}
\]

The calculation is performed over the common X range of the traces and the trace with the most dense data is used to determine the data points. The other traces are interpolated at the relevant data points.

**Boxcar Averaging**

This algorithm will take the number of points as defined by the Filter size, and average these, displaying the average at the middle X-point of the filter.

For example, for the following:

\[
\begin{align*}
\text{Data:} & \quad (x_1, y_1), (x_2, y_2), (x_3, y_3) \ldots \quad (x_n, y_n) \\
\text{Filter size:} & \quad 5 \\
\end{align*}
\]

The first smoothed data point will be \((x_3, \text{sum}(y_1:y_5)/5)\), the next \((x_8, \text{sum}(y_6:y_{10})/5)\) \ldots \((x_{n-2}, \text{sum}(y_{n-4}:y_{n})/5)\).

If there are not enough points at the end of the data set to complete the filter size, then these points will be ignored, hence, the gap at the end may be larger than the gap in the beginning. There will be a gap before the first point and after the last point of the smoothed continuum, with respect to the original unsmoothed data. This algorithm is only valid for evenly x-spaced data points.

Within a time scan, (e.g., Advanced collect in kinetics) the data points may be unevenly spaced between different stages. Each stage will be treated as a new collect, hence, there may be large gaps between stages. If there are not enough points at the end of a stage to complete the filter size, then these points are ignored, hence, there may be large gaps at the end of a stage. The filter size will be limited to odd numbers, with the minimum of 3 to a maximum of 101.

**Moving Average**

This algorithm will take the number of points as defined by the Filter size, and average these, displaying the average at the middle X-point of the filter.
For example, for the following:

Data: \((x_1, y_1), (x_2, y_2), (x_3, y_3) \ldots (x_n, y_n)\)

Filer size: \(5\)

The first smoothed data point will be \((x_3, \text{sum}(Y_1:y_5)/5)\), the next \((x_4, \text{sum}(y_2:y_6)/5)\)
\ldots \((x_{n-2}, \text{sum}(y_{n-4}:y_n)/5)\). There will be a gap before the first point and after the last point of the smoothed continuum, with respect to the original unsmoothed data. This algorithm is only valid for evenly x-spaced data points. Within a time scan, (e.g., Advanced collect in kinetics), the data points may be unevenly spaced between different stages. This will be treated by interpolation of data from the previous stage so as to give evenly spaced data points into the next stage. If there is not enough data for interpolation, then the next stage will be treated as a new collect, hence, there may be gaps and further delays in displaying the point. The filter size will be limited to odd numbers, with the minimum of 3 to a maximum of 101.

**Savitzky-Golay**

The Savitzky-Golay technique operation enables you to reduce the noise in the currently selected Maths scan by using smoothing.

When you select this operation you will be prompted to enter a Filter Size from 5 to 101. The larger the Filter Size, the more data points that will be smoothed.

Q. I cannot enter a particular value; it keeps changing to another number — why?
A. The Savitzky-Golay technique requires an odd number to be entered, not an even number.

You will also need to enter an Interval. The Interval refers to how many of the averaged points will be plotted on the smoothed trace. The smaller the interval, the more detail there will be in the trace.

Consider selecting an Interval of 1. This would mean that when the smoothed continuum is plotted, every smoothed point will be displayed.

Q. Why can't I see the Filter Size and Interval fields?
A. These will be displayed when you select Smooth in the Operation field.

**Spline**

The Deriv operations allow you to obtain derivatives of a selected scan. The derivative can be of the orders 1 to 4. The resulting trace is then plotted. The Savitzky-Golay technique is used for derivative calculations.

On selection of the Deriv operation you will be prompted to enter a Filter Size from 5 to 101. The Filter Size refers to the number of points around a particular point that will be averaged to produce a new derived Y value. The lower the Filter Size, the more data points that will be used in the derivative trace.

Q. I cannot enter a particular value; it keeps changing to another number — why?
A. The Savitzky-Golay technique requires an odd number to be entered, not an even number.
You will also need to enter an Interval. The Interval refers to how many of the derived points will be plotted on the resulting trace. The smaller the Interval, the more detail there will be in the trace.

Q. Why can't I see the Filter Size and Interval fields?
A. These will be displayed when you select one of the ‘Derivs’ in the Operation field.

See also: About smoothing

Maths How To…

- How to perform a spectral subtraction
- How to perform a mean calculation
- How to smooth a trace
- How to derive a trace

See also: Maths Home Page

User Data Form—Thermal

Use the User Data Form to enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data.

To access the User Data Form, select the User Data Form command from the Graph menu, or select the User Data Form button on the Graphics toolbar. The User Data Form command is only available if you have selected a graph box.

The dialog comprises the following:

- Data Name: This column lists the names of each visible trace in the highlighted graph box.

  Use the extra columns for recording additional information about the experimental conditions you used to collect the data, eg. pH.

- Other Columns: Use this to record the name of the parameter in which you are interested as a column heading, (for example, pH, ionic concentration, substrate concentration etc.).

  Note
  You must enter a column heading, or else the data in that column will not be saved.

See also: Graph menu, Graphics toolbar

Displaying data graphically—Thermal

- Graphics area
The Graphics area of the Thermal application appears in the top half of the Thermal Application window and is used to display traces of collected data. In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area surrounding the graph is referred to as the graph box. A graph will appear in the Graphics area when you select Start to commence a Thermal run.

To display or hide the Graphics area, select the Graphics command on the View menu. Double-clicking on the Graphics area enables you to toggle between half or full size display sizes.

You can access the functions associated with the Graphics area by doing the following:
- selecting one of the Graphics toolbar buttons;
- selecting a command from the Graph menu; or
- clicking the right mouse button in the Graphics area and select a command from the menu

**Note**
Not all functions are available on every menu.

You can customize the graphics area to suit your requirements in the following ways:

**Note**
All of the commands and dialogs listed are accessible from the Graph menu.

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<thead>
<tr>
<th>To…</th>
<th>Use the…</th>
<th>Tell me how…</th>
</tr>
</thead>
<tbody>
<tr>
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<td>Autoscale graph command</td>
<td>How to scale your graph automatically</td>
</tr>
<tr>
<td>Change the axes scaling manually</td>
<td>Axes Scales dialog</td>
<td>How to scale your graph manually</td>
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<tr>
<td>Modify the graph appearance (axes, trace)</td>
<td>Graph Preferences dialog</td>
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<td></td>
<td>How to specify the number of tick marks and decimal places for your X and Y axes</td>
</tr>
<tr>
<td>Add a new graph to the display</td>
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<td>View single or multiple graphs</td>
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<td>Resize a graph box</td>
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<td>Track the focused trace</td>
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<td>Action</td>
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<td>-------------------------</td>
<td>-----------------</td>
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<tr>
<td>Select the focused trace</td>
<td></td>
<td>select the</td>
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<td>Display a trace in the graph</td>
<td>Trace Preferences dialog</td>
<td>display a trace</td>
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<td>Remove a trace from a graph</td>
<td>Trace Preferences dialog</td>
<td>remove a trace</td>
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<tr>
<td>Remove a trace from the trace list</td>
<td>Trace Preferences dialog</td>
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<td>Zoom in and out on the graph</td>
<td>Zoom In and Zoom Out commands</td>
<td>zoom a graph</td>
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<tr>
<td>Add a label to the graph box</td>
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<tr>
<td>Edit labels in the graph box</td>
<td>Edit Label dialog</td>
<td>edit a label</td>
</tr>
<tr>
<td>Moving labels in the graph box</td>
<td>Cursor</td>
<td>move a label</td>
</tr>
<tr>
<td>Resizing labels in the graph box</td>
<td>Cursor</td>
<td>resize a label</td>
</tr>
<tr>
<td>Delete labels from the graph box</td>
<td>Delete Annotation command</td>
<td>delete a label</td>
</tr>
<tr>
<td>Add a picture to the graph box</td>
<td>Add Picture dialog</td>
<td>add a picture</td>
</tr>
<tr>
<td>Move pictures in the graph box</td>
<td>Cursor</td>
<td>move a picture</td>
</tr>
<tr>
<td>Resizing pictures in the graph box</td>
<td>Cursor</td>
<td>resize a picture</td>
</tr>
<tr>
<td>Replace pictures in the graph box</td>
<td>Edit Picture dialog</td>
<td>replace a</td>
</tr>
<tr>
<td>Delete pictures from the graph box</td>
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<td>---------------------------</td>
<td>------------------------</td>
</tr>
<tr>
<td>Save the Graphics area you have set up as a graphics template</td>
<td>Save As dialog (from the File menu)</td>
<td>How to create a graphics template</td>
</tr>
</tbody>
</table>

**Note**
The red trace in any graph box is the one on which all information appears and the one used for the cursor tracking. It is called the focused trace.

**See also:**
- Toolbar
- Graphics How To

# Graphics How to—Thermal

## Traces

- How to track the focused trace
- How to select the focused trace
- How to display a trace that is not appearing in a graph box
- How to remove a trace from a graph
- How to remove a trace from the trace List

## Sizing and scale

- How to resize graph boxes
- How to scale your graph manually
- How to scale your graph automatically
- How to alter the look of your graphs
- How to zoom a graph

## Labels

- How to add a label to a graph
- How to edit a label
- How to delete a label
- How to move a label

## Pictures

- How to add a picture to a graph
- How to move a picture
- How to delete a picture
- How to replace a picture
Graph menu—Thermal

Introduction

Use the Graph menu to view data in various graphical formats or in multiple graphs in the Graphics area. You can double-click on the Graphics area to toggle the display through graph and report to graph only.

In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area surrounding the graph is referred to as the graph box. A graph will appear in the Graphics area when you select Start to commence a Thermal run.

The Graph menu can be accessed in the following ways:

- from the main menu at the top of the Thermal Application window
- by clicking the right mouse button in a graph box or the Graphics area

Note

The red trace in any graph box is the one on which all information appears and the one used for the cursor tracking. It is called the focused trace.

Menu commands

The following commands can be accessed from the Graph menu:

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<thead>
<tr>
<th>Menu command</th>
<th>Button</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Print XY point</td>
<td><img src="image" alt="Print XY point" /></td>
<td>Select this to print the cursor's current X and Y coordinates in the report area.</td>
</tr>
<tr>
<td>Copy Graph</td>
<td><img src="image" alt="Copy Graph" /></td>
<td>Select <strong>Copy Graph</strong> to copy the entire Graphics area to the Windows Clipboard. All of the graphs in the Graphics area will be copied to the Clipboard.</td>
</tr>
<tr>
<td>Paste to Graph</td>
<td><img src="image" alt="Paste to Graph" /></td>
<td>Select <strong>Paste to Graph</strong> to paste the contents of the Windows Clipboard into the Graphics area. Text is pasted as a label, graphics are pasted as pictures.</td>
</tr>
<tr>
<td>Add Graph</td>
<td><img src="image" alt="Add Graph" /></td>
<td>Select <strong>Add Graph</strong> to add a new graph box to the Graphics area. You may want to do this if you have, for example, a graph containing existing data and you want to collect some new data in a new graph.</td>
</tr>
<tr>
<td>Remove Graph</td>
<td><img src="image" alt="Remove Graph" /></td>
<td>Select <strong>Remove Graph</strong> to delete the selected graph box from the Graphics area. The graph box is removed, but the data is not deleted and is still available from the Trace Preferences window. You may want to use remove graph if you have a large number of graph boxes open but only want to focus on some of these.</td>
</tr>
</tbody>
</table>

Note:

You can remove multiple graph boxes by first holding down the CTRL key and selecting the graphs to be removed and then selecting the **Remove Graph** button or menu item. Use the SHIFT key to highlight a range of
<table>
<thead>
<tr>
<th>Clear All Traces</th>
<th>Select <strong>Clear All Traces</strong> to delete all traces from the graph/s. Note that selecting this option simply turns off the <strong>Visible</strong> property for the traces in the <strong>Trace Preferences</strong> window. You can redisplay the traces by making the traces <strong>Visible</strong> again in the <strong>Trace Preferences</strong> window.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Add Label</td>
<td>Select <strong>Add Label</strong> to display the <strong>Add Label</strong> dialog where you can enter a text label to be placed on a graph or in the background of the Graphics area.</td>
</tr>
<tr>
<td>Add Picture</td>
<td>Select <strong>Add Picture</strong> to display the <strong>Add Picture</strong> dialog where you can select a picture, bitmap or chemical structure to be placed on a graph or in the background of the Graphics area.</td>
</tr>
</tbody>
</table>
| Edit Annotation  | Select **Edit Annotation** to alter the content or look of the selected annotation, (either a label or a picture) in the Graphics area.  
|                  | **Note:**  
|                  | This option can only be activated if an annotation is selected. |
| Delete Annotation| Select **Delete Annotation** to remove an annotation, (such as a label or a picture) from the Graphics area.  
|                  | You can use this menu option to remove highlighted annotations in the Graphics area such as labels, pictures and ruler/fit information.  
|                  | **Note:**  
|                  | This option can only be activated if an annotation is selected. |
| Trace Preferences| Select this command to display the **Trace Preferences** dialog where you can select the trace(s) to display in the selected graph. |
| Graph Preferences| Select this command to display the **Graph Preferences** dialog where you can change the appearance of your graph. Any changes you make to the appearance of the graph will be applied to all graphs. |
| Cursor Mode…     | Select this command to display the **Cursor Modes** dialog where you can choose the type of cursor to be used on the screen. For example, you can select to use a free cursor that can move anywhere in the graph or exactly trace the focused thermal (red thermal). Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the **Least Squares** method or the **Point to Point** method. |
| Axes Scales…     | Select this command to display the **Axes Scales** dialog where you can set the scale range for the X and Y axes. |
| Auto scale graph | To automatically scale the graph, point to one of the following options:  
|                  | **Autoscale[XY]**  
<p>|                  | Select Autoscale[XY] to automatically scale collected data by scaling the Y height and X width to full screen. |</p>
<table>
<thead>
<tr>
<th>Graphics Toolbar—Thermal</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Autoscale[X]</strong></td>
</tr>
<tr>
<td>Select Autoscale[X] to automatically scale collected data by scaling the X width to full screen.</td>
</tr>
<tr>
<td><strong>Autoscale[Y]</strong></td>
</tr>
<tr>
<td>Select Autoscale[Y] to automatically scale collected data by scaling the Y height to full screen.</td>
</tr>
<tr>
<td><strong>Zoom Out</strong></td>
</tr>
<tr>
<td>Select <strong>Zoom Out</strong> to Zoom back to the original X and Y ranges which were used to collect the data (i.e. the method setup conditions of the focused thermal). For example, the system will revert back to the X and Y ranges set in the method if you zoom in and out during a data collection.</td>
</tr>
<tr>
<td><strong>Single/Multi Graph</strong></td>
</tr>
</tbody>
</table>
| Select **Single/Multi Graph** to toggle between displaying one or all graphs.  
   **Note:**  
   The graph highlighted in the **Multi Graph** view will be displayed when you toggle the **Single Graph** option. |
| **Auto Arrange Graphs**  |
| Select **Auto Arrange Graphs** to view and arrange all graphs simultaneously.  
   **Note:**  
   You can move graph boxes manually by clicking the left mouse button outside the graph axes and dragging the graph box to a new position. You can also resize graphs. |
| **User Data Form**       |
| Select **User Data Form** to display the **User Data Form** where you can enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data, such as the pH at which data was collected. |
| **Maths**                |
| Select **Maths** to display the **Maths** window where you can perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator. |
| **History List**         |
| This displays a list of the current graphs. |

See also:
- Add Label
- Add Picture
- Edit Label
- Edit Picture
- Trace Preferences
- Graph Preferences
- Cursor Modes
- Axes Scales
- Single/Multi Graph
- User Data Form
- Maths
- Graphics toolbar
- Graphics area

**Graphics Toolbar—Thermal**

Use the Graphics toolbar buttons to gain quick access to many of the **Graph** menu options. You can also use the toolbar to select the graph you want to view and to change the look of your graphs.
To display or hide the toolbar, point to the Toolbars option in the View menu and select Graphics. To display or hide the toolbar's text labels, point to the Toolbars option in the View menu and select Text Labels.

**Hot Tip**
To see the name of a button, point to it with the mouse.

The following commands can be accessed from the Graphics toolbar:

<table>
<thead>
<tr>
<th>Button</th>
<th>Menu command</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>![Trace]</td>
<td>Trace Preferences</td>
<td>Click this button to display the Trace Preferences window where you can select the trace(s) to display in the selected graph.</td>
</tr>
<tr>
<td>![Graph]</td>
<td>Graph Preferences</td>
<td>Click this button to display the Graph Preferences dialog where you can change the appearance of your graph. Any changes you make to the appearance of the graph will be applied to all graphs.</td>
</tr>
<tr>
<td>![Cursor]</td>
<td>Cursor Mode</td>
<td>Click this button to display the Cursor Modes dialog where you can choose the type of cursor to be used on the screen. For example, you can select to use a free cursor that can move anywhere in the graph or exactly trace the focused scan (red scan). Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the Least Squares method or the Point to Point method.</td>
</tr>
<tr>
<td>![Axes]</td>
<td>Axes Scales…</td>
<td>Click this button to display the Axes Scales dialog where you can set the scale range for the X and Y axes.</td>
</tr>
<tr>
<td>![Autoscale]</td>
<td>Autoscale [XY]</td>
<td>Click the Autoscale[XY] button to automatically scale collected data by scaling the Y height and X width to full screen.</td>
</tr>
<tr>
<td>![Autoscale]</td>
<td>Autoscale [X]</td>
<td>Click the Autoscale[X] button to automatically scale collected data by scaling the X width to full screen.</td>
</tr>
<tr>
<td>![Autoscale]</td>
<td>Autoscale [Y]</td>
<td>Click the Autoscale[Y] button to automatically scale collected data by scaling the Y height to full screen.</td>
</tr>
<tr>
<td>![Zoom Out]</td>
<td>Zoom Out</td>
<td>Click the Zoom Out button to Zoom back to the original X and Y ranges which were used to collect the data (i.e. the method setup conditions of the focused scan). For example, the system will revert back to the X and Y ranges set in the method if you zoom in and out during a data collection.</td>
</tr>
<tr>
<td>![Add Label]</td>
<td>Add Label</td>
<td>Click this button to display the Add Label dialog where you can enter a text label to be placed on a graph or in the background of the Graphics area.</td>
</tr>
<tr>
<td>![Add Picture]</td>
<td>Add Picture</td>
<td>Click this button to display the Add Picture dialog where you can select a picture, bitmap or chemical structure to be placed on a graph or in the background of the Graphics area.</td>
</tr>
<tr>
<td>![Single/Multi Graph]</td>
<td>Single/Multi Graph</td>
<td>Select Single/Multi Graph to toggle between displaying one or all graphs. <strong>Note:</strong> The graph highlighted in the Multi Graph view will be displayed when you toggle the Single Graph option.</td>
</tr>
</tbody>
</table>
| **Auto Arrange Graphs** | Click the **Auto Arrange Graphs** button to view and arrange all graphs simultaneously.  
**Note:**  
You can move graph boxes manually by clicking the left mouse button outside the graph axes and dragging the graph box to a new position. You can also resize graphs. |
<table>
<thead>
<tr>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Add Graph</strong></td>
<td>Click this button to add a new graph box to the Graphics area. You may want to do this if you have, for example, a graph containing existing data and you want to collect some new data in a new graph.</td>
</tr>
</tbody>
</table>
| **Remove Graph** | Click this button to delete the selected graph box from the Graphics area. The graph box is removed, but the data is not deleted and is still available from the Trace Preferences window. You may want to use remove graph if you have a large number of graph boxes open but only want to focus on some of these.  
**Note:**  
You can remove multiple graph boxes by first holding down the CTRL key and selecting the graphs to be removed and then selecting the **Remove Graph** button or menu item. Use the SHIFT key to highlight a range of graphs for removal. |
| **Clear All Graphs** | Click this button to delete all traces from the graph/s. Note that selecting this option simply turns off the Visible property for the traces in the Trace Preferences window. You can redisplay the traces by making the traces Visible again in the Trace Preferences window. |
| **User Data Form** | Click this button to display the **User Data Form** where you can enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data, such as the pH at which data was collected. |
| **Calculator** | Click this button to display the **Maths** window where you can perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator. |

**See also:**  
Add Label  
Add Picture  
Edit Label  
Edit Picture  
Trace Preferences  
Graph Preferences  
Cursor Modes  
Axes Scales  
Single/Multi Graph  
User Data Form  
Maths  
Graphics area  
Graph menu  
Graphics How to
Graph Labels—Thermal

- Add Label dialog
- Edit Label dialog
- How to add a label to a graph
- How to edit a label
- How to move a label
- How to resize a label
- How to delete a label

Add Label—Thermal

Use the Add Label dialog to place a label on your graph or into the background of the Graphics area.

To add a label to a graph box, you must have at least one trace Visible in the graph box. (You can select which traces you want to make visible in the Trace Preferences dialog.) If you are viewing multiple graphs, the label will be placed in the graph box that is currently selected. However, you can add a label outside of a graph box in the background of the Graphics area at any time.

Q. Can I add a label that will print with all my graphs.
A. Yes, you can add your label to the background of the Graphics area. To do this, right-click on a section of the background (gray area) and select Add Label from the menu. (You will notice that this menu has less options than the menu that appears when you right-click on a graph.) In the Add Label dialog, enter the label to be added to all graphs and select OK. The label will appear in the background of the Graphics area.

To access the Add Label dialog, select the Add Label command from the Graph menu, or select the Add Label button on the Graphics toolbar.

The Add Label dialog comprises the following:

- To selected trace
  - Select this option to associate a label with the selected trace. When you associate a label with a trace and make the trace visible in the Graph box, the label will also be displayed with the trace.

- To selected graph
  - Select this option to associate the label with the selected graph. When you associate a label with a graph, the label will always be displayed in the Graph box, regardless of what traces are visible.

- Selected trace
  - This will identify the trace that is currently selected and display the name of the trace.

- Font...
  - Click Font to open the Windows Font dialog. In this dialog you can specify the font type, style and size to be used for the label.

- Label
  - Use the Label field to enter the text for the label. You can enter as many lines of text as you require.

- Hot Tip
  - If you wish to include certain XY co-ordinates on the graph as a label, position your cursor at the appropriate place, right-click and select Add Label from the menu. When the Add Label dialog appears, the co-ordinate positions will be added to the Label field.

- Note
When you save the data (as a Batch or Data file) the label is saved with its associated trace or graph. Labels outside graph boxes can be saved in a graphics template. To do this, choose **Graph Template** as the **Save As** type in the **Save As** dialog. See [Example of a Graphics Template](#).

See also:
- Trace Preferences dialog
- Graph menu
- Graphics toolbar
- How to add a label to a graph
- How to move a label
- How to edit a label
- How to resize a label
- How to delete a label

## Edit Label—Thermal

Use the **Edit Label** dialog to alter the content or look of your label.

To access the **Edit Label** dialog, right-click on the label you wish to change and select **Edit Annotation** from the menu.

The dialog comprises the following:

- **Font**
  - Select **Font** to open the **Font** dialog, where you can specify the font type, style and size to be used for the label.

- **Label**
  - Use the **Label** field to alter the text for the label.

See also:
- How to edit a label
- How to add a label to a graph
- How to move a label

## How to add a label to a graph—Thermal

You can add a label to your graph or Graphics area background. To do this:

**Step 1**

Click on the graph box you require or click outside the graph box(es) to choose the Graphics area background.

**Note**

There must be at least one trace Visible in the graph box before you can add a label to it.

**Step 2**

Click the **Add Label** button on the toolbar or right-click on the graph and select **Add Label** from the menu. The **Add Label** dialog will appear. The **Add Label** command is also accessible from the **Graph** menu.
Step 3

Click the **Font** button. The Windows **Font** dialog appears. Select the font type, size and style you require for your label. A sample of the font style appears in the **Sample** field. When you are satisfied with your selections, select **OK** to return to the **Add Label** dialog.

Step 4

Enter the label in the **Label** field and select **OK**. The label will be displayed on the selected graph. If necessary, you can move a label so that it is more appropriately placed on your graph.

How to edit a label—Thermal

Step 1

Place the cursor on the label and click the left mouse button to select the label.

**Hot Tip**

If you are having trouble selecting the label it may be that a trace is interfering with the label selection. Try zooming in and clicking in a different part of the label.

Step 2

Click the right mouse button and select **Edit Annotation** from the drop-down menu. Alternatively you can select the **Edit Annotation** command from the **Graph** menu. The **Edit Label** dialog is displayed.

Step 3

Use the **Edit Label** dialog to change the label.

For example, if you want to change the font size of the label, select the **Font** button. In the **Size** field, scroll through the available sizes for the font and select the size you require. Alternatively, highlight the currently selected size in the **Size** entry field and enter the size you require. Click the **OK** button in this dialog to return to the **Edit Label** dialog.

Step 4

Click **OK** to clear the **Edit Label** dialog. Your label will appear with the new font size.

See also:

- How to add a label to a graph
- How to move a label
- How to resize a label
- How to delete a label
How to move a label—Thermal

To move a label so that it is more appropriately placed on your graph or Graphics area background, do the following:

Step 1

Place the cursor on the label.

Step 2

Click and hold down the left mouse button.

**Hot Tip**

If you are having trouble selecting the label, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 3

Drag the label to the correct position and release the mouse button. The label will be displayed at the new position.

**Note**

Labels are transparent so as not to interfere with any traces on graphs.

See also:

- How to add a label to a graph
- How to edit a label
- How to delete a label
- How to resize a label

How to resize a label—Thermal

To resize a label so that it is more appropriately sized on your graph or Graphics area background, do the following:

Step 1

Select the label by clicking on it with the mouse. A border will be displayed around the label to indicate it is selected.

**Hot Tip**

If you are having trouble selecting the label, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Position the cursor over one of the corner edges of the label until the cursor becomes a diagonal double-headed arrow. Click and hold down the left mouse button.
Example of label showing the border that indicates it is selected and the arrow to be used for re-sizing

Step 3

Increase or decrease the size of the label by dragging the mouse. The label text will also be resized. Release the mouse button when the label and text are the correct size.

Note
Labels are transparent so as not to interfere with any traces on graphs.

See also:
- How to add a label to a graph
- How to edit a label
- How to delete a label

How to delete a label—Thermal

To delete a label from a graph or Graphics area background, do the following:

Step 1

Position the cursor on the label and click the left mouse button to select it.

Hot Tip
If you are having trouble selecting the label it may be that a trace is interfering with the label selection. Try zooming in and clicking in a different part of the label.

Step 2

Click with the right mouse button and select Delete Annotation from the drop-down menu displayed. Alternatively, you can select the Delete Annotation command from the Graph menu. The label will be deleted from the graph or Graphics area.

See also:
- How to add a label to a graph
- How to move a label
- How to edit a label
- How to resize a label

Graph Pictures—Thermal

- Add Picture dialog
- Edit Picture dialog
- How to add a picture
- How to replace a picture
- How to move a picture
- How to resize a picture
How to delete a picture

Add Picture—Thermal

Use the Add Picture dialog to insert a picture or bitmap into the Graphics area. The picture can be added as a background picture or to a particular graph box.

To add a picture to a graph box you must have at least one trace Visible in the graph box. (You can select which traces you want to make visible in the Trace Preferences dialog.) If you are viewing multiple graphs, the picture will be placed in the graph box that is currently selected. However, you can add a picture outside of a graph box in the Graphics area at any time.

Q. Can I add a picture that will print with all my graphs.

A. Yes, you can add your picture to the background of the Graphics area. To do this, right-click on a section of the background and select Add Picture from the menu. (You will notice that this menu has less options than the menu that appears when you right-click on a graph.) In the Add Picture dialog, load the picture to be added to all graphs and select OK. The picture will appear in the background of the Graphics area.

To access the Add Picture dialog, select the Add Picture command from the Graph menu, or select the Add Picture button on the Graphics toolbar.

The Add Picture dialog comprises the following commands:

- **To selected trace** Select this option to associate a picture with the selected trace. When you associate a picture with a trace and make the trace visible in the Graph box, the label will also be displayed with the trace.

- **To selected graph** Select this option to associate the picture with the selected graph. When you associate a picture with a graph, the label will always be displayed in the Graph box, regardless of what traces are visible.

- **Load** Click Load to open the Windows Open dialog. Use this dialog to browse your computer for the picture you want to insert. The picture must be one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.

  If you are viewing multiple graphs the picture will be placed on the graph that is currently selected. If no graph is selected, the picture will be placed on the Graphics area background.

**Note**
When you save the data (as a Batch or Data file) the label is saved with its associated trace or graph. Labels outside graph boxes can be saved in a graphics template. To do this, choose Graph Template as the Save As type in the Save As dialog. See 'Example of a Graphics Template'.

See also:
- Trace Preferences
- Graph menu
- Graphics toolbar
- How to add a picture to a graph
- How to move a picture
Edit Picture—Thermal

Use the **Edit Picture** dialog to change a picture you have inserted into the Graphics area.

To access the **Edit Picture** dialog, right-click on the picture you wish to change and select **Edit Annotation** from the menu.

The **Edit Picture** dialog comprises the following commands:

<table>
<thead>
<tr>
<th>Command</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>To selected trace/graph</strong></td>
<td>Displays whether you originally selected to add the picture to a trace or a graph. This option is not editable.</td>
</tr>
<tr>
<td><strong>Load</strong></td>
<td>Click <strong>Load</strong> to open the Windows <strong>Open</strong> dialog. Use this dialog to browse your computer for the picture you want to insert. The selected picture will replace the current picture. The picture must be one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.</td>
</tr>
</tbody>
</table>

See also:
- How to replace a picture
- How to add a picture
- How to move a picture
- How to resize a picture
- How to delete a picture

### How to add a picture to a graph—Thermal

You can add a picture to your graph or the Graphics area background. To do this:

**Step 1**

Click on the graph box you require or click outside the graph box(es) to choose the Graphics area background.

**Note**

There must be at least one trace Visible in the graph box before you can add a picture to it.

**Step 2**

Click the **Add Picture** button on the toolbar or right-click on the graph and select **Add Picture** from the menu. The **Add Picture** dialog will appear. The **Add Picture** command is also accessible from the **Graph** menu.

**Step 3**

Click the **Load** button. The Windows **Open** dialog appears.

**Step 4**
Browse through your directories to find the picture you want to insert. All pictures must be in the bitmap format and will have the extension .BMP.

Step 5

Highlight the .BMP that you want to insert and select **Open**.

Step 6

The **Add Picture** dialog will appear showing a preview of the bitmap. If the bitmap is the correct one select the **OK** button. The bitmap will be placed onto the graph.

**Note**

You should take care not to add a bitmap that is too big as it will obscure part or all of your graph.

You can move a picture so that it is more appropriately placed on your graph.

**See also:**
- How to move a picture to a graph
- How to delete a picture
- How to replace a picture
- How to resize a picture

**How to replace a picture—Thermal**

To change a picture that you have added to a graph or Graphics area background do the following:

Step 1

Move the cursor to the picture and select the left mouse button to select it.

**Hot Tip**

If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Click the right mouse button and select **Edit Annotation** from the drop-down menu or select **Edit Annotation** from the **Graph** menu. The **Edit Picture** dialog is displayed.

Step 3

From the **Edit Picture** dialog, select the **Load** button. The Windows **Open** dialog is displayed.

Step 4

Browse through your directories to find the picture to replace the current bitmap. Picture files must be in one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.

Step 5
Highlight the file that you want to insert and select **Open** to return to the **Edit Picture** dialog.

**Step 6**

The **Edit Picture** dialog will display a preview of the bitmap. If the bitmap is the correct one select the **OK** button. The bitmap will be placed onto the graph. If you need to, move the picture so that it is more appropriately placed on your graph.

**Note**

Do not to add a bitmap that is too big as it will obscure part or all of your graph.

**See also:**
- How to add a picture to a graph
- How to move a picture
- How to delete a picture
- How to resize a picture

**How to move a picture—Thermal**

To move a picture so that it is more appropriately placed on your graph or Graphics area background, do the following:

**Step 1**

Place the cursor on the picture.

**Step 2**

Click and hold down the left mouse button.

**Hot Tip**

If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

**Step 3**

Drag the picture to the correct position and release the mouse button. The picture will be displayed at the new position.

**See also:**
- How to add a picture to a graph
- How to delete a picture
- How to replace a picture
- How to resize a picture

**How to resize a picture—Thermal**

To resize a picture so that it is more appropriately sized on your graph or Graphics area background, do the following:
Step 1

Select the picture by clicking on it with the mouse. A border will be displayed around the picture to indicate it is selected.

Hot Tip
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Position the cursor over one of the corner edges of the picture until the cursor becomes a diagonal double-headed arrow. Click and hold down the left mouse button.

Example of picture showing the border that indicates it is selected and the arrow to be used for re-sizing

Step 3

Increase or decrease the size of the picture by dragging the mouse. The picture will also be resized. Release the mouse button when the picture is the correct size.

See also:
- How to add a picture to a graph
- How to move a picture to a graph
- How to delete a picture
- How to replace a picture
- How to resize a picture

How to delete a picture—Thermal

To delete a picture from a graph or Graphics area background, do the following:

Step 1

Position the cursor on the picture and click the left mouse button to select it.

Hot Tip
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Click with the right mouse button and select Delete Annotation from the drop-down menu displayed.
Alternatively, you can select the **Delete Annotation** command from the **Graph** menu. The picture will be deleted from the graph or Graphics area.

See also:
- How to add a picture to a graph
- How to move a picture
- How to zoom a graph
- How to resize a picture

**Trace Preferences—Thermal**

*Trace Preferences table* | *Trace Audit Log*

A single graph box can display many traces. This window enables you to select the trace(s) to display (make **Visible**) in the selected graph box.

To access the **Trace Preferences** dialog, select the **Trace Preferences** command from the **Graph** menu, or select the **Trace Preferences** button on the Graphics toolbar.

The dialog comprises the following:

**Hot Tip**
The **Trace Preferences** menu is also available on clicking the right mouse button anywhere in the trace listing.

**Trace Preferences table**

<table>
<thead>
<tr>
<th>Visible</th>
<th>Use the <strong>Visible</strong> field to select which traces will appear on the selected graph. To select or clear the <strong>Visible</strong> field, simply click the left mouse button in this column. A tick will appear next to traces that are to be displayed in the selected graph.</th>
</tr>
</thead>
</table>

If you are in single graph view, all selected traces will appear on the one graph. If you are in **Multi Graph** view the traces will appear on the graph that is currently selected.

**Hot Tip**
To make all traces Visible, right-click on the table and select the **Select All** command from the menu. Then right-click on the table again and select the **Show Traces** command from the menu.

<table>
<thead>
<tr>
<th>Color</th>
<th>The <strong>Color</strong> field enables you to view the color of each trace that appears on the selected graph. To change the color of a trace, highlight it, click the right mouse button in the <strong>Trace Preferences</strong> table and select <strong>Change Trace Color</strong>. You can then select a color from the <strong>Color</strong> dialog that is displayed.</th>
</tr>
</thead>
</table>

The red trace in any graph box is the one on which all information appears. This trace, which is used for the cursor tracking, is called the focused trace and it is *always* red. When you select a trace in the **Color** column it will become the focused trace in the selected graph box.

**Note**
When you change the trace line width in the **Graph Preferences** dialog it also will change the width of the color trace line in this dialog.

<table>
<thead>
<tr>
<th>Name of trace</th>
<th>The <strong>Name of Trace</strong> field contains a description of the traces currently available for display in the selected graph box.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Filename</td>
<td>This field displays the filename and path for the trace. If the name is too long to appear in the field you can use the keyboard arrow keys to scroll to the end of the filename or you can resize the <strong>Trace Preferences</strong> window. If the file path for a trace shows 'not saved' then the trace is unsaved data and is only stored in memory.</td>
</tr>
</tbody>
</table>

**Trace Audit Log**

The **Trace Audit Log** displays information about the highlighted trace. This includes instrument parameters and data form information relating to the trace that has been selected. It also logs all information about data manipulation. When more than one trace is highlighted, the **Trace Audit Log** will display information about the trace that was first selected.

**Trace Preferences menu—Thermal**

The **Trace Preferences** menu is displayed when you click the right mouse anywhere on the **Trace Preferences table**. The menu contains the following commands:

| Select All | This selects or highlights all the traces in the listed on the **Trace Preferences** page. |
| Select None| This de-selects all the traces in the listing. |
| Show Traces| Select **Show Traces** to make all of the traces currently highlighted **Visible**, meaning that they will be displayed in the current graph box. |
| Hide Traces| Select **Hide Traces** to remove the **Visible** property from all the traces currently highlighted, which will remove them from the current graph box. |
| Delete Traces| Select **Delete Traces** to delete all the highlighted traces in all graphs. Note that if you save the data file before you select **Delete Traces**, the traces are not deleted from the saved file, and they can be retrieved at a later date. Selecting **Delete Traces** will delete the traces from the application memory, and it is recommended that you select this option if you need to free up memory on your computer. This is especially relevant for the Thermal application where the data files can be quite large. |

**Note**

If the application is controlled by the GLP Administration application, you will be prompted to save an unsaved data file before it is removed from the application.

| Change | Select **Change Trace Color** to display the Windows **Color** dialog where you can |
**Trace Color** change the color of the currently highlighted trace(s). The focused trace will remain red while it retains the focus, and will revert to the selected color when the focus is moved.

See also:
- Trace Preferences page
- Graph Preferences page
- How to remove a trace from the trace list
- How to remove a trace from a graph

### How to select the focused trace—Thermal

To select the thermal that you want to be the focused (red) trace, do one the following:

- From the Graph, click the left mouse button on the trace that you want to make the focused trace.
- From the Graph Legend (if displayed), click the left mouse button on the trace that you want to make the focused trace.
- From the Trace Preferences dialog, click in the Color column of the trace that you want to make the focused trace.

**Note**
Select the Show Legend check box in the Graph Preferences dialog to display the Graph Legend.

See also:
- How to track the focused trace
- How to view a particular trace

### How to display a trace that is not appearing in the graph box—Thermal

**Step 1**

Click on the graph in the Graphics area.

**Step 2**

Select the Trace Preferences button from the Graphics toolbar, or select Trace Preferences from the Graph menu. The Trace Preferences dialog is displayed.

**Step 3**

Find that trace that you wish to see and select the Visible option. A tick will appear in the Visible column to indicate that it will now be displayed.

If you wish to make multiple traces visible, do the following:

- Hold down the CTRL key select all the traces that you want to display.
- Click the right mouse button and select the Show Traces command from the drop-down menu.
Ticks will appear next to the selected traces to indicate that they will now be displayed.

Step 4

Press **OK** to clear the **Trace Preferences** dialog. The selected trace(s) will be displayed.

See also:
- How to remove a trace from a graph
- How to remove a trace from the trace list

How to remove a trace from a graph—Thermal

Step 1

Select a graph in the Graphics area.

Step 2

Click the **Trace Preferences** button on the Graphics toolbar or select **Trace Preferences** from the **Graph** menu. The **Trace Preferences** dialog is displayed.

Step 3

Find that trace that you no longer wish to see and select the **Visible** option. The tick will be removed from the **Visible** column to indicate that it will not be displayed.

If you wish to make multiple traces visible, do the following:

a. Hold down the CTRL key select all the traces that you no longer want to display.

b. Click the right mouse button and select the **Hide Traces** command from the drop-down menu. The ticks will be removed from the selected traces to indicate that they will not be displayed.

Step 4

Select **OK** to clear the **Trace Preferences** dialog. The trace will be removed from the graph.

See also:
- How to display a trace that is not appearing in the graph box
- How to remove a trace from the trace list

How to remove a trace from the trace list—Thermal

If your graphs contain unwanted traces, you may wish to delete them (rather than simply making them invisible) in order to free up application memory and aid system performance.

Step 1

Select a graph in the Graphics area.
Step 2

Click the **Trace Preferences** button on the Graphics toolbar or select **Trace Preferences** from the **Graph** menu. The **Trace Preferences** dialog is displayed.

Step 3

Select the trace to be removed by clicking on it.

Step 4

Click the right mouse button to display the **Trace Preferences** menu.

Step 5

Select **Delete Traces** to delete the sample from the trace list. You will be prompted to confirm this action.

Step 6

Select **OK** to clear the **Trace Preferences** dialog. The trace will be removed from all graphs.

See also:

- **How to remove a trace from a graph**
- **How to display a trace that is not appearing in the graph box**

**Graph Preferences—Thermal**

**Introduction**

Use the **Graph Preferences** dialog to change the appearance of your graph.

All graphs will appear labeled with the X axis label being horizontal and the Y axis vertical. You cannot change the orientation of these labels. However you can alter the font and size of the axes and their labels.

Any changes you make to the appearance of the graph will apply to all graphs until the settings are changed again.

To access the **Graph Preferences** dialog, select the **Graph Preferences** command from the **Graph** menu, or select the **Graph Preferences** button on the Graphics toolbar.

**Dialog controls**

The **Graph Preferences** dialog comprises the following:

**Axes Preferences**
Axes Color  Select the color that you want to assign to the axes of the graph/s from the drop down list.

Axes Width  Select the width of the axes from the drop down list. The width, which is in pixels, ranges between one and five pixels.

Grid Style  Select the style of grid for the graph from the drop down list. If you do not wish to include a grid on the graph, select None.

Plot Area Color  Select the color to be used as the background of the graph from the drop down list. If you do not want a background color on the graph, select White.

Trace Width  Select the width of the trace from the drop down list. The width is given in pixels and ranges between one and five pixels.

Font  Click the Font button to display the Windows Font dialog where you can define the font style for the axes of the graph/s. The style and size of the font will be applied to all graph. The style, name and point size of the font appears on the font button.

Preview Box  Preview any changes you make to the axes preferences in the preview box. The changes shown in this box will not be made until you select OK.

Advanced Options

Auto tick marks  The Auto tick marks option enables you to approximately set how many tick marks (or points) will be included on the graph axes. The tick marks generated will be whole, rounded, even numbers. The Auto tick marks option is not available if you have selected Major ticks.

Y-axis  Use the Y-axis slider bar to increase or decrease the number of tick marks (or points) to be included on the Y axis.

X-axis  Use the X-axis slider bar to increase or decrease the number of tick marks (or points) to be included on the X axis.

Major ticks  Use the Major Ticks option to enable you to set exactly how many ticks (or points) will be added to the X and Y axes. The number of marks you specify will be divided by the axis range, which can result in odd numbers on the axis. Note that any changes are not reflected in the graph bitmap at the top of the screen.

Decimals  Use the Decimals option to set the number of X and Y decimal places to be displayed on the axes. To set the number of decimal places, select the Decimal option and enter the number in the Y-axis and X-axis combo boxes. Note that any changes are not reflected in the graph bitmap at the top of the screen.

Trace Style

Points  Select Points to display each individual data point collected by the Cary Eclipse on the graph.
How to alter the look of your graphs—Thermal

Step 1

Click the **Graph Preferences** button on the Graphics toolbar, or select **Graph Preferences** from the **Graph** menu. The **Graph Preferences** dialog is displayed.

Step 2

Use the available options available to choose your preferred appearance for the graphs. A preview box in the top right-hand corner will demonstrate how the graphs will appear.

Step 3

Select OK to clear the **Graph Preferences** dialog and update all graphs to reflect your selections.

**Note**

You can also add labels and pictures to your graphs to alter their appearance.
To re-use your graphics setup at a later date, save the setup as a graphics template.

See also:
How to add a label to a graph
How to add a picture to a graph
How to move a label
How to move a picture
How to resize graph boxes
How to make a graphics template

Cursor Modes—Thermal

Use the **Cursor Modes** dialog to choose the type of cursor to be used on screen. For example, you can select to use a free cursor that can move anywhere in the graph.

To access the **Cursor Modes** dialog, select the **Cursor Modes** command from the **Graph menu**, or select the **Cursor Modes** button on the **Graphics toolbar**.

The dialog comprises the following:

**Free**
The **Free** cursor is not confined to a particular trace on the graph. It can move anywhere in a graph box, and its ‘X,Y’ position is shown in the bottom right-hand side of the Graphics area. You can use the **Free** cursor to zoom in on a particular part of a graph.

*Note*
The Free cursor moves from point to point in the selected graph box. Hence you need to be careful when positioning the cursor over a thermal to obtain an accurate (X,Y) reading. It is recommended that you use the Track cursor mode for this.

**Track**
Use the **Track** cursor to exactly trace the focused (red) thermal in a graph box. The cursor position is displayed in the X and Y at the bottom right-hand side of the Graphics area.

*Hot Tip*
If you wish to fix the cursor on a particular position on the curve, move the cursor to the required position. Click the left mouse button, press ALT on the keyboard, then release the mouse button.

**Track Intensity**
Select the **Track Intensity** option if you want to associate an intensity value with the tracking. The cursor will then track from point to point on the continuum between the intensity value you enter. The default value will be 50% of the maximum intensity of the focused continuum.

**Peaks type**
Choose from the options of **Peaks**, **Valleys** or **Peaks Valleys** to move the cursor from peak to peak, valley to valley or from one peak/valley to the next.

See also:
How to zoom a graph
How to track the focused trace

**How to track the focused trace—Thermal**
To track the focused (red) trace in the graph:

Step 1

Click the **Cursor Modes** button from the Graphics toolbar, or select **Cursor Mode** from the **Graph** menu. The **Cursor Modes** dialog will appear.

Step 2

Select **Track** and select the **OK** button.

Step 3

Place the blue tracking cursor in the graph box you require.

Step 4

Move the cursor left and right across the graph box to trace the focused (red) thermal. The cursor coordinates change (in the X, Y field at the bottom left hand corner of the Graphics area) as you move along the curve.

**Hot Tip**

If you wish to fix the cursor on a particular position on the curve, move the cursor to the required position. Press 'Alt' on the keyboard, then release the mouse button.

**See also:**

- How to select the focused trace
- How to view a particular trace

**Axes Scales—Thermal**

Use the **Axes Scales** dialog to manually change the axes range. This can assist in zooming in on a particular part of the graph.

To access the **Axes Scales** dialog, select the **Axes Scales** command from the **Graph** menu, or select the **Axes Scales** button on the Graphics toolbar.

The dialog comprises the following:

**X Axis**

**Mode**

View the current units for the X axis.

**Scale**

Use the **Scale** field to enter a new range for the X axis, in the format n1 - n2 (where n1 is the start of the scale and n2 is the end of the scale).

**Hot Tip**

The Cary Eclipse will store the ten most recently entered ranges, which you can access by selecting the down arrow adjacent to the **Scale** field.
Y Axis

**Mode**
View the current units for the X axis.

**Scale**
Use the Scale field to enter a new range for the Y axis, in the format n1 - n2 (where n1 is the start of the scale and n2 is the end of the scale).

![Hot Tip]
The Cary Eclipse will store the ten most recently entered ranges, which you can access by selecting the down arrow adjacent to the Scale field.

See also:
- Graph menu
- Graphics toolbar
- How to scale your graph manually
- How to scale your graph automatically
- How to zoom a graph

**How to scale your graph manually—Thermal**

Step 1
Click on the graph whose axes you want to change to make it the focused trace.

Step 2
Click the Axes Scales button on the toolbar, or select Axes Scales from the Graph menu. The Axes Scales dialog is displayed.

Step 3
Type in the new ranges for the X and Y axes in the format ‘n-n’, or choose an option for each from the drop-down list.

Step 4
Select OK. The axes on the selected graph will change to reflect your chosen settings.

![Hot Tip]
Select the Zoom Out command to resize the graph to its original settings.

See also:
- How to zoom a graph
- How to scale a graph automatically

**How to scale your graph automatically—Thermal**

Step 1
Click on the graph whose axes you want to change to make it the focused trace.

Step 2
To autoscale the graph, select one of the following:

- Click the **Autoscale XY** button on the toolbar to autoscale both X and Y axes.
- Click the **Autoscale X** button on the toolbar to autoscale the X axis only.
- Click the **Autoscale Y** button on the toolbar to autoscale the Y axis only.

The focused trace will be scaled according to the option selected.

**Hot Tip**
Select the **Zoom Out** command to resize the graph to its original settings.

See also:
- How to zoom a graph
- How to scale your graph manually

**Single/Multi Graph—Thermal**

You can use the **Single/Multi Graph** command to toggle between a display of either one or multiple graph boxes in the Graphics area.

The command is accessible from the **Graph** menu or by selecting the **Single/Multi Graph** button from the Graphics toolbar.

Example of a single graph box display
Example of a multi graph box display

**Note**
Up to 100 graphs can be displayed in the Graphics area at the one time.

**See also:**
- Graph menu
- Graphics toolbar
- Graphics How To

### How to make a graphics template—Thermal

A graphics template lets you set up and store the style of a graphics report for future use. You can do this by adding labels and pictures to the background of the Graphics area and altering any axes styles etc. One advantage of this over the application's built-in reporting features is that you have more flexibility as to where you can position graph boxes and any text that you may want to appear (e.g. operator name, batch number etc.) and also where you add pictures, e.g. company logos etc.

**Step 1**

Start the application and open the data file from which you want to make a graphics template. Alternatively, collect some new data.

**Step 2**

Make as many graph boxes as you require in your template by using the **Add Graph** function.

**Step 3**

Adjust the graph boxes to the correct size and position them as you want them to appear in the template.

**Step 4**

Do any of the following, if required:

- Change the appearance of the graph boxes in the **Graph Preferences** dialog
- Use the **Add Label** function to append text to the Graphics area
- Use the **Add Picture** function to add pictures, such as your company's logo to the Graphics area

**Step 5**

From the **File** menu, select **Print Preview**. Use the **Print Preview** dialog displayed to check the template that you have created.

**Step 6**

When you are satisfied with the appearance of the graphics, select the **Save As**... command from the **File** menu. The **Save As** dialog is displayed.

**Step 7**

Note
Up to 100 graphs can be displayed in the Graphics area at the one time.
A list of stored data files will appear. Select the down arrow at the right of the **Files of type** field and select 'Graph Template' to list all the template files. Ensure that the **Save only focused trace** check box is cleared.

**Step 8**

Enter a name for the template in the **File name** field.

**Step 9**

Select **OK** to save the graphic template. The template will be used each time you create a new file.

**Note**

You can also make a graphic template in a similar manner from existing data. No data will be saved in the template.

**See also:**
- Example of a Graphics Template
- How to add a label to a graph
- How to add a picture to a graph
- How to move a label
- How to move a picture

**Example of a Graphics Template—Thermal**

By using the **Add Label** command in the Cary Eclipse graphics system, you can customize a graphics template such as the one shown below:

The above is created as follows:
Examples—Thermal

- **User Collect**
- **Multi-stage thermal run**
- **Example of a graphics template**

Example of a multi-stage thermal run—Thermal

Number of Stages = 3

The **Ave Time** is set to 1.0 s

*Graphical representation of data points for the above example*

**Stage 1:** 10°C - 30°C. The run will commence taking data readings at the set Ave Time of 1.0 s every 0.5°C. Once the stage one end temperature of 30°C is achieved it will hold this temperature for 5.0 minutes before proceeding to stage two.

**Stage 2:** 30°C - 65°C. The run will continue taking data readings at the set Ave Time of 1.0 s every 0.1°C. Once the stage two end temperature of 65°C is achieved it will hold this temperature for 2.0 minutes before proceeding to stage three.

**Stage 3:** 65°C - 85°C. The run will continue taking data readings at the set Ave Time of 1.0 s every 0.5°C. Once the stage three end temperature of 85°C is achieved it will hold this temperature for 2.0 minutes before proceeding to the return to temperature of 20°C.
See also:
Example of a User Collect

File Management—Thermal

- **File menu**
- **Open dialog**
- **Save As dialog**
- **Filename extensions**
- **ASCII spreadsheet file**
- **Auto Store page**
- **How to perform post run calculations on previously stored data**
- **How to combine data files into a batch file**
- **How to export collected data**
- **How to combine data files into a batch file**

**File menu—Thermal**

Use the **File** menu to open or save a method or data file, print or preview a report, or exit the Cary Eclipse application.

**Hot Tip**
Highlight a method or data file in Explorer and simply drag and drop it onto the application to open it. Any data will be displayed in the Graphics area.

The following commands can be accessed from the **File** menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>New</strong></td>
<td>Select <strong>New</strong> to clear all method, report and graphics information and create a new file containing the default settings.</td>
</tr>
<tr>
<td><strong>Open Method…</strong></td>
<td>Select <strong>Open Method</strong> to display the <strong>Open</strong> dialog where you can search for and load a stored method.</td>
</tr>
<tr>
<td><strong>Open…</strong></td>
<td>Select <strong>Open</strong> to display the <strong>Open</strong> dialog where you can search for and load method, data, report, graph template and batch files, as well as other file types compatible with the Cary Eclipse software. When you load a batch file, the method which collected the data is loaded as well as the associated report and graphics layout.</td>
</tr>
<tr>
<td><strong>Save Method As…</strong></td>
<td>Select this option to display the <strong>Save As</strong> dialog and store the current method. You can use the Save As dialog to store the method, data, report, or graphics template separately or together as a Batch file. The data can also be saved as a comma delimited ASCII Spreadsheet file.</td>
</tr>
<tr>
<td><strong>Save As…</strong></td>
<td>Select this option to display the <strong>Save As</strong> dialog and store the collected data. You can use the Save As dialog to store the method, data, report, or graphics template separately or together as a Batch file. The data can also be saved as a comma delimited ASCII file.</td>
</tr>
<tr>
<td><strong>Print Setup…</strong></td>
<td>Select this option to display the Windows <strong>Print Setup</strong> dialog where you can select the printer, printer page size and orientation.</td>
</tr>
<tr>
<td><strong>Print</strong></td>
<td>Select this option to display the <strong>Print Preview</strong> dialog where you can view the current</td>
</tr>
</tbody>
</table>
Look in

The Look In field lists the available folders and files. Click the down arrow on the right of this field to view the location of the current folder in your computer's hierarchy. You can also select a new location simply by selecting a different folder. The folders and files in the selected folder are displayed in the list box below the Look In field.

Note
To open the folder in the level that is one higher than the current folder, select on the toolbar.

Look in list box
The Look In List Box, which is directly below the Look In field, displays the folders and files in the selected location. To view the contents of a folder, double-click it.

Click to create a new folder. Type the name of the new folder in the highlighted text entry field that appears and press Enter. Then double-click on the new folder to open it.

Click to display the directory structure of the drive.

Click to display files as a list.

Click to display files as a detailed list including the file size, file type and the date the file was last modified.

Click to preview the selected trace before you load it into the application.

File name
Enter the name of the file you want to open in this field. The easiest way to do this is to find the file in the Look in List box and then select the file. However, you can type the
full path of a file and then select Open to open the file.

**Files of type**  
Click on the arrow on the right of this field to view a list of the different types of application files that can be opened in the list box.

**Methods (*.FMTM)**  
Choose this file type to open a Method file. Only the method is loaded. No data is loaded.

**Data (*.FDTM)**  
Choose this file type to open a Data file. Only the data is loaded. The method that was used to collect the data is not loaded. The last used method or default method will be displayed.

**Report (*.FRTM)**  
Choose this file type to open a Report file. You can view the report in the Report area.

**Graph Template (*.FGTM)**  
Choose this file to open a Graph Template file. The file will set up the layout of the Graphics area including any bitmaps as well as the range and style of the graph axes.

**Batch (*.FBTM)**  
Choose this file type to open a Batch file. A Batch file includes data, the method used to collect the data, report and graphics setup.

**Spreadsheet Ascii (*.CSV)**  
This will open ASCII files that contain single comma delimited files with two or more columns.

**GRAMS (*.SPC)**  
Choose this file type to open a Cary Eclipse GRAMS data file. This allows you to use data files collected using a Cary Eclipse GRAMS system. The method that was used to collect the data is not loaded.

**ADL (*.ADL)**  
This will open and run program files generated in the Applications Development Language (ADL).

**All files (*.*)**  
This enables you to view all files in the directory so that you can open any compatible file.

**Tell me more about Filename Extensions**

**Overlay Data**  
Select this check box to overlay individual data files in one graph box. If this is not checked the data from each file will appear in individual graph boxes. This option is only available if the Open dialog has been accessed by selecting 'Open…' from the File menu.

**Hot Tip**  
Use the SHIFT or Control key to select multiple data files.

**Open**  
This button opens the highlighted file/s or the files listed in the File name field.

---

See also:  
File menu  
ASCII Spreadsheet File

**Save As—Thermal**
Use the **Save As** dialog to save the data, method, report or graphics template individually or together as a Batch file.

**Note**
This dialog appears when you select either the **Save Method As** or **Save As** menu option in the **File** menu. When you select **Save Method As** this dialog appears with 'Methods' displayed in the **Files of type** field. When you select **Save As** this dialog appears with 'Batch' displayed in the **Files of type** field.

Traces (or data) that are currently ‘Visible’ in the Graphics area are saved, regardless of how they are displayed (full screen graph box or tile).

For example, if six graph boxes are displayed in the Graphics area and each graph box has one Visible trace then all six traces will be saved.

If you do not wish to save particular traces, make them not Visible in the **Trace Preferences** dialog. To save only the focused trace, select the **Save only focused trace** check box.

The **Save As** dialog comprises the following:

- **Look In**
  The **Look In** field lists the available folders and files. Click the down arrow on the right of this field to view the location of the current folder in your computer's hierarchy. You can also select a new location simply by selecting a different folder. The folders and files in the selected folder are displayed in the list box below the **Look In** field.

  **Note**
  To open the folder in the level that is one higher than the current folder, select on the toolbar.

- **Look in list box**
  The **Look In** list box, which is directly below the **Look In** field, displays the folders and files in the selected location. To view the contents of a folder, double-click it.

  - Click to create a new folder. Type the name of the new folder in the highlighted text entry field that appears and press ENTER. Then double-click on the new folder to open it.
  - Click to display the directory structure of the drive.
  - Click to display the files as a list.
  - Click to display files as a detailed list including the file size, file type and the date the file was last modified.
  - Click to preview the selected trace before you load it into the application.

- **File Name**
  Enter the name of the file you want to save in this field. You can also type the full path of a file then select the **Save** button to save the file.

- **Files of Type**
  Click on the arrow on the right of this field to view a list of the different types of application files that can be opened in the list box.

  **Methods (*.FMTM)** Choose this file type to only save the current method. Any
collected data is not stored.

**Data (*.FDTM)** Choose this file type to only save the data currently Visible in the Trace Preferences dialog (for any graph box). Note that the method used to collect the data is not stored with the data.

**Report (*.FRTM)** Choose this file type to only save the contents currently shown in the Report area.

**Graph Template (*.FGTM)** Choose this file type to save the layout of the Graphics area as a template. For example, any bitmaps you have added to your Graphics area will be saved as well as the range and style of the graph axes.

**Batch (*.FBTM)** Choose this file type to save the data that is currently Visible in the Trace Preferences dialog (for any graph box), the method used to collect the data and the currently displayed report all together as a batch file. The graphics setup is also stored.

**Spreadsheet Ascii (*.CSV)** Choose this file type to save the raw intensity data currently Visible in the Trace Preferences dialog (for any graph box) as a comma delimited ASCII Spreadsheet file (*.CSV) file. The data is saved as XY pairs delimited by a comma.

*Note* This file type can be easily imported into a spreadsheet program. For example, Microsoft Excel. You can also double-click on the stored *.CSV file from Microsoft Explorer to load Excel automatically with the ASCII XY data displayed.

**GRAMS (*.SPC)** Choose this file type to only save the data currently Visible in the Trace Preferences dialog (for any graph box) as a Cary Eclipse GRAMS data file.

Tell me more about Filename Extensions

**Save only focused trace** Select this option to save only the focused trace in the graph box. If this option is not selected, all visible traces in the graph box will be saved.

**Save** Click this button to save the file currently present in the File name field in the directory specified in the Look in field.

See also: File menu ASCII Spreadsheet file (*.CSV)

### Filename extensions—Thermal

The Cary Eclipse system uses the filename extension to distinguish between data types (e.g. method or data) and also to differentiate applications. (e.g. Concentration or Thermal).

The Syntax for file extensions is:

<table>
<thead>
<tr>
<th>Letter</th>
<th>Represents</th>
<th>Available Options</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
The following files may be opened and saved in the Thermal Application:

<table>
<thead>
<tr>
<th>Extension</th>
<th>Available action</th>
<th>File type</th>
</tr>
</thead>
<tbody>
<tr>
<td>*.FMTM</td>
<td>Open/Save</td>
<td>A method file (*.FMTM) consists of the software setup by which data is collected.</td>
</tr>
<tr>
<td>*.FDTM</td>
<td>Open/Save</td>
<td>A data file (*.FDTM) contains data collected that is currently visible in the Trace Preferences dialog (for any graph box).</td>
</tr>
<tr>
<td>*.FRTM</td>
<td>Open/Save</td>
<td>A report file (*.FRTM) contains the report generated by the Cary Eclipse about the data.</td>
</tr>
<tr>
<td>*.FGTM</td>
<td>Open/Save</td>
<td>A graph template file (*.FGTM) contains information that sets up the layout of the Graphics area including any bitmaps as well as the range and style of the graph axes.</td>
</tr>
<tr>
<td>*.FBTM</td>
<td>Open/Save</td>
<td>A batch file (*.FBTM) contains the collected data that was Visible in the Trace Preferences dialog (for any graph box), the method by which the data was collected and the report and graphics template.</td>
</tr>
<tr>
<td>*.CSV</td>
<td>Open/Save</td>
<td>An ASCII spreadsheet file (*.FCSV) has the same extension for all applications. It consists of raw intensity data (that was currently Visible in the Trace Preferences dialog (for any graph box) saved as XY comma-delimited pairs. This file can be imported into a spreadsheet program such as Microsoft Excel.</td>
</tr>
<tr>
<td>Data.*</td>
<td>Open</td>
<td>A Cary DOS (Data.*) file is the equivalent of a Cary Windows Data file—it does not contain any Batch information.</td>
</tr>
</tbody>
</table>
An ASCII Spreadsheet file (*.CSV) consists of raw intensity data saved as XY comma-delimited pairs. You can choose to import or export Cary Eclipse files to ASCII files, which can then be imported in spreadsheet programs such as Microsoft Excel.

The format is as follows:

- Line 1 contains the sample name
- Line 2 contains X and Y labels. The name of the X label in Thermal is Temperature (°C). The Y label should be intensity (a.u.)
- Lines 3 and onward contain the X,Y data
- After the last line of X,Y pairs there must be a blank line separating the data from any text audit strings or the end of the file.

For example, if a thermal file was collected as:  
Temp (°C) = 94.99, Intensity = 196.775  
Temp (°C) = 94.55, Intensity = 196.620  
Temp (°C) = 94.03, Intensity = 197.256  
etc.

It would appear as follows in the ASCII file:

```
Thermal,  
Temp (°C); Intensity (a.u.)  
30, 0.0255,  
60, 0.0500,  
80, 0.0750,  
```
Microsoft’s Excel generates a CSV file with the above format.

There is also an alternative format for CSV files. For example,

<table>
<thead>
<tr>
<th>Thermal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Temp (°C); Intensity (a.u.)</td>
</tr>
<tr>
<td>30, 0.0255</td>
</tr>
<tr>
<td>60, 0.0500</td>
</tr>
<tr>
<td>80, 0.0750</td>
</tr>
</tbody>
</table>

See also:
- Save As
- Open
- How to export collected data

**How to combine data files into a batch file—Thermal**

To combine a number of data files into a batch file, do the following:

Step 1

Select **Open…** from the **File** menu to display the Windows **Open** dialog.

Step 2

A list of stored batch files will appear. Select the down arrow to the right of the **Files of type** field and select 'Data' to list all the data files.

Step 3

Select the **Overlay Data** check box.

Step 4

Highlight the data files that you require. Hold down the CTRL key to select multiple files.

Step 5

Select **Open**. The highlighted data files will load into the application and appear in the same graph box.

Step 6
Select **Save As**… from the **File** menu to display the Windows **Save As** dialog.

**Step 7**

A list of stored data files will appear. Select the down arrow at the right of the **Files of type** field and select 'Batch' to list all the batch files. Ensure that the **Save only focused trace** check box is cleared.

**Step 8**

In the **File name** field, type in the file name for your new batch file.

**Step 9**

Select **Save** to create the new batch file. The current method will be stored with the batch file.

All the data files are now combined into the one batch file.

**How to export collected data—Thermal**

If you want to export data from a Cary Eclipse application into a third-party application, then you simply need to save your data as an Spreadsheet ASCII (*.CSV) file or a Rich Text Format (*.RTF) file. Spreadsheet ASCII files are recommended if you are planning to use the data in a spreadsheet program such as Microsoft® Excel. Rich Text Format files are recommended if you are planning to open the data in a word processor program such as Microsoft® Word.

To export collected data:

**Step 1**

From the **File** menu, select **Save As**. The **Save As** dialog is displayed.

**Step 2**

Click on the down arrow next to the **Files of type** list box and select the **Spreadsheet ASCII (*.CSV)** or **Rich Text Format (*.RTF)** option.

**Step 4**

Type the name of your new file in the **File name** field.

**Step 5**

Select **Save** to export your data to this new file.

**Software and Screen features—Thermal**

- **Menu line**
- **Status display**
- **Status line**
Menu line—Thermal

- File menu
- Edit menu
- View menu
- Commands menu
- Setup menu
- Graph menu
- Help menu

Status Line—Thermal

The Status Line is displayed at the very bottom of the Thermal Application window. The line will display the following information (from left to right):

<table>
<thead>
<tr>
<th>Hardware Status</th>
<th>This field displays status messages indicating the current instrument activity.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sequence Information</td>
<td>This field displays status messages indicating the current system activity.</td>
</tr>
<tr>
<td>Current Results</td>
<td>This field displays information on the last reading.</td>
</tr>
</tbody>
</table>

Thermal Application window

Buttons—Thermal

The major commands in this application can be accessed both from the menu items at the top of the main window and from the application buttons. These buttons, which appear in the left side of the Thermal Application window, comprise the following:

Hot Tip

If you wish to access these commands from the main menus, rather than from these buttons, you can remove the Command buttons from your application window. To do this, clear the Buttons command in the View menu. This will enlarge the Graphics area, making viewing and selection of thermals easier.

<table>
<thead>
<tr>
<th>Button</th>
<th>Enables you to...</th>
</tr>
</thead>
<tbody>
<tr>
<td>Setup</td>
<td>Click this button to display the Setup dialog where you can set up your Cary Eclipse for the collection, storage and analysis of the thermal run. You can also set up any accessories that you may have and define the reporting options.</td>
</tr>
<tr>
<td>Zero</td>
<td>Select this to zero the current ordinate value. This command can also be accessed by pressing F5. Note: Before you select Zero, ensure that you have placed the blank solution in the cell...</td>
</tr>
</tbody>
</table>
positions that are currently in the cell path.

<table>
<thead>
<tr>
<th>Function key</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>This displays the Help for the current dialog or window.</td>
</tr>
<tr>
<td>F4</td>
<td>Press F4 to display the Go to Wavelength dialog.</td>
</tr>
<tr>
<td>F5</td>
<td>Press F5 to perform an instrument Zero. The current intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before selecting Start if you want your data to start at 0 intensity. This command is also accessible from the Commands menu or by selecting the Zero button.</td>
</tr>
</tbody>
</table>

Note
Before you select Zero, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.
Right mouse menu—Thermal

The following commands are accessible by clicking the right mouse button on editable fields in the Cary Eclipse software:

<table>
<thead>
<tr>
<th>Command</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Undo</strong></td>
<td>If available, this reverts to the previous value before the Edit on the field.</td>
</tr>
<tr>
<td><strong>Cut</strong></td>
<td>If available, this cuts the highlighted text to the clipboard.</td>
</tr>
<tr>
<td><strong>Copy</strong></td>
<td>If available, this copies the highlighted text to the clipboard.</td>
</tr>
<tr>
<td><strong>Paste</strong></td>
<td>If available, this pastes the text from the clipboard to the cursor position.</td>
</tr>
<tr>
<td><strong>Delete</strong></td>
<td>If available, this deletes the highlighted text/number.</td>
</tr>
<tr>
<td><strong>Select All</strong></td>
<td>If available, this selects all entries.</td>
</tr>
</tbody>
</table>

How to remove or display the command buttons—Thermal

The command buttons (Setup, Zero, Pre-thermal, Set Temp, Stop Time, Clear Report, Recalculate and Print) appear on the left side of the Thermal Application window.

To remove the command buttons

Step 1

From the Thermal Application window, select the View menu.

Step 2

Clear the Buttons menu item.
The buttons will disappear from the application screen, giving you more room to display reports.

To show the command buttons

Step 1

From the Thermal Application window, select the View menu.

Step 2

Select the Buttons menu item.

The buttons will re-appear in the application screen.

ADL—Thermal

- [ADL Command Line](#)
- [ADL Hooks](#)
- [How to insert ADL commands](#)

ADL Command Line—Thermal

Introduction to ADL

Use the ADL Command Line to run Single-line or Multi-line Applications Development Language (ADL) commands that enable you to perform functions over and above that of the existing program functionality.

ADL is not a general purpose programming language. It has been developed specifically by Varian to meet the needs of spectroscopists. It can be used to handle all the data collected from the Cary Eclipse—both photometric values and entire data collections. ADL programs that you develop should be stored with the extension 'ADL'.

To access the ADL Command Line, select the ADL Command item from the Commands menu. You can also press ALT+Y to open the dialog.

**Note**

Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.

Software controls

| Single Line | Selecting this lets you enter ADL commands on a single line into the Command Line entry box. |
Q. Can I enter more than one ADL command on a single line?
A. Yes, provided that the commands are separated by a semicolon ‘;’.

Multi-line
The Multi-line capability enlarges the Command Line entry box. Use the Multi-line capability to create a more complex ADL program by adding a series of commands over multiple lines. When Multi-line mode is selected, the Quick Reference option becomes available.

Quick Reference
Select Quick Reference to open the Quick Reference dialog where you can easily add a command in the correct syntax to your ADL program. This option is only available for Multi-line commands.

Show me How to insert ADL commands using the Quick Reference
A detailed description of all the commands in the Quick Reference is available by pressing the Help button on the Quick Reference dialog.

Browse
Select Browse to access the Open dialog where you can browse for and select an existing ADL program to load and run.

Command Line (entry box)
This entry box enables you to use ADL commands. You can enter single ADL commands into this field, or you can enter a series of ADL commands. If you have the entry type set to Single Line, you can review previous ADL entries by selecting the down arrow to the right of the list box.

The ADL commands in the Command Line are executed when you select OK.

Note
On entering this window, the Cary Eclipse will recall the last command or program executed from the Command Line entry box. The ADL commands entered in the last executed program are typically stored in the Varian\Cary Eclipse WinFLR\ADL directory on the drive on which the Cary Eclipse software is installed.

Hot Tip
You can rerun the ADL commands in the Command Line entry box from any screen in the Thermal application by pressing Alt-X or selecting Re-run ADL from the Commands menu.

See also:
ADL Hooks

ADL Hooks—Thermal

The ADL Hooks command is accessible from the View menu.

Instead of writing a full ADL program you can use ADL hooks to link into an existing application. This is particularly useful if you want to make small changes to the way an application works, e.g. you want to pop up a dialog for the user to enter some specific data before the run starts. ADL hooks are links at specific points in an application at which you can run an ADL program. For example, ADL hooks have been supplied to run the following ADL programs:

Note
Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.
<table>
<thead>
<tr>
<th>Hook</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>TM_Startup.FADL</td>
<td>This is run when the Thermal application starts. For this to to work the application must be started from a batch or method file that has ADL Hooks on.</td>
</tr>
<tr>
<td>TM_PreSeq.FADL</td>
<td>This runs when the <strong>Start</strong> button is selected.</td>
</tr>
<tr>
<td>TM_PostSeq.FADL</td>
<td>This runs at the end of the <strong>Start</strong> button sequence.</td>
</tr>
<tr>
<td>TM_Online.FADL</td>
<td>This runs when Cary Eclipse is detected online.</td>
</tr>
<tr>
<td>TM_OffLine.FADL</td>
<td>This runs when Cary Eclipse goes offline.</td>
</tr>
<tr>
<td>TM_Zero.FADL</td>
<td>This runs when the Zero button is selected.</td>
</tr>
<tr>
<td>TM_Recalc.FADL</td>
<td>This runs when the <strong>Recalculate</strong> button is selected.</td>
</tr>
<tr>
<td>TM_Shutdown.FADL</td>
<td>This runs when the application is shutting down.</td>
</tr>
<tr>
<td>TM_PreRead.FADL</td>
<td>This runs just before a read is taken.</td>
</tr>
<tr>
<td>TM_PostRead.FADL</td>
<td>This runs just after a read is taken (It has the reading result stored in the UserResult variable.)</td>
</tr>
<tr>
<td>TM_PreSetup.FADL</td>
<td>This runs when the <strong>Setup</strong> button is selected.</td>
</tr>
<tr>
<td>TM_PostSetup.FADL</td>
<td>This runs when the <strong>OK</strong> button on the <strong>Setup</strong> dialog is selected.</td>
</tr>
<tr>
<td>TM_CellChange.FADL</td>
<td>This runs instead of the Cary Eclipse driving the Multicell Holder. It enables you to add your own instructions for the Cary Eclipse Multicell Holder or to use your own cell changing accessory.</td>
</tr>
<tr>
<td>TM_PreStage.FADL</td>
<td>This runs prior to each collection stage. It determines the stage by checking the UserResult variable as UserResult = stage number.</td>
</tr>
<tr>
<td>TM_PreUserRecalc.FADL</td>
<td>This runs just after the <strong>Recalculate</strong> button has been selected and performs any pre-processing required prior to a recalculation. If the ADL Variable UserBoolean is set to FALSE, it can also replace the normal recalculations.</td>
</tr>
<tr>
<td>TM_UserRecalc.FADL</td>
<td>This runs just prior to the calculations and can also be used to replace the calculations.</td>
</tr>
<tr>
<td>TM_PostUserRecalc.FADL</td>
<td>Runs after the <strong>Recalculate</strong> sequence has been completed</td>
</tr>
<tr>
<td>TM_TraceName.FADL</td>
<td>Allows the name of the sample to be obtained from an ADL file, instead of being entered by typing into the sample loading dialog. This only works for single samples.</td>
</tr>
<tr>
<td>TM_UserPreCalc.FADL</td>
<td>This runs just prior to the calculations. Any pre-processing of data should be done here.</td>
</tr>
<tr>
<td>TM_UserPostCalc.FADL</td>
<td>This runs after the calculations. Any post processing of data should be</td>
</tr>
</tbody>
</table>
### How to insert ADL commands using the Quick Reference—Thermal

The Quick Reference window is accessed from the ADL Command window. It enables you to easily add a command in the correct syntax to your ADL program. To do this:

**Step 1**
Press Alt-Y to open the **ADL Command Line** window.

**Step 2**
Select the **Multi-line** option. The Command Line entry box will expand to take multiple lines of ADL commands and the Quick Reference check box is displayed.

**Step 3**
Select the **Quick Reference** check box. The Quick Reference window is displayed.

**Step 4**
If necessary, use the **Go To** field to jump to the appropriate part of the Quick Reference listing.

**Step 5**
Highlight the command you require in the Quick Reference list.

**Step 6**
Click the **Insert in ADL** button. The selected ADL command will appear in the Command Line entry box.

**Step 7**
Where necessary, alter the defaults supplied by the inserted Quick Reference command in the Command Line entry box to suit your program.

**Step 8**
Repeat steps 4–7 to add more commands as required.

---

<table>
<thead>
<tr>
<th><strong>Command</strong></th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>TM_DuringSeq.FADL</td>
<td>Runs when the collect sequence has been paused</td>
</tr>
<tr>
<td>TM_PreStage.FADL</td>
<td>Runs prior to each stage in a multistage (advanced) run</td>
</tr>
<tr>
<td>TM_PostStage.FADL</td>
<td>Runs after each stage in a multistage (advanced) run</td>
</tr>
</tbody>
</table>

---

See also:
ADL Command Line

Help and Safety information—Thermal

- Help menu
- Safety home page

Help menu—Thermal

Use the Help menu to view the online Help and software information for this application.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Thermal Help</td>
<td>Select this to display the Thermal Home Page. This contains a list of online Help topics specific to the Thermal Application.</td>
</tr>
<tr>
<td>Help Topics</td>
<td>Select this to display the Cary Eclipse Windows Online Help Home Page.</td>
</tr>
<tr>
<td>Tip of the day</td>
<td>View useful tips about using your Cary Eclipse instrument and software.</td>
</tr>
<tr>
<td>Eclipse Tour</td>
<td>Select this to display the Cary Eclipse Multimedia Online Help. This will give you an animated introduction to the Cary Eclipse software and hardware.</td>
</tr>
<tr>
<td>About…</td>
<td>Select About to access a dialog that displays information about the application as well as Varian email and Internet contact information. To clear this About dialog, select OK.</td>
</tr>
</tbody>
</table>

Safety Home Page

Click on the desired index entry to view the Help.

- About Safety
- Lamp module
- Ultraviolet radiation
- Electrical hazards — Cary Eclipse
- Panels, covers and modules
- Other precautions
- Warnings and cautions
- Warning symbols
- Color coding
- Information symbols
- FCC advisory
- CE compliant products

Thermal How to…

Select the procedure you require below for a step-by-step informative guide on running the application software.
Procedural

- How to perform a run and determine Tm using Derivative calculations
- How to perform Derivative calculations on previously stored data
- How to perform a run and determine Tm using Hyperchromicity calculations
- How to perform Hyperchromicity calculations on previously stored data
- How to use the ruler during Hyperchromicity calculations

General

- How to email your results
- How to export collected data
- How to combine data files into a batch file
- How to remove or display the command buttons
- How to insert ADL commands using the Quick Reference

See also:
Thermal Home Page
Graphics How To

How to resize graph boxes—Thermal

Step 1
Click on the graph that you want to resize so that the highlight box appears around it.

Step 2
Move the cursor to the edge of the highlight box and place it over one of the square marks (e.g. ■) in the box until it turns into a double headed arrow (←→).

Step 3
Click and hold the left mouse button and drag the box to the desired size.

Step 4
Release the left mouse button.

See also:
How to alter the look of your graphs

How to zoom a graph—Thermal

To zoom a graph, do the following:

Step 1
Click on the button on the toolbar, or select Cursor Mode from the Graph menu. The Cursor Modes dialog will appear.
Step 2

Select **Free** and select the **OK** button.

Step 3

Click and hold down left mouse button in the highlighted graph box.

Step 4

Drag the mouse sideways and up or down. You will see a zoom box made up of a dotted line appear on the screen. (See picture below.)

![The dotted line is a zoom box.](image)

**Hot Tip**

To move the box on the screen, hold down the left and right mouse buttons together. You can then position the box anywhere in the graph box. Releasing the right mouse button will still allow you to resize the zoom box.

Step 5

Release the left mouse button. The area defined by the zoom box will be redrawn to fill the entire graph box, and the thermals within the box will be enlarged accordingly. The abscissa (X) and ordinate (Y) coordinates displayed below the graph will also be changed to reflect the positions of the corners of the zoom box.

You can repeat this process if you need to zoom in even further.

**Note**

The system may limit the extent to which you can Zoom.

See also:

- How to scale your graph manually
- How to scale your graph automatically

**How to take the derivative of a trace**

Some uses of derivatives are:

- To resolve partly overlapping peaks
- To quantitatively determine trace compounds
- To characterize pure compounds
- Purity testing

To take the derivative of a collected trace and plot the result, follow these steps:
Step 1

Click on the trace to be smoothed. It will become red (highlighted).

Step 2

Click the Selected Trace or Focused Trace button. The name of the highlighted trace will appear in the calculation display field.

Step 3

In the Trace group, select your derivative order in the Operation field. Choose from derivatives 1 to 4 (Deriv1, Deriv2, Deriv3, Deriv4). You may have to scroll to find the derivative you require.

Step 4

Enter a Filter Size and an Interval in the corresponding fields.

Step 5

In the Display Options group, select New Graph to plot the result in a new graph box, or Selected Graph to plot the result in the current graph box.

Step 6

Click the Apply button in the Trace group. Your Deriv selection will be displayed before the trace name in the calculation display field.

Step 7

Click on the equals (=) button. The result will be displayed in the Graphics area.

See also:

About derivatives

**How to smooth a trace**

To smooth a collected trace and plot the result, follow these steps:

Step 1

Click on the trace to be smoothed. It will become red (highlighted).

Step 2

Click on the Maths button in the toolbar (it's on the far right) or select the Maths menu.

Step 3

Click the Selected Trace or Focused Trace button. The name of the highlighted trace will appear in the
calculation display field.

Step 4

In the Trace group, select Smooth in the Operation field. (You may have to scroll to find it.)

Step 5

Enter a Filter Size and an Interval in the corresponding fields.

Step 6

In the Display Options group select New Graph to plot the result in a new graph box, or Selected Graph to plot the result in the current graph box.

Step 7

Click the Apply button in the Trace group. The word ‘Smooth’ will appear before the trace name in the calculation display field.

Step 8

Click on the equals (=) button. The result will be displayed in the Graphics area.
# Validate Home Page

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<th>Use the…</th>
<th>Show me…</th>
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</thead>
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                    • Configure page | • How to setup and perform Validation tests  
                                                  • How to care for standard and reference materials |
| Performing Tests | • Installation Tests  
                    • Instrument Tests  
                    • ASTM  
                    • All Tests | • How to setup and perform Validation tests |
| Viewing run progress and results | • View menu  
                   • Status Display  
                   • Reports  
                   • Print Preview | • How to use advanced reporting options |
| Performing calculations | • Maths Calculator  
                    • User Data Form | • Maths How To |
| Display data graphically | • Graph menu  
                    • Graphics area | • Graphics How To |

<table>
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<tr>
<th>To find out about…</th>
<th>See the…</th>
<th>Show me…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Validate Application</td>
<td>• Validate Application</td>
<td></td>
</tr>
</tbody>
</table>
| File management | • File menu  
                   • Open  
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| Software and screen features | • Menu Line  
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| ADL | • ADL Command Line  
                    • ADL Hooks | • How to insert ADL commands |
| Help and safety information | • Help menu  
                    • Safety Home Page | |

See also:  
[Validate How To](#)  

**Validate Application**
Introduction

Use the Validate Application to optimize the settings and validate the accuracy of your Cary Eclipse Instrument by executing a number of pre-defined tests. The tests are preset with default parameters that comply with international standards for Good Laboratory Practices. You can use this application to perform wavelength checks and calibrations of various Cary Eclipse functions. The Validate Application can only be used for validating your Cary Eclipse instrument. The application cannot be used to validate accessories.

This application runs the tests in a sequence that ensures that tests requiring your intervention are run first. Tests that do not require your intervention are performed last. Consequently you can leave the instrument unattended once you no longer needed to change filters or solutions in the sample compartments.

Certified test kits are available from Varian for use with the Validate application. Refer to the Cary Eclipse Eclipse Parts and Supplies list for details.

Note:
Due to the sensitivity of the fluorescence technique, Validate tests must be performed with the covers on the instrument so as to minimize detection of room light.

To access the Validate Application, click the Windows Start button, point to Programs, Cary Eclipse Eclipse and then select Validate.

Software controls

The application comprises the following:

<table>
<thead>
<tr>
<th>Instrument Status Readings</th>
<th>The Instrument Status Readings, which display the Y and X values of the graph axes are displayed in the top left and right corners of the Validate Application window.</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Y value</strong></td>
<td>Displays the current Y value, usually intensity. This value is continually updated during a data collection.</td>
</tr>
<tr>
<td><strong>X value</strong></td>
<td>Displays the current X values, in nanometers, for both excitation and emission.</td>
</tr>
<tr>
<td>Start and Stop buttons</td>
<td>The Start and Stop buttons at the top of the Validate Application window are used to start and stop instrument analyses. These buttons can also be accessed from the Commands menu.</td>
</tr>
<tr>
<td></td>
<td><strong>Tell me more about the Start and Stop buttons</strong></td>
</tr>
<tr>
<td>Graph area</td>
<td>The Graphics area refers to the plotting area of the application. It can contain traces of collected data, labels and pictures. In the Cary Eclipse Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area defined by the axes is referred to as a graph box.</td>
</tr>
<tr>
<td></td>
<td>To display or hide the Graphics area, select the Graphics command on the View menu. Double-clicking on the Graphics area enables you to toggle between half or full size</td>
</tr>
</tbody>
</table>
display sizes. To manually size the Graphics area, place the mouse cursor over the border between the Graph and Report areas until the cursor changes to a double arrow (↕). Then hold down the mouse and drag it up or down to resize the areas.

**Tell me more about the Graph area**

**Report area**
Use the Validate Report area to view the current report. The report may include the results of your data collection, the method parameters used to collect the results and any graphics you have included. You can select the information to include in your report on the Reports page of the Setup dialog.

You can copy the generated report onto the Windows clipboard. To do this, highlight the report (or the relevant section) and press CTRL+C. You can then paste the report into a word processor such as Microsoft® Word.

**Hot Tip**
You can edit the Validate Report by pressing CTRL+R or selecting the Edit Report command from the Edit menu.

**Commands**
The Validate Application window contains the following commands, all of which can also be accessed from the various menus in the application.

*Click on the buttons to view information about them:*

- Tests...
- Setup ...
- Zero
- Clear report
- Print ...

**Hot Tip**
By becoming familiar with these commands (Tests, Setup, Zero, etc.), you can remove these buttons from the Validate window. This will enlarge the Graphics and Report areas and make it easier to view results. To do this, clear the Buttons item in the View menu.

**Menus**
The Concentration Application contains the following menus:

- File, Edit, View, Commands, Setup, Graph, Calibration, Help

See also:

- Validate Home Page
- Keyboard function keys

**Tests—Validate**

Use the Tests dialog to set up your instrument for a performance tests. You can use the dialog to configure your instrument to check its performance based on the regulatory specifications you select.

**Tip:**
To access the Tests option, select the Tests button from the Validate Application window.

Select the tab you require to display Varian specifications and instrument setup information for each test.

**Note:**
The tests in the Validate application will have most of their fields disabled, as they will be set to
values that are appropriate for achieving guaranteed instrument specifications. However, if the Cary Eclipse software is under GLP control and you have administrator privileges, you will be able to alter these parameters.

- Configure
- Wavelength Accuracy (Xe)
- Wavelength Accuracy (Ho)
- Wavelength Reproducibility (Xe)
- Wavelength Reproducibility (Ho)
- Zero %T Accuracy
- Spectral Bandwidth Accuracy
- Sensitivity
- Stray Light
- Maximum Signal to Noise
- Stability
- ASTM (Detection Limits)

See also:
Regulations
Validate Application window

**Regulations—Validate**

Regulatory authorities demand that generated analytical data complies with Good Laboratory Practices (GLP). A range of performance tests for fluorescence spectrophotometers is specified by the British Pharmacopoeia (BP), European Pharmacopoeia, the US Pharmacopoeia (USP) and the Australian Code of GMP for Therapeutic Goods (TGA)—Guidelines for Laboratory Instrumentation Appendix D2.

The specifications in these tests are a minimum set of criteria for a general purpose UV-Vis spectrophotometer. You need to use an instrument that meets these specifications for your analyses.

These performance tests should be performed routinely on a monthly basis or whenever the instrument has been disturbed or serviced, or when malfunctioning is suspected.

See also:
Tests
Configure
Validate Application
How to perform a validation test

**Configure—Validate**

Use the **Configure** page to select the Validate tests you require.

The page comprises the following:

<table>
<thead>
<tr>
<th>Enable Test Suite</th>
<th>The following test suites are available for selection. The number of tabbed pages displayed and the <strong>Test Order</strong> list will change depending upon the test suite selection.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>To view the tests contained in each of the following suites, select on the links below:</td>
</tr>
</tbody>
</table>
The following Installation Tests are available in the Validate application:

- Wavelength Accuracy Ex. Mono (Xe)
- Wavelength Accuracy Em. Mono (Xe)
- Spectral Bandwidth Accuracy Ex. Slit
- Spectral Bandwidth Accuracy Em. Slit
- Sensitivity 350/397
- Stray Light

See also:
- Configure
- Validate Application

Wavelength Accuracy (Xe)—Validate
Poor wavelength accuracy can produce low quantitative results as the intensity measurement may have been made on the side of the peak rather than at the peak. Wavelength accuracy is determined by scanning a known wavelength peak and calculating the difference between the theoretical wavelength peak and the wavelength peak as reported by the instrument. The tests are each divided into two tests, one for the emission monochromator and one for the excitation monochromator.

There are two methods used to test for wavelength accuracy:

- Xenon (Xe) emission line method
- Holmium perchlorate method

The first involves scanning two peaks in the xenon lamp spectrum. The second uses the transmission minima in the spectrum of a solution of holium perchlorate.

**Note:**
The parameters on this page will have most of their fields disabled, as they will be automatically set to values that are appropriate for achieving guaranteed instrument specifications. Simply select the test check box to include it in the Test Order list on the Configure page.

It is recommended that you leave the Validate parameters at their default settings, however if the Cary Eclipse software is under GLP control and you have administrator privileges, you will be able to alter these parameters.

**Xenon (Xe) emission line method**

This method scans the Xenon (Xe) source emission line at 541.92 nm and 260.54 nm. A ground glass filter is placed in the sample position when measuring the wavelength accuracy of the emission monochromator. The filter is supplied with the instrument. The application compares the expected wavelength peak to that of the theoretical position. The procedure is separated into two tests, consisting of the following:

- Validation of the excitation monochromator
- Validation of the emission monochromator

**Sample required**

*Excitation monochromator:*
No sample is required for this tests as it is the signal from the reference PMT that is being monitored.

*Emission monochromator:*
A ground silica screen diffuser must be placed in the sample position.

**Specifications required to pass the test**

Peak $< \pm 0.5$ nm from 541.92 nm
Peak $< \pm 1.0$ nm from 260.54 nm

**What if my instrument fails to meet the specification?**

- Check that you have set the tolerances to values that are within the specifications of the instrument.
This test is dependent on correct alignment and function of the Xenon source lamp. Check the lamps and alignment.

- Use the wavelength calibration command from within Validate to recalibrate the instrument.
- Perform the wavelength accuracy test again.

If it still fails, contact your local Varian Service office, as a service call may be required.

See also:
- Configure
- Regulations
- Validate Application

Spectral Bandwidth Accuracy—Validate

This test checks the positional accuracy of the slit mechanism. The test is performed by scanning the zero order peak and then determining the peak width at half peak height. This is performed on either the emission or excitation monochromator in separate tests.

The emission and excitation monochromators both use a number of slits and they are measured separately. All five available slit options are tested on the emission monochromator. Only three slits can be tested on the excitation monochromator, as scanning at zero order at the two largest slit widths (10 nm and 20 nm) would result in too much light which could flood the detector.

Note:
The parameters on this page will have most of their fields disabled, as they will be automatically set to values that are appropriate for achieving guaranteed instrument specifications. Simply select the test check box to include it in the Test Order list on the Configure page.

It is recommended that you leave the Validate parameters at their default settings, however if the Cary Eclipse software is under GLP control and you have administrator privileges, you will be able to alter these parameters.

Sample required

Excitation monochromator:
No sample is required for this tests as it is the signal from the reference PMT that is being monitored.

Emission monochromator:
A ground silica screen diffuser must be placed in the cell holder.

Note:
It is recommended that the Auto excitation PMT voltage option on the Configure page is selected on for the Emission test. The PMT voltages used in the test will be displayed in the report.

Specifications required to pass the tests

Excitation and emission monochromators

<table>
<thead>
<tr>
<th>SBW (nm)</th>
<th>Minimum</th>
<th>Maximum</th>
</tr>
</thead>
</table>
The most common method of determining the sensitivity of a Cary Eclipse instrument is to monitor the intensity of the Raman band of water as a function of time and determine the signal-to-noise ratio, defined as (mean signal/RMS noise).

An alternate method for determining instrumental sensitivity involves measuring the signal to noise ratio from low concentrations of fluorescent solutions such as fluorescein in 0.01M NaOH.

**Note:**
The parameters on this page will have most of their fields disabled, as they will be automatically set to values that are appropriate for achieving guaranteed instrument specifications. Simply select the test check box to include it in the Test Order list on the Configure page.

It is recommended that you leave the Validate parameters at their default settings, however if the Cary Eclipse software is under GLP control and you have administrator privileges, you will be able to alter these parameters.

The tests used are:

- Raman water 350/397, Raman 2, Raman 3
- Fluorescein detection limit
Raman water 350/397, Raman 2, Raman 3

This method uses the Raman band of water to determine the instrument sensitivity. Distilled water in a clean quartz fluorescence cuvette is required to perform this test. The test will report the signal to noise ratio.

What if the signal to noise is lower than expected?

- Ensure that the instrument has been given adequate time to warm up.
- Ensure the cell containing water has been placed in the sample position in a clean quartz cuvette.
- Perform the sensitivity test again.

If it still fails contact your local Varian Service office as a service call may be required

Fluorescein detection limit

This test method employs a low concentration aqueous solution of the fluorescent dye fluorescein to assess the instrumental sensitivity. Effectively the test is a limit of detection measurement. The test uses parameters similar to the ASTM limit of detection test using quinine sulfate described in section X.XX.

A solution of fluorescein (acid yellow 73, FW 376.3) in 0.01M NaOH at a concentration of around 1nM (1E-09 M) is prepared and the intensity from 10 individual readings averaged. The intensity of the solvent blank (0.01 M NaOH) is then measured 10 times and the standard deviation calculated. The limit of detection is calculated as

\[
\frac{SD_b \times 3}{(M_{FI} - M_B)} \times F_{conc}
\]

Where
- \(SD_b\) = Standard deviation of the blank measurement
- \(M_{FI}\) = Mean fluorescence intensity
- \(M_B\) = Mean Blank intensity
- \(F_{conc}\) = Fluoroscein concentration

Three times the standard deviation of the blank measurement is considered to give a minimum sample signal.

What if my detection limit is too high?

- Check that you have entered the correct concentration for the fluorescein solution you are using into the validate application.
- Check the solution used has been prepared correctly and freshly and the cuvette used is clean.
- Perform the sensitivity test again.
If it still fails contact your local Varian Service office as a service call may be required.

See also:
Configure
Regulations
Validate Application

**Stray Light—Validate**

Stray light is defined as any light that gets to the detector that is not of the selected wavelength. The detector cannot distinguish between stray light and light of the selected wavelength and will add the two together to give a higher intensity reading than expected. The stray light test measures the ratio of the intensity of scattered light detected by the sample PMT at a wavelength which is in the “wings” of the excitation peak, as compared to the intensity detected when the emission wavelength is identical to the excitation wavelength.

The stray light test is performed by first setting both excitation and emission wavelengths to 350 nm. A reading is then taken before the emission monochromator is set to 371 nm, where no emission would be expected to be observed and the values ratioed.

**Note:**
The parameters on this page will have most of their fields disabled, as they will be automatically set to values that are appropriate for achieving guaranteed instrument specifications. Simply select the test check box to include it in the Test Order list on the Configure page.

It is recommended that you leave the Validate parameters at their default settings, however if the Cary Eclipse software is under GLP control and you have administrator privileges, you will be able to alter these parameters.

**Sample required**

A ground silica screen must be placed in the cell holder.

**Specifications required for passing the test**

The stray light ratio should be in the range of -0.0005–0.0005 for the instrument to pass the test.

**What if my instrument fails to meet the specification?**

- Ensure that the ground glass filter is in place.
- Ensure that the sample compartment is tightly sealed. Repeat the test with the room lights turned off. If the instrument passes the test then there is a light leak in the instrument cover somewhere.
- Ensure that the filter is clean and free of any contamination that may fluoresce.

If it still fails, contact your local Varian Service office as a service call may be needed.

See also:
Configure
Regulations
Validate Application
Instrument Tests—Validate

The following Installation Tests are available in the Validate application:

- Wavelength Accuracy Ex. Mono (Xe)
- Wavelength Accuracy Em. Mono (Xe)
- Wavelength Accuracy Ex. Mono (Ho)
- Wavelength Accuracy Em. Mono (Ho)
- Wavelength Reproducibility Ex. Mono (Xe)
- Wavelength Reproducibility Em. Mono (Xe)
- Wavelength Reproducibility Ex. Mono (Ho)
- Wavelength Reproducibility Em. Mono (Ho)
- 0%T Accuracy (Rayleigh scatter)
- 0%T Accuracy (Raman band)
- Sensitivity 350/397
- Sensitivity Raman 2
- Sensitivity Raman 3
- Fluorescein detection limit
- Stray Light
- Maximum Signal to Noise
- Stability

See also:
- Configure
- Validate Application

Wavelength Accuracy (Ho)—Validate

Poor wavelength accuracy can produce low quantitative results as the intensity measurement may have been made on the side of the peak rather than at the peak. Wavelength accuracy is determined by scanning a known wavelength peak and calculating the difference between the theoretical wavelength peak and the wavelength peak as reported by the instrument. The tests are each divided into two tests, one for the emission monochromator and one for the excitation monochromator.

There are two methods used to test for wavelength accuracy:

- Xenon (Xe) emission line method
- Holmium perchlorate method

The first involves scanning two peaks in the xenon lamp spectrum. The second uses the transmission minima in the spectrum of a solution of holium perchlorate.

Note:
The parameters on this page will have most of their fields disabled, as they will be automatically set to values that are appropriate for achieving guaranteed instrument specifications. Simply select the test check box to include it in the Test Order list on the Configure page.
Holmium perchlorate method

This method involves scanning the spectrum of an aqueous solution of 4% w/v holmium oxide in 1.4 M perchloric acid and comparing the wavelengths of the transmission minima with those reported in the Certificate of Analysis for Holmium (III) oxide in perchloric acid. This test provides a wavelength accuracy check independent of the light source.

A scan of the spectrum of holmium perchlorate is made and the positions of the peaks at the following wavelengths: 640.70, 536.70, 485.30, 451.30, 416.40, 361.10, 287.40 and 278.0 nm, are determined.

A ground glass filter, supplied with the Cary Eclipse is placed in the sample position to perform the wavelength accuracy test. After collecting a baseline the holmium perchlorate solution is placed in the special holder on the excitation side of the monochromator and a second scan takes place. A new continuum is created by taking the ratio of the Holmium perchlorate continuum and the blank continuum. From this continuum the positions of the peak maxima, close to the set wavelengths, are determined.

Sample required

A sealed 4% w/v aqueous solution of holium (III) oxide (99.5%) dissolved in perchloric acid contained in a sealed quartz cuvette, and a ground silica screen diffuser.

Specification required to pass the test

Peaks < ± 1.0 nm from certified wavelengths

What if my instrument fails to meet the specification?

- Check that you have set the tolerances to values that are within the specifications of the instrument.
- Use the wavelength calibration command from within Validate to recalibrate the instrument.
- Perform the wavelength accuracy test again.

If it still fails, contact your local Varian Service office, as a service call may be required.

See also:
- Configure
- Regulations
- Validate Application

Wavelength Reproducibility (Xe)—Validate

There are two tests for wavelength reproducibility. The first involves scanning the 541.92 nm peak in the xenon lamp spectrum. The second involves scanning the 451.30 nm transmission minima in the spectrum of a solution of holium perchlorate.

Note:
The parameters on this page will have most of their fields disabled, as they will be automatically set to values that are appropriate for achieving guaranteed instrument specifications. Simply select the test check box to include it in the Test Order list on the Configure page.

It is recommended that you leave the Validate parameters at their default settings, however if the Cary Eclipse software is under GLP control and you have administrator privileges, you will be able to alter these parameters.

The test procedure consists of repeating the wavelength accuracy test ten times to validate the wavelength reproducibility of each monochromator independently. The procedure is separated into two tests, consisting of the following:

- Validation of the excitation monochromator wavelength reproducibility
- Validation of the emission monochromator wavelength reproducibility

The test for for xenon consists of repetitively measuring the wavelength of the 541.92 nm xenon lamp peak for 10 scans and calculating the difference between the largest and smallest measured wavelength.

**Sample required**

*Excitation monochromator:*
No sample is required for this tests as it is the signal from the reference PMT that is being monitored.

*Emission monochromator:*
A ground silica screen diffuser must be placed in the sample position.

**Specifications required to pass the tests**

Difference < 0.2 nm

See also:
- Configure
- Regulations
- Validate Application

**Wavelength Reproducibility (Ho)—Validate**

There are two tests for wavelength reproducibility. The first involves scanning the 541.92 nm peak in the xenon lamp spectrum. The second involves scanning the 451.30 nm transmission minima in the spectrum of a solution of holium perchlorate.

**Note:**
The parameters on this page will have most of their fields disabled, as they will be automatically set to values that are appropriate for achieving guaranteed instrument specifications. Simply select the test check box to include it in the Test Order list on the Configure page.

It is recommended that you leave the Validate parameters at their default settings, however if the Cary Eclipse software is under GLP control and you have administrator privileges, you will be able to alter these parameters.
The test procedure consists of repeating the wavelength accuracy test ten times to validate the wavelength reproducibility of each monochromator independently. The procedure is separated into two tests, consisting of the following:

- Validation of the excitation monochromator wavelength reproducibility
- Validation of the emission monochromator wavelength reproducibility

The test for holium perchlorate consists of repetitively scanning from 448–454 nm to measure the transmission minimum in the region. This scan is performed 10 times, and the maximum difference must have a magnitude of less than 0.2 nm.

**Sample required**

*Excitation monochromator:*
No sample is required for this test as it is the signal from the reference PMT that is being monitored.

*Emission monochromator:*
A ground silica screen diffuser must be placed in the sample position.

**Specifications required to pass the tests**

Difference < 0.2 nm

See also:
- Configure
- Regulations
- Validate Application

**Zero% T Accuracy—Validate**

There are two tests for Zero% T accuracy. The tests measure the 'blackness' or absence of light on the detection side of the Cary Eclipse instrument. The amount of 'blackness' is affected by the amount of light that leaks from outside the instrument as well as the amount of light leaking from the sample chamber when the lamp is flashing but the emission filter is in the **Closed** position.

**Note:**
The parameters on this page will have most of their fields disabled, as they will be automatically set to values that are appropriate for achieving guaranteed instrument specifications. Simply select the test check box to include it in the **Test Order** list on the **Configure** page.

It is recommended that you leave the Validate parameters at their default settings, however if the Cary Eclipse software is under GLP control and you have administrator privileges, you will be able to alter these parameters.

To evaluate the 0% T accuracy, two tests are performed. The first, in which the light levels in the sample chamber are low, such as looking at the Raman band of water, examines the shot noise of the instrument at high sample PMT voltage settings. A second test in which excitation light is scattered off a ground silica screen diffuser with the emission wavelength set to equal that of excitation (simulating high scattered light levels in the sample compartment) is required to examine the electronic noise at low
sample PMT voltages. In both cases the 0%T measurements are taken with the instrument's emission filter closed.

The two tests used are:

- Raleigh scattering
- Raman band of water

**Rayleigh scattering**

Raleigh scattering is phenomenon explained by considering a liquid or gas as being made up of discrete molecules containing minute regions of fluctuating reflective index. Any light passing through the medium will thus be refracted an number of times, when passing through the medium and continue at angles at other than that of the exciting beam. This type of scattering causes light to be emitted from the sample at the same wavelength as the excitation wavelength.

**Sample required**

A ground silica screen diffuser is placed in the cell holder to create a highly scattered source. The emission filter is closed and the reading from the detector taken.

**Specifications required to pass the test**

-0.01 < Signal < +0.01

**What if my instrument fails to meet the specification?**

- Check the water was placed in the sample position in a clean quartz fluorescence cuvette.
- Check the sample compartment lid was fully closed during the test.
- Repeat the test.

If the instrument still does not meet the specification, please contact your local Varian Service office as a service call may be required.

**Raman water**

The Raman band of water simulates the fluorescence effect. The Raman effect is observed as a low level signal produced at a higher wavelength to that of the excitation wavelength. The effect is caused when molecular vibrational energy caused by light scattering is released as a photons at a wavelength different to the excitation wavelength.

As the light emitted at the shifted is of a low intensity the instrument detector PMT voltage is increased. This makes the Raman band ideal as a measurement of low signals at a high PMT voltage.

**Sample required**

A clean quartz cuvette containing distilled water in the cell holder to perform this test. The emission filter is closed and the reading from the detector taken.

**Specifications required to pass the test**
-0.01 < Signal < +0.01

What if my instrument fails to meet the specification?

- Check the water was placed in the sample position in a clean quartz fluorescence cuvette.
- Check the sample compartment lid was fully closed during the test.
- Repeat the test.

If the instrument still does not meet the specification, please contact your local Varian Service office as a service call may be required.

See also:
Configure
Regulations
Validate Application

Maximum Signal to Noise—Validate

This test measures the maximum instrumental signal-to-noise ratio, which involves measuring the signal-to-noise from a highly fluorescent sample such as Rhodamine B.

Note:
The parameters on this page will have most of their fields disabled, as they will be automatically set to values that are appropriate for achieving guaranteed instrument specifications. Simply select the test check box to include it in the Test Order list on the Configure page.

It is recommended that you leave the Validate parameters at their default settings, however if the Cary Eclipse software is under GLP control and you have administrator privileges, you will be able to alter these parameters.

Sample required

A Rhodamine B in PMMA block must be placed in the sample position.

Specifications required to pass the test

S/N > 1000:1

What if my instrument fails to meet the specification?

- Check that you have set the tolerances to values that are within the specifications of the Cary instrument.
- Check that the PMMA block was inserted into position.
- Perform the maximum signal to noise test again.

If it still fails, contact your local Varian Service office as a service call may be required.

See also:
Configure
The Cary Eclipse Stability test measures for instrument stability as a function of time. The stability test involves measuring the signal from a highly fluorescent sample for an extended period of time. The test is similar to the maximum signal-to-noise test except that during the stability test, the signal averaging time is increased to 10 s and the collection time is increased to 1 hour.

Instrument Stability is particularly important when the instrument is to be used over long time periods for example when performing repetitive sample scans or when using a Wellplate reader accessory.

**Note:**

- The parameters on this page will have most of their fields disabled, as they will be automatically set to values that are appropriate for achieving guaranteed instrument specifications. Simply select the test check box to include it in the **Test Order** list on the **Configure** page.

- It is recommended that you leave the Validate parameters at their default settings, however if the Cary Eclipse software is under GLP control and you have administrator privileges, you will be able to alter these parameters.

**Sample required**

A Rhodamine B in PMMA block must be placed in the sample position.

**Specification required to pass the test**

T stab < 0.5%

**What if my instrument fails to meet the specification?**

- Ensure that the PMMA block was placed in the sample position
- Check that you have set the tolerances to values that are within the specifications of the instrument
- Check that the instrument was warmed up before the test commenced
- Perform the stability test again

If it still fails, contact your local Varian Service office as a service call may be required.

**See also:**

- **Configure**
- **Regulations**
- **Validate Application**

**ASTM Tests—Validate**

The following ASTM Tests are available in the Validate application:
Detection Limits

The Detection Limits test adheres to the requirements for ASTM standard E 579-84 "Standard Test Method for Limit of Detection of Fluorescence of Quinine Sulfate". The method employs the signal-to-noise ratio theory to determine the sensitivity of a fluorescence measuring system in testing for the limit of detection (LOD) of quinine sulfate.

The ASTM standard states that “The practical lower limit of this test method is 1 ng of quinine sulfate per ml (one part per billion; 1 ppb”).

Varian does not provide a specification for this test, however it can be used to monitor instrument performance over time.

**Note:**
The parameters on this page will have most of their fields disabled, as they will be automatically set to values that are appropriate for achieving guaranteed instrument specifications. Simply select the test check box to include it in the Test Order list on the Configure page.

It is recommended that you leave the Validate parameters at their default settings, however if the Cary Eclipse software is under GLP control and you have administrator privileges, you will be able to alter these parameters.

**Sample required**

Quinine sulfate in 0.1 N H₂SO₄ at a concentration of 10⁻³ µg/ml (1 ng/ml; 1 ppb) as well as the reagent blank (0.1 N H₂SO₄).

**What if my limit of detection is higher than expected?**

- Ensure that the Quinine sulfate was placed in the sample position
- Check that the solution was prepared correctly and the cuvette was clean
- Repeat the test
- Perform the limit of detection test again.

If it still fails, contact your local Varian Service office as a service call may be required

**Enable All Tests—Validate**
The following tests are available in the Validate application:

- Wavelength Accuracy Ex. Mono (Xe)
- Wavelength Accuracy Em. Mono (Xe)
- Wavelength Accuracy Ex. Mono (Ho)
- Wavelength Accuracy Em. Mono (Ho)
- Wavelength Reproducibility Ex. Mono (Xe)
- Wavelength Reproducibility Em. Mono (Xe)
- Wavelength Reproducibility Ex. Mono (Ho)
- Wavelength Reproducibility Em. Mono (Ho)
- 0%T Accuracy (Rayleigh scatter)
- 0%T Accuracy (Raman band)
- Spectral Bandwidth Accuracy Ex. Slit
- Spectral Bandwidth Accuracy Em. Slit
- Sensitivity 350/397
- Sensitivity Raman 2
- Sensitivity Raman 3
- Fluorescein detection limit
- Stray Light
- Maximum Signal to Noise
- Stability
- Detection Limits (ASTM)

See also:
Configure
Validate Application

**Setup—Validate**

Use the **Setup** dialog to set up your report and email options.

To access the **Setup** dialog select the **Setup** button from the Validate Application window or the **Setup** command from the **Setup** menu.

The **Setup** dialog comprises the following tabbed pages:

- **Reports**
- **Auto store**

### Setup menu—Validate

Use the Setup menu to access the following pages.

<table>
<thead>
<tr>
<th>Command</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Setup</strong></td>
<td>Select <strong>Setup</strong> to display the Setup dialog where you can access the <strong>Reports</strong> and <strong>Auto Store</strong> pages.</td>
</tr>
<tr>
<td><strong>Reports</strong></td>
<td>Choose <strong>Reports</strong> to display the <strong>Reports</strong> page. You can use this page to set up the report</td>
</tr>
</tbody>
</table>


Use the Reports page to set up the report parameters associated with data collection.

From the Setup menu, select Reports to access the Reports page. You can also access the page from the Setup dialog by selecting the Reports tab. To access the Setup dialog, select the Setup button from the Validate Application window.

The page comprises the following:

### Operator

**Name**
Enter the instrument operator name in this text entry field. Use the arrow to the right of this field to view and select one of the last ten entries in this field.

**Note**
In GLP mode the system will automatically enter the operator’s name and this field will not be editable.

**Comment**
Enter operator details and comments in this text entry field.

### Options

This group enables you to select the information to include in your report.

**Auto Print**
Select Auto Print to print a report automatically during the data collection. If Auto Print is selected, then the system will send the report information to the specified printer(s) as well as the Report area. However if Auto Print is not selected, the report will only be sent to the Report area and can be viewed by selecting Report in the View menu.

To automatically obtain a copy of the graphics, you will need to select both the Auto Print and Graph options (see below). In this case, all graphical information displayed on screen will be printed with the report. Therefore it is advisable to arrange the graphical information as you require on screen, prior to printing.

For example, if you require a single graph print out, then the Single/Multi Graph option in the Graph menu should be changed to the correct setting.

**Graph**
Select Graph to include any graphs or graphic information associated with your report. If you require a printout of the displayed graphics only, you must ensure that only this
Report Toolbar—Validate

Use the Report toolbar buttons to gain quick access to many of the Edit menu options. You can also use the toolbar to cut, copy and paste information and to change the look of your report.

To display or hide the toolbar, point to the Toolbars option in the View menu and select Reports. To display or hide the toolbar's text labels, point to the Toolbar option in the View menu and select Text Labels.

Hot Tip
To see the name of a button, point to it with the mouse.

The following commands can be accessed from the Report toolbar:

<table>
<thead>
<tr>
<th>Button</th>
<th>Menu command</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Edit Report</td>
<td></td>
<td>Click this button to edit the report.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>This command is also accessible by pressing CTRL+R.</td>
</tr>
</tbody>
</table>
Clear Report—Validate

Select Clear Report to remove the contents of the Report area. If you have saved the report, selecting this option does not delete the contents of the report, the information is simply removed from the Report Area.

You can select the Clear Report button from the Validate Application window or the Clear Report command from Commands menu.

How to use advanced reporting options—Validate

This outlines ways to fully customize your reports. Options available include:

- Including header and footer information
- Including a logo
- Editing the text in the report
- Viewing the report on screen before printing
• Cutting and pasting to a word processor

Including header and footer information

You can have text appearing at the top and bottom of every page in your report. To specify what the text should be, do the following:

Step 1

On the Windows Taskbar, click the Windows Start button, point to Programs, Cary Eclipse and then select System Information. The System Information application appears with the Company page displayed.

Step 2

Enter the header and footer information in the Header and Footer fields.

Step 3

Click OK to implement your changes.

Including a logo

You can include a logo on the first page of your report. To specify what the logo should be, do the following:

Step 1

On the Windows Taskbar, click the Windows Start button, point to Programs, Cary Eclipse and then select System Information. The System Information application appears with the Company page displayed.

Step 2

Enter a logo using the Add Logo button. This logo will always be included on the front page of reports.

Step 3

Click OK to implement your changes.

Q. Why isn't the logo I selected included in my report?
A. If you are running the Cary Eclipse software under Windows NT you need to be logged on as an Administrator in order to specify the logo in the System Information application.

Editing the text in the report

You can edit any of the text in the Report area. Just select Edit Report from the Edit menu. You can then add, delete and change text in the Report area.
Q. The Edit Report option is not available, why not?
A. You do not have reporting editing privileges set in the GLP Administration application. The Administrator of the system will have to change your privileges before you can edit reports.

Viewing the report on the screen before printing

You can view a report before printing it. To do this, from the File menu select Print Preview. You can then ensure that the report format is correct before you print it.

Hot Tip
Use the arrow keys at the bottom of the Print Preview screen to change pages.

Cutting and pasting to a word processor

To have complete control over how your report looks you may wish to copy it to a word processor program, such as Microsoft Word. To do this:

Step 1
Open a report.

Step 2
From the Edit menu, choose Edit Report.

Step 3
Click in the Report area and highlight the text you want to export to your word processor.

Step 4
Right-click on the Report area and select Copy from the menu.

Step 5
Start up your word processor and use the Paste function (usually under the Edit menu) to place the text into a document.

See also:
How to add a label to a graph
How to add a picture to a graph
How to alter the look of your graphs
How to display a trace that is not appearing

Auto Store—Validate

Use the Auto store page to set up the system to automatically send an email at the end of a read.
To access the **Auto store** page, select the **Auto store** command from the **Setup** menu. You can also access the page from the **Setup** dialog, by selecting the **Auto store** tab. To access the **Setup** dialog, select the **Setup** button from the Validate Application window or the **Setup** command from the **Setup** menu.

The page comprises the following:

**Email**

Select the **Email** check box and enter the Email details to send an email to a person once the read is completed. This useful facility can be used to alert you to the completion of the read, particularly if you are working away from the laboratory where the Cary Eclipse instrument is. You can also attach the collected data to the email.

![Note]

You must set up the email **Host** and **Port** options on the **Email options** page of the System Information application in order for the email facility to work correctly.

**To** Enter the recipient's email address. You can enter more than one recipient in this field.

**From** Enter your email address (or the email address associated with the computer on which the Cary Eclipse software is installed).

**Subject** Enter the subject of your email (or leave this field blank if you do not wish to include a subject).

**Cc** Enter the email address of any recipient you want to copy the email to.

**Message** Enter your message.

**Attached…** Use the Attached button include a file attachment in your email. To do this, select Attached and browse your computer to select a file, using the Windows® Open dialog that appears.

**Attach current data** Select this option to attach the data that has been collected during the run to the email.

See also: How to email your results

**How to email your results—Validate**

You may wish to send an email to a person once the read is completed. This useful facility can be used to alert you to the completion of the read, particularly if you are working away from the laboratory where the Cary Eclipse instrument is.

To do this:

**Step 1**

Ensure that you have set up the **Host** and **Port** options on the **Email options** page of the System Information application in order for the email facility to work correctly.

**Step 2**

From the **Setup** menu, select **Auto Store**. The **Auto Store** page is displayed.
Step 3

Select the **Email** check box to enable the email options.

Step 4

In the **To** field, enter the recipient's email address (for example, *rchandler@varianinc.com*). To enter multiple recipients, simply type a comma ',' between each email address.

Step 5

In the **From** field, enter the email address of the computer on which the Cary Eclipse software is installed. This may be your email address, or a communal email address, if the Cary Eclipse software is installed on a computer that has multiple users.

Step 6

Enter information in the **Subject** and **Message** fields as required.

Step 7

Select **Attach current data**. The data that has been collected during the run will be attached to the email.

**Hot Tip**
You can attach other documents to your email. To do this, select the **Attached** button and browse your computer to select a file, using the Windows® **Open** dialog that is displayed.

Step 8

Select **OK** to clear the **Auto Store** dialog. Your email will be sent once the run has been completed.

See also:
- [Auto Store](#)

### How to set up tests for validation—Validate

- Start the Validate application
- Set up test parameters
- Start the Validate run
- After the run

**Start the Validate application**

Step 1

On the Windows Taskbar, click the **Start** button, point to **Programs, Cary Eclipse** and then select **Validate**. The Validate application appears.
(Alternatively, you can double-click the Validate button in the Cary Eclipse folder on the desktop.)

**Note**

If you are running a GLP system, you may be prompted to enter a password before accessing the Validate application.

**Set up test parameters**

Step 2

From the Validate Application, select the **Tests** button to open the **Tests** dialog. The **Configure** page is displayed.

Step 3

On the **Configure** page, ensure that the **Auto Excitation PMT voltage** option is selected. Selecting this option enables the instrument to set the optimum photomultiplier voltage for the system. The voltages used during the test will be displayed on the report.

Step 4

Select the type of instrument performance tests that you require. The various tests that are available will appear as tabs across the top of this dialog.

If your access privileges are restricted by the GLP application you may find that fields and buttons in this dialog are not accessible. See your administrator to gain access.

Step 5

Click on each tab and select the tests that you require to be performed by ticking the appropriate checkbox.

Change any parameters that you require to be changed to suit your laboratory in each test.

If you wish to save the tests you have set up for future use select **Save Method As** from the **File** menu. The **Save As** dialog will be displayed to enable you to the method with an FMVO extension.

**Start the Validate run**

Step 6

Click the **Start** button or select **Start** from the **Commands** menu to commence the testing procedure.

Follow the instructions on the prompts that appear and select the **OK** button to continue with the performance tests.

You can select the **Cancel** button at any time to stop the Validate run.

**Note**
The results for each test are displayed in the Report area immediately after the test is completed.

**After the run**

Step 7

At the end of the Validate run, the Cary system automatically generates a report file. The report file is stored in the "Cary Eclipse WinFLR" directory.

The Report file will be saved with the following format:

DATE TIME.FRVO

For example,

16 Apr 00 3:57:48 PM.FRVO

The date and time will appear in the same form that is set in the Windows Regional Settings Properties dialog box that is accessed from the Windows Control Panel.

At the end of the Validate run, you can also save your data. To do this, select Save As... from the File menu. The Save As dialog will be displayed to enable you to save the current method, data and report in a batch file (FBVO).

See also:

- **Save As**

**How to care for standard and reference materials—Validate**

- General care
- Cleaning instructions for liquid samples
- Re-calibration of liquid samples
- Ordering NIST reference materials
- Recalibration of NIST filter standards

To maintain optimum optical and reflectance properties of the optical standards, the following care and cleaning instructions must be followed.

**General care**

Although the materials used for these standards are quite durable, care should be taken to prevent contaminants from contacting the materials surface, such as finger oils from contacting the material surface. Always wear clean gloves when handling these materials.
The liquid standards must not be allowed to freeze and it is recommended that they be stored within the temperature range of 5–50 °C.

**Cleaning instructions for liquid standards**

Dirty cells are the greatest single source of error in spectrophotometry. Avoid handling the cells by the polished surfaces. In the event of the faces becoming contaminated, clean by soaking in a neutral detergent solution according to the manufacturers instructions. Rinse thoroughly with deionised water and dry with ethanol. DO NOT use ACETONE. Solution spillage should never be allowed to dry on the cells and should be wiped off with a soft tissue paper or lint free cloth.

**Recalibration of liquid standards**

Starna, the recommended supplier of liquid standards, recommend that their liquid standards sets be recalibrated every two years. However, most accreditation authorities require that standards be recalibrated at least once per year. The calibration of the liquid standards does not usually change significantly within this time interval unless the cell becomes cracked, marked or scratched. If this occurs, the cell will require re-polishing to restore its accuracy.

For recalibration of the Starna liquid standards, please contact your nearest Starna distributor. Alternatively you can contact:

<table>
<thead>
<tr>
<th>Starna Pty Ltd.</th>
<th>Optiglass Ltd.</th>
</tr>
</thead>
<tbody>
<tr>
<td>PO Box 113, Thornleigh, NSW 2120, AUSTRALIA</td>
<td>52/54 Fowler Road, Hainault Industrial Estate, HAINAULT, ESSEX 1G6 3UT, ENGLAND</td>
</tr>
<tr>
<td>Email: <a href="mailto:sales@starna.com.au">sales@starna.com.au</a></td>
<td>Telephone: +44 181 501 5550</td>
</tr>
<tr>
<td>FREE CALL: 1 800 252 284</td>
<td>Facsimile: +44 181 501 1118</td>
</tr>
<tr>
<td>Telephone: +61-2-9484-0033</td>
<td></td>
</tr>
<tr>
<td>Facsimile: +61-2-9484-0055</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Starna Cells Inc.</th>
<th>Starna GmbH</th>
</tr>
</thead>
<tbody>
<tr>
<td>5950 Traffic Way, ATASCADERO, CA 93423, USA</td>
<td>Postfach 1206, D-64311 Pfungstadt, WEST GERMANY</td>
</tr>
<tr>
<td>Telephone: 1 805 466 8855</td>
<td>Telephone: +49 6157 7953</td>
</tr>
<tr>
<td>Facsimile: 1 805 461 1575</td>
<td>Facsimile: +49 6157 85564</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Starna Ges.mbH Wien</th>
<th>Starna S A R L</th>
</tr>
</thead>
<tbody>
<tr>
<td>Skallgasse 20, A-3403 Klosterneuberg, AUSTRIA</td>
<td>Sopares SA, Societe Parisienne D'Equipement Scientifique, 51 Bix, Rue Raymond-Lefevre, 94527 Gentilly, CEDEX, FRANCE</td>
</tr>
<tr>
<td>Telephone: 02243 85009</td>
<td>Telephone: +33 1 4986 4401</td>
</tr>
</tbody>
</table>
Ordering NIST reference materials

Purchase orders (in English) for all NIST SRMs/ RMs should be directed to:

<table>
<thead>
<tr>
<th>National Institute of Standards and Technology Standard</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reference Materials Program</td>
</tr>
<tr>
<td>Room 204, Building 202</td>
</tr>
<tr>
<td>Gaithersburg, MD 20899-0001</td>
</tr>
<tr>
<td>USA</td>
</tr>
<tr>
<td>Telephone: +1-301-975-6776</td>
</tr>
<tr>
<td>Facsimile: +1-301-948-3730</td>
</tr>
<tr>
<td>Email: <a href="mailto:SRMINFO@enh.nist.gov">SRMINFO@enh.nist.gov</a></td>
</tr>
</tbody>
</table>

Each purchase order should give the number of units, catalogue number, and name of each reference material requested.

Example: 1 each, SRM 930e, Glass Filter Set, Transmittance

The following information must be included with each order:
a billing address, a shipping address, name of customer, telephone number, fax number, purchase order number, a customer identification number, i.e., a social security number (SSN) for consumer customers, tax identification number (TIN) for commercial customers, or agency code (ALC) for U.S. government customers.

Recalibration for the NIST Filter standards

For recalibration of the NIST filter standards, please contact your nearest NIST distributor. Alternatively you can contact:

<table>
<thead>
<tr>
<th>Calibrations Program</th>
</tr>
</thead>
<tbody>
<tr>
<td>National Institute of Standards and Technology</td>
</tr>
<tr>
<td>Room A104, Building 411</td>
</tr>
<tr>
<td>Gaithersburg, MD 20899-0001</td>
</tr>
<tr>
<td>USA</td>
</tr>
<tr>
<td>Telephone: +1-301-975-2002</td>
</tr>
<tr>
<td>Facsimile: +1-301-926-2884</td>
</tr>
<tr>
<td>Email: <a href="mailto:CALIBRATIONS@enh.nist.gov">CALIBRATIONS@enh.nist.gov</a></td>
</tr>
</tbody>
</table>

Performing tests—Validate

- [Calibration](#)
- [Start/Stop buttons](#)
- [Go To dialog](#)
Calibration menu—Validate

Use the Calibration menu to open or save a method or data file, print or preview a report, or exit the Cary Eclipse application.

The following commands can be accessed from the Calibration menu:

- **Wavelength Calibration**: Select this option to perform a wavelength calibration. The Xenon emission lines at 0 nm and 541.92 nm are scanned with both the emission and excitation monochromators to ensure that the wavelength scale is accurate. You will be prompted to insert a diffuser during the calibration of the emission monochromator. Note that you should perform an EHT calibration before a wavelength calibration.

- **EHT Calibration**: Select this option to perform an EHT calibration. This will calibrate the signal on the excitation reference detector. The instrument will scan across the whole wavelength range, at each slit width, using the Xenon emission lines to ensure that the detector response is within the required range.

See also: Tests

Start/Stop buttons—Validate

The following buttons may be displayed at the top of the Validate Application window. The Start and Connect buttons will change depending upon the status of the instrument. For example, if the instrument is ready for a run, the Start button will be visible to indicate that the system is ready to commence a run.

- **Start**: Select Start to commence a Validate run using the selected tests. If any unsaved data exists in the Graphics area you will be prompted to save the data. If you choose not to save, any unsaved data, pictures and labels will be deleted when you start the run.

- **Connect**: If another application is already online with the Cary Eclipse, the Start button will change to Connect, and you will need to select Connect to bring this application online before the Start button will re-appear. (You will not be able to connect if the other application is collecting data.)

- **Stop**: Select Stop to end a Validate run.

See also: Graphics area
          Commands menu
          Auto Store page
          Validate Application
Go To Wavelength—Validate

Use the **Go To Wavelength** dialog to temporarily change the instrument to a new excitation or emission wavelength. You would mainly use this dialog if you want to quickly go to a wavelength and check the intensity reading of the sample. The lamp will flash continuously in this mode. This option is only available if the instrument is not currently collecting data.

To access the **Go To Wavelength** dialog, select the **Go To Wavelength** command from the **Commands** menu. You can also select F4 to open the dialog.

**Note**
This is not the wavelength used in the application method. If your method uses a wavelength that is different from the **Go To Wavelength** setting you select, then when you select **Start** the system will change to the wavelength selected (as set on the **Cary** page of the **Setup** dialog).

The Go to Wavelength page comprises the following:

**Excitation**  
**Wavelength**
Enter the new wavelength in this numeric entry field. When you select **Go To** the instrument will change to the new value.

**Zero Order**
Select **Zero Order** to drive the Cary Eclipse fluorescence spectrophotometer to 0 nm (white light). You can then use this light for aligning accessories.

**Emission**  
**Wavelength**
Enter the new wavelength in this numeric entry field. When you select **Go To** the instrument will change to the new value.

**Zero Order**
Select **Zero Order** to drive the Cary Eclipse fluorescence spectrophotometer to 0 nm (white light). You can then use this light for aligning accessories.

**Go To**
Click **Go To** once you have entered a new wavelength, to change the instrument to the entered value.

See also:
- **Commands menu**
- **Function Keys**

Rapid Result—Validate

Use the **Rapid Result** dialog to take a reading of a blank and sample/s, without having to change the current application method. This means that between runs, while new samples are being prepared and loaded, other samples can quickly be measured. The result is printed in the Report area.

To access the **Rapid Result** dialog, select the **Rapid Result** command from the **Commands** menu or press F6.

**Excitation**  
**Wavelength**
Enter the wavelength at which you want to take a reading.
Zero order
Select this option to set the excitation monochromator to zero order (white light).

Emission

Wavelength
Enter the wavelength at which you want to take a reading.

Zero order
Select this option to set the excitation monochromator to zero order (white light).

Zero
Select Zero to change the wavelength to the entered value and measure a blank.

Result
Select Result to change the wavelength to the entered value and measure a sample. The X,Y results are corrected for the blank and appear in the reports area.

See also
Commands menu

Zero—Validate

Use the Zero button to zero the current intensity value. The intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before commencing a data collection if you want your data to start at 0 intensity. The Cary Eclipse will prompt you to load a blank before performing the zero.

Before you select Zero, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.

When the result is zeroed, the word 'Zeroed' will appear in the Y display box in the top left corner of the Validate Application window.

Select the Clear Zero command from the Commands menu to remove zero correction and thus remove the subtraction effect.

To access the Zero option, select the Zero command from the Commands menu, or select the Zero button from the Validate Application window.

See also:
Validate Application
Commands menu

Commands menu—Validate

Use the Commands menu to access major operational commands in the Validate application. The menu is accessible from the menu line at the top of the Validate Application window.

Alternatively you can use function key shortcuts for some of the commands. These appear next to the item in the Commands menu. Note that not all commands appear as both button and menu items.

Hot Tip
If you access these commands using the Commands menu, rather than from the application buttons, you can remove these buttons from your application window. To do this, clear the Buttons command in the View menu. This will enlarge the Graphics area, making viewing and selection of scans easier.
The following commands are accessible from the **Commands** menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Start</td>
<td>Select this to commence the selected tests.</td>
</tr>
<tr>
<td></td>
<td><strong>Note</strong></td>
</tr>
<tr>
<td></td>
<td>If the <strong>Connect</strong> command is displayed in place of the <strong>Start</strong> command, this indicates that another Cary Eclipse application is in use and has control of the Cary Eclipse instrument. To make the Scan application active so that you can perform a read, you will need to select <strong>Connect</strong>. However, you will not be able to select <strong>Connect</strong> if another application is collecting data.</td>
</tr>
<tr>
<td>Stop</td>
<td>Select this to stop the Validate run.</td>
</tr>
<tr>
<td>Clear Zero</td>
<td>Select this option to clear the <strong>Zero</strong> command. When you select the <strong>Zero</strong> command (from the <strong>Commands</strong> menu or by pressing F5), the intensity at that time will be reset to zero and all subsequent results will have the intensity value that was reset to zero subtracted from them. Selecting <strong>Clear zero</strong> cancels this zeroing (and thus subtraction) effect.</td>
</tr>
<tr>
<td>Zero Order</td>
<td>Select the <strong>Zero order</strong> command to set one of the monochromators to zero order. The Cary Eclipse software will use the zero setting when collecting data.</td>
</tr>
<tr>
<td></td>
<td>The <strong>Zero order</strong> command is generally used during alignment procedures.</td>
</tr>
<tr>
<td></td>
<td><strong>Excitation</strong></td>
</tr>
<tr>
<td></td>
<td>Select this command to set the excitation monochromator to zero order (white light).</td>
</tr>
<tr>
<td></td>
<td><strong>Emission</strong></td>
</tr>
<tr>
<td></td>
<td>Select this command to set the emission monochromator to zero order (white light).</td>
</tr>
<tr>
<td>Reset</td>
<td>Select this command to re-initialize the instrument. This will perform the same initializing procedure that is performed when the instrument is powered on.</td>
</tr>
<tr>
<td>Rapid Result…</td>
<td>Select this to display the <strong>Rapid Result</strong> dialog. Use this dialog to take a reading of a blank and multiple sample readings without changing the application method. You can only do this when the Validate application is not performing instrument tests. This command can also be accessed by pressing F6.</td>
</tr>
<tr>
<td>Zero</td>
<td>Select this to zero the current intensity value. The intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before selecting <strong>Start</strong> if you want your data to start at 0 intensity. This command is also accessible by pressing F5.</td>
</tr>
<tr>
<td></td>
<td><strong>Note</strong></td>
</tr>
<tr>
<td></td>
<td>Before you select <strong>Zero</strong>, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.</td>
</tr>
<tr>
<td>Go to…</td>
<td>Select <strong>Go To…</strong> to display the <strong>Go To Wavelength</strong> dialog where you can change the instrument to a new wavelength. Use this command if you want to quickly go to a wavelength to check the intensity reading. The command is only available if the instrument is not currently collecting data. It can</td>
</tr>
</tbody>
</table>
Viewing run progress and results—Validate

- Reports page
- Reports toolbar
- View menu
- Edit menu
- Status display
- Print Preview dialog
- Print
- How to use the advanced reporting options

View menu—Validate

The View menu is accessible from the menu line at the top of the Validate Application window.

The following commands can be accessed from the View menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>Use this to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Toolbars</td>
<td>Select this option to toggle the Graphics and Reports toolbar on and off. You can also select to display or hide the toolbar Text Labels.</td>
</tr>
<tr>
<td>Buttons</td>
<td>Select this option to toggle the left-hand button display (button area) on and off.</td>
</tr>
<tr>
<td>Graphics</td>
<td>Select this option to toggle the Graphics area on and off.</td>
</tr>
<tr>
<td>Report</td>
<td>Select this option to toggle the Report area on and off.</td>
</tr>
<tr>
<td>Report Vertical</td>
<td>If the Graphics option is selected, select Report Vertical to display the Graphics area, Report area and/or ADL Editor side by side vertically.</td>
</tr>
<tr>
<td>ADL</td>
<td>Select this option to toggle the ADL Editor on and off.</td>
</tr>
<tr>
<td>ADL Vertical</td>
<td>If the ADL and Report options are selected, select ADL Vertical to display the ADL</td>
</tr>
<tr>
<td><strong>Vertical</strong></td>
<td>Editor vertically on the right hand side of the Report area.</td>
</tr>
<tr>
<td><strong>ADL Quick Reference</strong></td>
<td>This displays the Quick Reference window which you can use to easily add a command in the correct syntax to your ADL program. A detailed description of all the commands in the Quick Reference is available by selecting the Help button on this window.</td>
</tr>
</tbody>
</table>
| **ADL Hooks** | Select this to activate a link (or hook) to an ADL program at a specific point in the application, and then run that ADL program. For example, ADL hooks have been supplied to run specific programs:  
  - on start-up of the Cary Eclipse software;  
  - when the system goes online or offline;  
  - when the Start button is selected;  
  - when a run finishes;  
  - when a zero is performed etc.  
  
  For the hook to work, ADL Hooks must be selected, and the appropriate ADL file for the application must be present in the Varian/Cary Eclipse WinFLR/ADL folder on the drive on which the Cary Eclipse software is installed. |
| **Hints** | Select this option to toggle the Hints bubble text on and off. Hint text appears when you place the arrow cursor over a field, button or text entry field for a short period. It offers a quick explanation of the item’s function. |
| **Status Display** | Select this option to display the Status Display window, where you can view the status of various parameters during the data collection. |

See also:
- Validate Application
- Graphics area
- ADL Hooks
- How to insert ADL commands using the Quick Reference
- Status Display

## Edit menu—Validate

Use the **Edit** menu to copy, cut, paste and delete information. You can also use the menu to access the Report editing commands.

The following commands can be accessed from the **Edit** menu:

**Note**

These commands are also accessible from the **Report Toolbar**. To display the **Report Toolbar**, from the **View** menu, point to **Toolbars** and select **Report**.
**Status Display—Validate**

Use the **Status Display** window to view the status of your scan during the data collection. Depending upon settings you have for the scan, the following parameters may be displayed:

- Data Mode
- Scan Mode
- Average time
- Current Excitation Wavelength
- Reference PMT Voltage (V)
Excitation Slit width
Excitation Filter
Data Interval

To access the **Status Display** window, select **Status Display** from the **View** menu, or from the Configure page of the **Tests** dialog. You can choose to view the Status Display window at any time during a data collection.

**Print Preview—Validate**

Use the **Print Preview** dialog to view the current report before you print it. From the **File** menu, select **Print Preview** to access the dialog.

The contents of the Graphics area appear first in the preview area followed by the contents of the Report area. You can reduce or enlarge the print preview on screen. To do this, click the left mouse button in the preview area to toggle through the five available views. Click the right mouse button in the preview area to return to the full page view.

The page comprises the following commands:

**First arrow**  Use this arrow to view the first page of the report.

**Previous arrow**  Use this arrow to view the previous page of the report.

**Next arrow**  Use this arrow to view the next page of the report.

**Last arrow**  Use this arrow to view the last page of the report.

**Print**  Use this to display the Windows **Print** dialog where you can choose the printer and pages to print as well as the number of copies you wish to print.

**Q.** Why are the arrow buttons grayed out?

**A.** The buttons will be grayed out if the report has only one page of data.

**Print—Validate**

Use the **Print** dialog to select the printer and pages to print of the currently displayed report as well as the number of copies you wish to print. From the **File** menu, select **Print** to access the dialog.

All graphical information displayed on screen will be printed with the report, therefore it is advisable to edit the report on the screen prior to printing. You can also preview the report prior to printing by using the **Print Preview** command.

If you require a printout of the graphics in the Graphics area only, select the **Clear Report** button to erase the contents of the Report area before you print.
Performing calculations—Validate

- Maths Calculator
- Maths Operations
- Maths How To

Maths window

Use the Maths window to perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator.

To access the Maths window, select the Calculator command from the Graph menu, or select the Calculator button on the Graphics toolbar.

The Maths window is only available in the Kinetics, Lifetimes, Scan and Validate applications.

The Maths window comprises the following:

- **Calculation display field**: View the current calculation, including the trace/s being used, the trace operation being carried out and any functions that have been applied to the trace.

  **Hot Tip**
  Click on the down arrow adjacent to this field to view the last ten unique calculations that have taken place. Use this list to keep track of previous manipulations and repeat calculations quickly by simply selecting and then applying the desired calculation.

- **Selected Trace/Focused Trace**: Choosing either of these options selects the highlighted (red) trace in the current Graph box for manipulation. When choosing between these options, use this simple rule:
  - Use the **Selected Trace** option when the selected calculation is only to be applied to one trace.
  - Use the **Focused Trace** option when the same calculation is to be applied to a number of traces.

The following example illustrates the difference between these options.

To perform the same manipulation on several traces:
a. Highlight a trace by clicking on it with the mouse. The trace will become red.
b. Select **Selected Trace, Apply** and then = to plot the result of the calculation
c. Highlight the next trace and repeat step b.

OR

a. Highlight the first trace.
b. Select **Focused Trace, Apply** and then = to plot the result of the calculation;
c. Highlight the next trace and select the = button to perform the same calculation
on the newly selected trace.

**Trace**
Use the Trace group options to select the operation to be performed on the traces, the
filter size and the interval (where applicable).
Click Apply to insert the selected operation into the equation
Click = (equals) to display the result of the calculations.

**Operation**
Click on an operation to find out more about it

- Anisotropy
- Polarization
- Sum Function
- Mean
- Boxcar Averaging
- Moving Average
- Savitzky-Golay
- Spline
- Deriv 1
- Deriv 2
- Deriv 3
- Deriv 4

**Display Options**
Select whether the results of the operation performed on the trace will be viewed in a
New Graph box or in the Selected Graph box. If you choose the Selected Graph option,
you will be able to directly compare the results with existing traces.

**Functions**
This group contains number and function buttons that can be used as a standard
calculator in addition to being applied to traces.

To use the functions:

1. select a trace or value
2. select either a second trace or number
3. select the equals (=) button to display the result

All the basic mathematical functions are available (addition, subtraction, multiplication
and division), as well as logarithm (Log) and square root (Sqrt) functions. When used
as a calculator, the results of equations appear in the Report area.

**Additional buttons**
Click C to clear the entire operation in the calculation display field.
**CE**
Click CE to clear the last entry entered in the calculation display field.
Click = (equals) to start the operation shown in the calculation display field.

See also:
Maths Home Page
Maths How To

Maths Operations

The following operations can be selected on the Maths window. To access the Maths window, select the Calculator command from the Graph menu, or select the Calculator button on the Graphics toolbar.

Anisotropy, Polarization and Sum Function

The term Anisotropy refers to an unequal angular distribution about a point in space. Anisotropy also refers specifically to a simple expression for measuring the degree of polarization of the fluorescence. It is almost identical to the expression for polarization apart from having a factor of two in the denominator. Anisotropy is usually represented by the lowercase letter "r", while polarization by an uppercase "P".

The two expressions are listed below:

Anisotropy
\[ r = \frac{I_{vv} - G I_{vh}}{I_{vv} + 2G I_{vh}} \]

Polarisation
\[ P = \frac{I_{vv} - G I_{vh}}{I_{vv} + G I_{vh}} \]

where

\( I_{vv} \) equals the intensity of fluorescence with excitation polarizer = vertical, emission polarizer = vertical
\( I_{vh} \) equals the intensity of fluorescence with excitation polarizer = vertical, emission polarizer = horizontal

\( G \) is a factor that accounts for the polarization bias of the instrument. In particular it accounts for the wavelength-dependent polarization bias of the emission side of the instrument (dominated by the polarization bias of the emission grating).

The G factor is given by:
\[ G = \frac{I_{hv}}{I_{hh}} \]

where

\( I_{hv} \) equals the intensity of fluorescence with excitation polarizer = horizontal, emission polarizer = vertical
\( I_{hh} \) equals the intensity of fluorescence with excitation polarizer = horizontal, emission polarizer = horizontal

The expression for the anisotropy is a quotient. The denominator of the expression, as given by \( I_{vv} + 2G I_{vh} \) is the ‘sum’ function, and actually represents the total emission...
unbiased by any sample polarization effects.

**Mean**
The Mean operation allows the average of two or more traces to be taken and the resulting trace to be plotted. The mean of the traces can also be used in conjunction with other functions.

The equation used in mean calculations is:

\[(\text{scan 1 + scan 2...+ scan } x)/x\]

The calculation is performed over the common X range of the traces and the trace with the most dense data is used to determine the data points. The other traces are interpolated at the relevant data points.

**Boxcar Averaging**
This algorithm will take the number of points as defined by the Filter size, and average these, displaying the average at the middle X-point of the filter.

For example, for the following:

Data: \((x_1, y_1), (x_2, y_2), (x_3, y_3) \ldots (x_n,y_n)\)

Filer size: \(5\)

The first smoothed data point will be \((x_3, \text{sum}(Y_1:y_5)/5)\), the next \((x_8, \text{sum}(y_6:y_{10})/5)\) \ldots \((x_{n-2}, \text{sum}(y_{n-4}:y_{n})/5)\).

If there are not enough points at the end of the data set to complete the filter size, then these points will be ignored, hence, the gap at the end may be larger than the gap in the beginning. There will be a gap before the first point and after the last point of the smoothed continuum, with respect to the original unsmoothed data. This algorithm is only valid for evenly x-spaced data points.

Within a time scan, (e.g., Advanced collect in kinetics) the data points may be unevenly spaced between different stages. Each stage will be treated as a new collect, hence, there may be large gaps between stages. If there are not enough points at the end of a stage to complete the filter size, then these points are ignored, hence, there may be large gaps at the end of a stage. The filter size will be limited to odd numbers, with the minimum of 3 to a maximum of 101.

**Moving Average**
This algorithm will take the number of points as defined by the Filter size, and average these, displaying the average at the middle X-point of the filter.

For example, for the following:

Data: \((x_1, y_1), (x_2, y_2), (x_3, y_3) \ldots (x_n,y_n)\)

Filer size: \(5\)

The first smoothed data point will be \((x_3, \text{sum}(Y_1:y_5)/5)\), the next \((x_4, \text{sum}(y_2:y_6)/5)\) \ldots \((x_{n-2}, \text{sum}(y_{n-4}:y_{n})/5)\). There will be a gap before the first point and after the last point of the smoothed continuum, with respect to the original unsmoothed data. This algorithm is only valid for evenly x-spaced data points. Within a time scan, (e.g., Advanced collect in kinetics), the data points may be unevenly spaced between different
stages. This will be treated by interpolation of data from the previous stage so as to give evenly spaced data points into the next stage. If there is not enough data for interpolation, then the next stage will be treated as a new collect, hence, there may be gaps and further delays in displaying the point. The filter size will be limited to odd numbers, with the minimum of 3 to a maximum of 101.

Savitzky-Golay

The Savitzky-Golay technique operation enables you to reduce the noise in the currently selected Maths scan by using smoothing.

When you select this operation you will be prompted to enter a Filter Size from 5 to 101. The larger the Filter Size, the more data points that will be smoothed.

Q. I cannot enter a particular value; it keeps changing to another number — why?
A. The Savitzky-Golay technique requires an odd number to be entered, not an even number.

You will also need to enter an Interval. The Interval refers to how many of the averaged points will be plotted on the smoothed trace. The smaller the interval, the more detail there will be in the trace.

Consider selecting an Interval of 1. This would mean that when the smoothed continuum is plotted, every smoothed point will be displayed.

Q. Why can't I see the Filter Size and Interval fields?
A. These will be displayed when you select Smooth in the Operation field.

Spline

Deriv 1–4

The Deriv operations allow you to obtain derivatives of a selected scan. The derivative can be of the orders 1 to 4. The resulting trace is then plotted. The Savitzky-Golay technique is used for derivative calculations.

On selection of the Deriv operation you will be prompted to enter a Filter Size from 5 to 101. The Filter Size refers to the number of points around a particular point that will be averaged to produce a new derived Y value. The lower the Filter Size, the more data points that will be used in the derivative trace.

Q. I cannot enter a particular value; it keeps changing to another number — why?
A. The Savitzky-Golay technique requires an odd number to be entered, not an even number.

You will also need to enter an Interval. The Interval refers to how many of the derived points will be plotted on the resulting trace. The smaller the Interval, the more detail there will be in the trace.

Q. Why can't I see the Filter Size and Interval fields?
A. These will be displayed when you select one of the ‘Derivs’ in the Operation field.

See also:
About smoothing

Maths How To…
Displaying data graphically—Validate

- Graphics area
- Graphics How To
- Graph menu
- Graphics toolbar
- Graph Labels
  - Add Label dialog
  - Edit Label dialog
  - How to add a label to a graph
  - How to edit a label
  - How to move a label
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- Graph Pictures
  - Add Picture dialog
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- Trace Preferences dialog
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  - How to select the focused trace
  - How to display a trace
  - How to remove a trace from a graph
  - How to remove a trace from the trace list
- Graph Preferences dialog
  - How to alter the look of your graphs
- Cursor Modes dialog
  - How to track the focused trace
- Axes Scales dialog
  - How to scale your graph manually
  - How to scale your graph automatically
- User Data Form
- Peak Labels

Graphics area—Validate

The Graphics area of the Validate application appears in the top half of the Validate Application and is used to display traces of collected data. In the Cary Eclipse system, the term 'graph' refers to the plotting
area and is represented by a set of axes. The rectangular area surrounding the graph is referred to as the graph box. A graph will appear in the Graphics area when you select **Start** to commence a scan.

To display or hide the Graphics area, select the Graphics command on the **View** menu. Double-clicking on the Graphics area enables you to toggle between half or full size display sizes.

You can access the functions associated with the Graphics area by doing the following:

- selecting one of the Graphics toolbar buttons;
- selecting a command from the **Graph** menu; or
- clicking the right mouse button in the Graphics area and select a command from the menu

**Note**

Not all functions are available on every menu.

You can customize the graphics area to suit your requirements in the following ways:

**Note**

All of the commands and dialogs listed are accessible from the **Graph** menu.

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**How to set and perform a Validation test**

**How to care for standard and reference materials**

**How to combine data files into a batch file**

**How to email your results**

**How to use advanced reporting options**

**How to remove the command buttons from the application window**

**How to insert ADL commands using the Quick Reference**

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<td>Cursor</td>
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</tr>
<tr>
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<td>How to delete a label</td>
</tr>
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<tr>
<td>Delete pictures from the graph box</td>
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</tr>
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<td>Save the Graphics area you have set up as a graphics template</td>
<td>Save As dialog (from the File menu)</td>
<td>How to create a graphics template</td>
</tr>
</tbody>
</table>

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**Note**

The red trace in any graph box is the one on which all information appears and the one used for the cursor tracking. It is called the focused trace.

---

See also:

- Graphics Toolbar
- Graph menu
- Validate Application Home Page
Graph menu—Validate

Introduction

Use the Graph menu to view data in various graphical formats or in multiple graphs in the Graphics area. You can double-click on the Graphics area to toggle the display through graph and report to graph only.

In the Cary Eclipse system, the term 'graph' refers to the plotting area and is represented by a set of axes. The rectangular area surrounding the graph is referred to as the graph box. A graph will appear in the Graphics area when you select Start to commence a scan.

The Graph menu can be accessed in the following ways:

- from the main menu at the top of the Validate Application window
- by clicking the right mouse button in a graph box or the Graphics area

Note
The red trace in any graph box is the one on which all information appears and the one used for the cursor tracking. It is called the focused trace.

Menu commands

The following commands can be accessed from the Graph menu:

<table>
<thead>
<tr>
<th>Menu command</th>
<th>Button</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Print XY point</td>
<td>Select this to print the cursor's current X and Y coordinates in the report area. This option is only available from the Graph menu that appears when you right-click in a graph box.</td>
<td></td>
</tr>
<tr>
<td>Copy Graph</td>
<td>Select Copy Graph to copy the entire Graphics area to the Windows Clipboard. All of the graphs in the Graphics area will be copied to the Clipboard.</td>
<td></td>
</tr>
<tr>
<td>Paste to Graph</td>
<td>Select Paste to graph to paste the contents of the Windows Clipboard into the Graphics area. Text is pasted as a label, graphics are pasted as pictures.</td>
<td></td>
</tr>
<tr>
<td>Add Graph</td>
<td>Select Add graph to add a new graph box to the Graphics area. You may want to do this if you have, for example, a graph containing existing data and you want to collect some new data in a new graph.</td>
<td></td>
</tr>
<tr>
<td>Remove Graph</td>
<td>Select Remove graph to delete the selected graph box from the Graphics area. The graph box is removed, but the data is not deleted and is still available from the Trace Preferences window. You may want to use remove graph if you have a large number of graph boxes open but only want to focus on some of these.</td>
<td></td>
</tr>
</tbody>
</table>
You can remove multiple graph boxes by first holding down the CTRL key and selecting the graphs to be removed and then selecting the **Remove Graph** button or menu item. Use the SHIFT key to highlight a range of graphs for removal.

| Clear All Traces | Select Clear All Traces to delete all traces from the graph/s. Note that selecting this option simply turns off the Visible property for the traces in the Trace Preferences window. You can redisplay the traces by making the traces Visible again in the Trace Preferences window. |
| Add Label | Select Add Label to display the Add Label dialog where you can enter a text label to be placed on a graph or in the background of the Graphics area. |
| Add Picture | Select Add Picture to display the Add Picture dialog where you can select a picture, bitmap or chemical structure to be placed on a graph or in the background of the Graphics area. |
| Edit Annotation | Select Edit Annotation to alter the content or look of the selected annotation, (either a label or a picture) in the Graphics area. This option is only available if an annotation is selected. If the selected annotation is a label, selecting Edit Annotation will open the Edit Label dialog where you can change the text of the selected label. If the selected annotation is a picture, selecting Edit Annotation will open the Edit Picture dialog where you can replace the selected picture with a new picture. |
| Delete Annotation | Select Delete Annotation to remove an annotation, (such as a label or a picture) from the Graphics area. This option is only available if an annotation is selected. You can use this menu option to remove highlighted annotations in the Graphics area such as labels, pictures and ruler/fit information. |
| Trace Preferences | Select this command to display the Trace Preferences window where you can select the trace(s) to display in the selected graph. |
| Graph Preferences | Select this command to display the Graph Preferences dialog where you can change the appearance of your graph. Any changes you make to the appearance of the graph will be applied to all graphs. |
| Cursor Mode... | Select this command to display the Cursor Modes dialog where you can choose the type of cursor to be used on the screen. For example, you can select to use a free cursor that can move anywhere in the graph or exactly trace the focused scan (red scan). Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the Least Squares method or the Point to Point method. |
| Axes Scales... | Select this command to display the Axes Scales dialog where you can set the scale range for the X and Y axes. |
| Auto scale graph | To automatically scale the graph, point to one of the following options: |
### Autoscale [XY]
Select Autoscale [XY] to automatically scale collected data by scaling the Y height and X width to full screen.

### Autoscale [X]
Select Autoscale [X] to automatically scale collected data by scaling the X width to full screen.

### Autoscale [Y]
Select Autoscale [Y] to automatically scale collected data by scaling the Y height to full screen.

<table>
<thead>
<tr>
<th>Zoom Out</th>
<th>Select Zoom Out to Zoom back to the original X and Y ranges which were used to collect the data (i.e. the method setup conditions of the focused scan). For example, the system will revert back to the X and Y ranges set in the method if you zoom in and out during a data collection.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Single/Multi Graph</td>
<td>Select Single/Multi Graph to toggle between displaying one or all graphs. The graph highlighted in the Multi Graph view will be displayed when you toggle the Single Graph option.</td>
</tr>
<tr>
<td>Auto Arrange Graphs</td>
<td>Select Auto Arrange Graphs to view and arrange all graphs simultaneously. You can move graph boxes manually by clicking the left mouse button outside the graph axes and dragging the graph box to a new position. You can also resize graphs.</td>
</tr>
<tr>
<td>User Data Form</td>
<td>Select User Data Form to display the User Data Form where you can enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data, such as the pH at which data was collected.</td>
</tr>
<tr>
<td>Peak Labels</td>
<td>Select this to the Peak Labels dialog where you can select various labeling and peak reporting functions.</td>
</tr>
<tr>
<td>Calculator</td>
<td>Select Calculator to display the Maths window where you can perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator.</td>
</tr>
<tr>
<td>History List</td>
<td>This displays a list of the current graphs.</td>
</tr>
</tbody>
</table>

See also:
- Graphics toolbar
- Add Label
- Add Picture
- Edit Label
- Edit Picture
- Trace Preferences
- Graph Preferences
- Cursor Modes
- Axes Scales
- User Data Form
Graphics Toolbar—Validate

Use the Graphics toolbar buttons to gain quick access to many of the Graph menu options. You can also use the toolbar to select the graph you want to view and to change the look of your graphs.

To display or hide the toolbar, point to the **Toolbars** option in the **View** menu and select **Graphics**. To display or hide the toolbar's text labels, point to the **Toolbars** option in the **View** menu and select **Text Labels**.

**Hot Tip**
To see the name of a button, point to it with the mouse.

The following commands can be accessed from the Graphics Toolbar:

<table>
<thead>
<tr>
<th>Button</th>
<th>Menu command</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="Image" alt="Trace Preferences" /></td>
<td>Trace Preferences</td>
<td>Click this button to display the <strong>Trace Preferences</strong> window where you can select the trace(s) to display in the selected graph.</td>
</tr>
<tr>
<td><img src="Image" alt="Graph Preferences" /></td>
<td>Graph Preferences</td>
<td>Click this button to display the <strong>Graph Preferences</strong> dialog where you can change the appearance of your graph. Any changes you make to the appearance of the graph will be applied to all graphs.</td>
</tr>
<tr>
<td><img src="Image" alt="Cursor Mode" /></td>
<td>Cursor Mode</td>
<td>Click this button to display the <strong>Cursor Modes</strong> dialog where you can choose the type of cursor to be used on the screen. For example, you can select to use a free cursor that can move anywhere in the graph or exactly trace the focused scan (red scan). Alternatively, you can select a cursor that enables you to draw a line from one point to another to determine the slope using either the Least Squares method or the Point to Point method.</td>
</tr>
<tr>
<td><img src="Image" alt="Axes Scales…" /></td>
<td>Axes Scales…</td>
<td>Click this button to display the <strong>Axes Scales</strong> dialog where you can set the scale range for the X and Y axes.</td>
</tr>
<tr>
<td><img src="Image" alt="Autoscale[XY]" /></td>
<td>Autoscale[XY]</td>
<td>Click the <strong>Autoscale[XY]</strong> button to automatically scale collected data by scaling the ordinate (Y) height and abscissa (X) width to full screen.</td>
</tr>
<tr>
<td><img src="Image" alt="Autoscale[X]" /></td>
<td>Autoscale[X]</td>
<td>Click the <strong>Autoscale[X]</strong> button to automatically scale collected data by scaling the abscissa (X) width to full screen.</td>
</tr>
<tr>
<td><img src="Image" alt="Autoscale[Y]" /></td>
<td>Autoscale[Y]</td>
<td>Click the <strong>Autoscale[Y]</strong> button to automatically scale collected data by scaling the ordinate (Y) height to full screen.</td>
</tr>
<tr>
<td><img src="Image" alt="Zoom Out" /></td>
<td>Zoom Out</td>
<td>Click the <strong>Zoom Out</strong> button to Zoom back to the original X and Y ranges which were used to collect the data (i.e. the method setup conditions of the focused scan). For example, the system will revert back to the X and Y ranges set in the method if you zoom in and out during a data collection.</td>
</tr>
<tr>
<td><strong>Add Label</strong></td>
<td>Click this button to display the <strong>Add Label</strong> dialog where you can enter a text label to be placed on a graph or in the background of the Graphics area.</td>
<td></td>
</tr>
<tr>
<td><strong>Add Picture</strong></td>
<td>Click this button to display the <strong>Add Picture</strong> dialog where you can select a picture, bitmap or chemical structure to be placed on a graph or in the background of the Graphics area.</td>
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<tr>
<td><strong>Single/Multi Graph</strong></td>
<td>Select <strong>Single/Multi Graph</strong> to toggle between displaying one or all graphs. The graph highlighted in the Multi Graph view will be displayed when you toggle the <strong>Single Graph</strong> option.</td>
<td></td>
</tr>
<tr>
<td><strong>Auto Arrange Graphs</strong></td>
<td>Click the <strong>Auto Arrange Graphs</strong> button to view and arrange all graphs simultaneously. You can move graph boxes manually by clicking the left mouse button outside the graph axes and dragging the graph box to a new position. You can also resize graphs.</td>
<td></td>
</tr>
<tr>
<td><strong>Add Graph</strong></td>
<td>Click this button to add a new graph box to the Graphics area. You may want to do this if you have, for example, a graph containing existing data is displayed and you want to collect some new data in a new graph.</td>
<td></td>
</tr>
<tr>
<td><strong>Remove Graph</strong></td>
<td>Click this button to delete the selected graph box from the Graphics area. The graph box is removed, but the data is not deleted and is still available from the <strong>Trace Preferences</strong> window. You may want to use remove graph if you have a large number of graph boxes open but only want to focus on some of these. <strong>Note</strong> You can remove multiple graph boxes by first holding down the CTRL key and selecting the graphs to be removed and then selecting the <strong>Remove Graph</strong> button or menu item. Use the SHIFT key to highlight a range of graphs for removal.</td>
<td></td>
</tr>
<tr>
<td><strong>Clear All Graphs</strong></td>
<td>Click this button to delete all traces from the graph/s. Note that selecting this option simply turns off the <strong>Visible</strong> property for the traces in the <strong>Trace Preferences</strong> window. You can redisplay the traces by making the traces <strong>Visible</strong> again in the <strong>Trace Preferences</strong> window.</td>
<td></td>
</tr>
<tr>
<td><strong>User Data Form</strong></td>
<td>Click this button to display the <strong>User Data Form</strong> where you can enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data, such as the pH at which data was collected.</td>
<td></td>
</tr>
<tr>
<td><strong>Calculator</strong></td>
<td>Click this button to display the <strong>Maths</strong> window where you can perform mathematical manipulations on collected traces. In addition, it can act as a simple numeric calculator.</td>
<td></td>
</tr>
</tbody>
</table>

**See also:**
- Trace Preferences
- Graph Preferences
- Cursor Modes
- Axes Scales
- Add Label
- Add Picture
Graph Labels—Validate

- Add Label dialog
- Edit Label dialog
- How to add a label to a graph
- How to edit a label
- How to move a label
- How to resize a label
- How to delete a label

Add Label—Validate

Use the Add Label dialog to place a label on your graph or into the background of the Graphics area.

To add a label to a graph box, you must have at least one trace Visible in the graph box. (You can select which traces you want to make visible in the Trace Preferences dialog.) If you are viewing multiple graphs, the label will be placed in the graph box that is currently selected. However, you can add a label outside of a graph box in the background of the Graphics area at any time.

To access the Add Label dialog, select the Add Label command from the Graph menu, or select the Add Label button on the Graphics toolbar.

The Add Label dialog comprises the following:

- **To selected trace**
  Select this option to associate a label with the selected trace. When you associate a label with a trace and make the trace visible in the Graph box, the label will also be displayed with the trace.
  When you save the data (as a Batch or Data file) the label is saved with its associated trace.

- **To selected graph**
  Select this option to associate the label with the selected graph. When you associate a label with a graph, the label will always be displayed in the Graph box, regardless of what traces are visible.
  When you save the data (as a Batch or Data file) the label is saved with its associated graph.

- **Selected trace**
  This will identify the trace that is currently selected and display the name of the trace.

- **Font…**
  Click Font to open the Windows Font dialog. In this dialog you can specify the font type, style and size to be used for the label.

- **Label**
  Use the Label field to enter the text for the label. You can enter as many lines of text as you require.

Q. Can I add a label that will print with all my graphs.
A. Yes, you can add your label to the background of the Graphics area. To do this, right-click on a section of the background (gray area) and select **Add Label** from the menu. (You will notice that this menu has less options than the menu that appears when you right-click on a graph.) In the **Add Label** dialog, enter the label to be added to all graphs and select **OK**. The label will appear in the background of the Graphics area.

**Hot Tip**
If you wish to include certain XY co-ordinates on the graph as a label, position your cursor at the appropriate place, right-click and select **Add Label** from the menu. When the **Add Label** dialog appears, the co-ordinate positions will be added to the **Label** field.

**Edit Label—Validate**

Use the **Edit Label** dialog to alter the content or look of your label.

To access the **Edit Label** dialog, right-click on the label you wish to change and select **Edit Annotation** from the menu.

The dialog comprises the following:

- **Font**
  Select **Font** to open the **Font** dialog, where you can specify the font type, style and size to be used for the label.

- **Label**
  Use the **Label** field to alter the text for the label.

**How to add a label to a graph—Validate**

You can add a label to your graph or Graphics area background. To do this:

Step 1

Click on the graph box you require or click outside the graph box(es) to choose the Graphics area background.

**Note**
There must be at least one trace Visible in the graph box before you can add a label to it.

Step 2

Click the **Add Label** button on the toolbar or right-click on the graph and select **Add Label** from the menu. The **Add Label** dialog will appear. The **Add Label** command is also accessible from the **Graph** menu.

Step 3

Click the **Font** button. The Windows **Font** dialog appears. Select the font type, size and style you require for your label. A sample of the font style appears in the **Sample** field. When you are satisfied with your selections, select **OK** to return to the **Add Label** dialog.

Step 4

Enter the label in the **Label** field and select **OK**. The label will be displayed on the selected graph. If necessary, you can move a label so that it is more appropriately placed on your graph.

See also:
- How to move a label
- How to edit a label
- How to resize a label

**How to move a label—Validate**

To move a label so that it is more appropriately placed on your graph or Graphics area background, do the following:

Step 1

Place the cursor on the label.

Step 2

Click and hold down the left mouse button.

**Hot Tip**

If you are having trouble selecting the label, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 3

Drag the label to the correct position and release the mouse button. The label will be displayed at the new position.

**Note**

Labels are transparent so as not to interfere with any traces on graphs.
How to resize a label—Validate

To resize a label so that it is more appropriately sized on your graph or Graphics area background, do the following:

Step 1

Select the label by clicking on it with the mouse. A border will be displayed around the label to indicate it is selected.

**Hot Tip**

If you are having trouble selecting the label, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Position the cursor over one of the corner edges of the label until the cursor becomes a diagonal double-headed arrow. Click and hold down the left mouse button.

![Example of label showing the border that indicates it is selected and the arrow to be used for re-sizing](image)

Step 3

Increase or decrease the size of the label by dragging the mouse. The label text will also be resized. Release the mouse button when the label and text are the correct size.

**Note**

Labels are transparent so as not to interfere with any traces on graphs.

How to delete a label—Validate

To delete a label from a graph or Graphics area background, do the following:

Step 1

Position the cursor on the label and click the left mouse button to select it.
Step 2

Click with the right mouse button and select **Delete Annotation** from the drop-down menu displayed. Alternatively, you can select the **Delete Annotation** command from the **Graph** menu. The label will be deleted from the graph or Graphics area.

**See also:**
- How to add a label to a graph
- How to move a label
- How to edit a label
- How to resize a label

**Graph Pictures—Validate**

- **Add Picture** dialog
- **Edit Picture** dialog
- How to add a picture
- How to replace a picture
- How to move a picture
- How to resize a picture
- How to delete a picture

**Add Picture—Validate**

Use the **Add Picture** dialog to insert a picture or bitmap into the Graphics area. The picture can be added as a background picture or to a particular graph box.

To add a picture to a graph box you must have at least one trace **Visible** in the graph box. (You can select which traces you want to make visible in the **Trace Preferences** dialog.) If you are viewing multiple graphs, the picture will be placed in the graph box that is currently selected. However, you can add a picture outside of a graph box in the Graphics area at any time.

To access the **Add Picture** dialog, select the **Add Picture** command from the **Graph** menu, or select the **Add Picture** button on the Graphics toolbar.

The **Add Picture** dialog comprises the following commands:

- **To selected trace**
  Select this option to associate a picture with the selected trace. When you associate a picture with a trace and make the trace visible in the Graph box, the label will also be displayed with the trace.

- **To selected graph**
  Select this option to associate the picture with the selected graph. When you associate a picture with a graph, the label will always be displayed in the Graph box, regardless of what traces are visible.

- **Load**
  Click **Load** to open the Windows **Open** dialog. Use this dialog to browse your computer...
for the picture you want to insert. The picture must be one of the following formats: DIB, JPG, PGE, BMP, EMF or WMF.

If you are viewing multiple graphs the picture will be placed on the graph that is currently selected. If no graph is selected, the picture will be placed on the Graphics area background.

When you save the data (as a Batch or Data file) the picture is saved with its associated trace or graph.

Q. Can I add a picture that will print with all my graphs.
A. Yes, you can add your picture to the background of the Graphics area. To do this, right-click on a section of the background and select Add Picture from the menu. (You will notice that this menu has less options than the menu that appears when you right-click on a graph.) In the Add Picture dialog, load the picture to be added to all graphs and select OK. The picture will appear in the background of the Graphics area.

See also:
- Trace Preferences dialog
- Graph menu
- Graphics toolbar
- How to add a picture to a graph
- How to move a picture
- How to resize a picture
- How to delete a picture
- How to replace a picture

**Edit Picture—Validate**

Use the Edit Picture dialog to change a picture you have inserted into the Graphics area.

To access the Edit Picture dialog, right-click on the picture you wish to change and select Edit Annotation from the menu.

The Edit Picture dialog comprises the following commands:

- **To selected trace/ graph**
- **Load**

See also:
- How to replace a picture
- How to add a picture
- How to move a picture
- How to resize a picture
- How to delete a picture

**How to add a picture to a graph—Validate**
You can add a picture to your graph or the Graphics area background. To do this:

Step 1

Click on the graph box you require or click outside the graph box(es) to choose the Graphics area background.

Note
There must be at least one trace Visible in the graph box before you can add a picture to it.

Step 2

Click the Add Picture button on the toolbar or right-click on the graph and select Add Picture from the menu. The Add Picture dialog will appear. The Add Picture command is also accessible from the Graph menu.

Step 3

Click the Load button. The Windows Open dialog appears.

Step 4

Browse through your directories to find the picture you want to insert. All pictures must be in the bitmap format and will have the extension .BMP.

Step 5

Highlight the .BMP that you want to insert and select Open.

Step 6

The Add Picture dialog will appear showing a preview of the bitmap. If the bitmap is the correct one select the OK button. The bitmap will be placed onto the graph.

Note
You should take care not to add a bitmap that is too big as it will obscure part or all of your graph.

You can move a picture so that it is more appropriately placed on your graph.

See also:
How to move a picture to a graph
How to delete a picture
How to replace a picture
How to resize a picture

How to replace a picture—Validate

To change a picture that you have added to a graph or Graphics area background do the following:

Step 1
Move the cursor to the picture and select the left mouse button to select it.

**Hot Tip**
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2

Click the right mouse button and select **Edit Annotation** from the drop-down menu or select **Edit Annotation** from the **Graph** menu. The **Edit Picture** dialog is displayed.

Step 3

From the **Edit Picture** dialog, select the **Load** button. The Windows **Open** dialog is displayed.

Step 4

Browse through your directories to find the picture to replace the current bitmap. Picture files must be in one of the following formats: DIB, JPG, JPEG, BMP, EMF or WMF.

Step 5

Highlight the file that you want to insert and select **Open** to return to the **Edit Picture** dialog.

Step 6

The **Edit Picture** dialog will display a preview of the bitmap. If the bitmap is the correct one select the **OK** button. The bitmap will be placed onto the graph. If you need to, move the picture so that it is more appropriately placed on your graph.

**Note**
Do not to add a bitmap that is too big as it will obscure part or all of your graph.

See also:
- [How to add a picture to a graph](#)
- [How to move a picture](#)
- [How to delete a picture](#)
- [How to resize a picture](#)

**How to move a picture—Validate**

To move a picture so that it is more appropriately placed on your graph or Graphics area background, do the following:

Step 1

Place the cursor on the picture.

Step 2
Click and hold down the left mouse button.

**Hot Tip**
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 3
Drag the picture to the correct position and release the mouse button. The picture will be displayed at the new position.

**See also:**
- [How to add a picture to a graph](#)
- [How to delete a picture](#)
- [How to replace a picture](#)
- [How to resize a picture](#)

**How to resize a picture—Validate**

To resize a picture so that it is more appropriately sized on your graph or Graphics area background, do the following:

Step 1
Select the picture by clicking on it with the mouse. A border will be displayed around the picture to indicate it is selected.

**Hot Tip**
If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

Step 2
Position the cursor over one of the corner edges of the picture until the cursor becomes a diagonal double-headed arrow. Click and hold down the left mouse button.

*Example of picture showing the border that indicates it is selected and the arrow to be used for re-sizing*

Step 3
Increase or decrease the size of the picture by dragging the mouse. The picture will also be resized. Release the mouse button when the picture is the correct size.
See also:
- How to add a picture to a graph
- How to move a picture to a graph
- How to delete a picture
- How to replace a picture

**How to delete a picture—Validate**

To delete a picture from a graph or Graphics area background, do the following:

**Step 1**

Position the cursor on the picture and click the left mouse button to select it.

**Hot Tip**

If you are having trouble selecting the picture, it may be that a trace is interfering with the graphic selection. Try zooming in and clicking in a different part of the picture.

**Step 2**

Click with the right mouse button and select **Delete Annotation** from the drop-down menu displayed. Alternatively, you can select the **Delete Annotation** command from the **Graph** menu. The picture will be deleted from the graph or Graphics area.

See also:
- How to add a picture to a graph
- How to move a picture
- How to resize a picture
- How to zoom a graph

**Trace Preferences—Validate**

A single graph box can display many traces. Use the **Trace Preferences** dialog to select the trace(s) to display (make **Visible**) in the selected graph box.

To access the **Trace Preferences** dialog, select the **Trace Preferences** command from the **Graph** menu, or select the **Trace Preferences** button on the Graphics toolbar.

The dialog comprises the following:

**Hot Tip**

The **Trace Preferences** menu is also available on clicking the right mouse button anywhere in the trace listing.

**Trace Preferences table**

**Visible**

Use the **Visible** field to select which traces will appear on the selected graph. To select
or clear the **Visible** field, simply click the left mouse button in this column. A tick will appear next to traces that are to be displayed in the selected graph.

If you are in single graph view, all selected traces will appear on the one graph. If you are in **Multi Graph** view the traces will appear on the graph that is currently selected.

**Hot Tip**

To make all traces Visible, right-click on the table and select the **Select All** command from the menu. Then right-click on the table again and select the **Show Traces** command from the menu.

**Color**

The **Color** field enables you to view the color of each trace that appears on the selected graph. To change the color of a trace, highlight it, click the right mouse button in the **Trace Preferences** table and select **Change Trace Color**. You can then select a color from the **Color** dialog that is displayed.

The red trace in any graph box is the one on which all information appears. This trace, which is used for the cursor tracking, is called the focused trace and it is always red. When you select a trace in the **Color** column it will become the focused trace in the selected graph box.

**Note**

When you change the trace line width in the **Graph Preferences** dialog it also will change the width of the color trace line in this dialog.

**Name of trace**

The **Name of Trace** field contains a description of the traces currently available for display in the selected graph box.

**Filename**

This field displays the filename and path for the trace. If the name is too long to appear in the field you can use the keyboard arrow keys to scroll to the end of the filename or you can resize the **Trace Preferences** window. If the file path for a trace shows 'not saved' then the trace is unsaved data and is only stored in memory.

**Trace Audit Log**

The **Trace Audit Log** displays information about the highlighted trace. This includes instrument parameters and data form information relating to the trace that has been selected. It also logs all information about data manipulation. When more than one trace is highlighted, the **Trace Audit Log** will display information about the trace that was first selected.

**See also:**

- **Graph menu**
- **Graphics toolbar**
- **Trace Preferences menu**
- **Graph Preferences**
- How to make multiple traces visible in a graph
- How to remove a trace from the trace list
- How to remove a trace from a graph

**Trace Preferences menu—Validate**

The Trace Preferences menu is displayed when you click the right mouse anywhere on the **Trace Preferences table**.
The following commands can be accessed from the menu:

Select All This selects or highlights all the traces in the listed on the Trace Preferences page.
Select None This de-selects all the traces in the listing.
Show Traces Select Show Traces to make all of the currently highlighted traces ‘Visible’, meaning that they will be displayed in the current graph box.
Hide Traces Select Hide Traces to remove the ‘Visible’ property from all the traces currently highlighted. The traces will be removed them from the current graph box.
Delete Traces Select Delete Traces to delete all the highlighted traces in all graphs. Note that if you save the data file before you select Delete Traces, the traces are not deleted from the saved file, and they can be retrieved at a later date.
Selecting Delete Traces will delete the traces from the application memory, and it is recommended that you select this option if you need to free up memory on your computer.

Note
If the application is controlled by the GLP Administration application, you will be prompted to save an unsaved data file before it is removed from the application.

Change Trace Color Select Change Trace Color to display the Windows Color dialog where you can change the color of the currently highlighted trace(s). The focused trace will remain red while it retains the focus, and will revert to the selected color when the focus is moved.

See also:
Trace Preferences
Graph Preferences
How to remove a trace from the trace list
How to remove a trace from a graph

How to select the focused trace—Validate

To select the scan that you want to be the focused (red) trace, do one the following:

- From the Graph, click the left mouse button on the trace that you want to make the focused trace.
- From the Graph Legend (if displayed), click the left mouse button on the trace that you want to make the focused trace.
- From the Trace Preferences dialog, click in the Color column of the trace that you want to make the focused trace.

Note
Select the Show Legend check box in the Graph Preferences dialog to display the Graph Legend

See also:
How to track the focused trace
How to view a particular trace

How to display a trace that is not appearing in the graph box—Validate
Step 1
Click on the graph in the Graphics area.

Step 2
Select the Trace Preferences button from the Graphics toolbar, or select Trace Preferences from the Graph menu. The Trace Preferences dialog is displayed.

Step 3
Find that trace that you wish to see and select the Visible option. A tick will appear in the Visible column to indicate that it will now be displayed.

If you wish to make multiple traces visible, do the following:

a. Hold down the CTRL key select all the traces that you want to display.

b. Click the right mouse button and select the Show Traces command from the drop-down menu. Ticks will appear next to the selected traces to indicate that they will now be displayed.

Step 4
Press OK to clear the Trace Preferences dialog. The selected trace(s) will be displayed.

See also:
How to remove a trace from a graph
How to remove a trace from the trace list

How to remove a trace from a graph—Validate

Step 1
Select a graph in the Graphics area.

Step 2
Click the Trace Preferences button on the Graphics toolbar or select Trace Preferences from the Graph menu. The Trace Preferences dialog is displayed.

Step 3
Find that trace that you no longer wish to see and select the Visible option. The tick will be removed from the Visible column to indicate that it will not be displayed.

If you wish to make multiple traces visible, do the following:

a. Hold down the CTRL key select all the traces that you no longer want to display.

b. Click the right mouse button and select the Hide Traces command from the drop-down menu. The ticks will be removed from the selected traces to indicate that they will not be displayed.
Step 4

Select OK to clear the Trace Preferences dialog. The trace will be removed from the graph.

See also:
How to display a trace that is not appearing in the graph box
How to remove a trace from the trace list

How to remove a trace from the trace list—Validate

If your graphs contain unwanted traces, you may wish to delete them (rather than simply making them invisible) in order to free up application memory and aid system performance.

Step 1

Select a graph in the Graphics area.

Step 2

Click the Trace Preferences button on the Graphics toolbar or select Trace Preferences from the Graph menu. The Trace Preferences dialog is displayed.

Step 3

Select the trace to be removed by clicking on it.

Step 4

Click the right mouse button to display the Trace Preferences menu.

Step 5

Select Delete Traces to delete the sample from the trace list. You will be prompted to confirm this action.

Step 6

Select OK to clear the Trace Preferences dialog. The trace will be removed from all graphs.

See also:
How to remove a trace from a graph
How to display a trace that is not appearing in the graph box

Graph Preferences—Validate

Introduction

Use the Graph Preferences dialog to change the appearance of your graph.
All graphs will appear labeled with the X axis label being horizontal and the Y axis vertical. You cannot change the orientation of these labels. However you can alter the font and size of the axes and their labels.

Any changes you make to the appearance of the graph will apply to all graphs until the settings are changed again.

To access the Graph Preferences dialog, select the Graph Preferences command from the Graph menu, or select the Graph Preferences button on the Graphics toolbar.

**Dialog controls**

The Graph Preferences dialog comprises the following:

**Axes Preferences**

- **Axes Color**: Select the color that you want to assign to the axes of the graph/s from the drop down list.
- **Axes Width**: Select the width of the axes from the drop down list. The width, which is in pixels, ranges between one and five pixels.
- **Grid Style**: Select the style of grid for the graph from the drop down list. If you do not wish to include a grid on the graph, select None.
- **Plot Area Color**: Select the color to be used as the background of the graph from the drop down list. If you do not want a background color on the graph, select White.
- **Trace Width**: Select the width of the trace from the drop down list. The width is given in pixels and ranges between one and five pixels.
- **Font**: Click the Font button to display the Windows Font dialog where you can define the font style for the axes of the graph/s. The style and size of the font will be applied to all graph. The style, name and point size of the font appears on the font button.
- **Preview Box**: Preview any changes you make to the axes preferences in the preview box. The changes shown in this box will not be made until you select OK.

**Advanced Options**

- **Auto tick marks**: The Auto tick marks option enables you to approximately set how many tick marks (or points) will be included on the graph axes. The tick marks generated will be whole, rounded, even numbers. The Auto tick marks option is not available if you have selected Major ticks.

  - **Y-axis**: Use the Y-axis slider bar to increase or decrease the number of tick marks (or points) to be included on the Y axis.

  - **X-axis**
Use the X-axis slider bar to increase or decrease the number of tick marks (or points) to be included on the X axis.

**Major ticks**  Use the **Major Ticks** option to enable you to set exactly how many ticks (or points) will be added to the X and Y axes. The number of marks you specify will be divided by the axis range, which can result in odd numbers on the axis. Note that any changes are not reflected in the graph bitmap at the top of the screen.

**Decimals**  Use the **Decimals** option to set the number of X and Y decimal places to be displayed on the axes. To set the number of decimal places, select the **Decimal** option and enter the number in the Y-axis and X-axis combo boxes. Note that any changes are not reflected in the graph bitmap at the top of the screen.

**Trace Style**

**Points**  Select **Points** to display each individual data point collected by the Cary Eclipse on the graph.

**Points and solid lines**  Select **Points and Solid Lines** to display each individual data point collected by the Cary Eclipse on the graph. Additionally, an interpolated line that connects the points will be drawn.

**Solid lines**  If you select **Solid Lines**, the Cary Eclipse system will interpolate the data between the actual, collected data points and show the data as a continuous line.

**Style cycles automatically**  If you select **Style Cycles Automatically**, the Cary Eclipse system will display each of the above three trace styles. This makes it easier to differentiate between traces, especially on black-and-white printers.

**Show Legend**

**Show Legend**  Select **Show Legend** to display a legend in the right side of each graph box. A single left click on a sample name or example line within the legend will make it the focused or red scan. Information about the trace will be displayed below the X axis.

**Hot Tip**

Double-clicking on the legend will automatically display the **Trace Preferences** window, allowing you to view more detailed information about your trace (e.g. audit log, File name, etc.).

See also:
- **Graph menu**
- **Graphics toolbar**
- **Graphics area**
- **Graphics How To**

**How to alter the look of your graphs—Validate**

**Step 1**

Click the **Graph Preferences** button on the Graphics toolbar, or select **Graph Preferences** from the
Graph menu. The Graph Preferences dialog is displayed.

Step 2

Use the available options available to choose your preferred appearance for the graphs. A preview box in the top right-hand corner will demonstrate how the graphs will appear.

Step 3

Select OK to clear the Graph Preferences dialog and update all graphs to reflect your selections.

Note
You can also add labels and pictures to your graphs to alter their appearance.

Step 4

To re-use your graphics setup at a later date, save the setup as a graphics template.

See also:
How to add a label to a graph
How to add a picture to a graph
How to move a label
How to move a picture
How to resize graph boxes

Cursor Modes—Validate

Use the Cursor Modes dialog to choose the type of cursor to be used on screen. For example, you can select to use a free cursor that can move anywhere in the graph.

To access the Cursor Modes dialog, select the Cursor Modes command from the Graph menu, or select the Cursor Modes button on the Graphics toolbar.

The dialog comprises the following:

Free
The Free cursor is not confined to a particular trace on the graph. It can move anywhere in a graph box, and its ‘X,Y’ position is shown in the bottom right-hand side of the Graphics area. You can use the Free cursor to zoom in on a particular part of a graph.

Note
The Free cursor moves from point to point in the selected graph box. Hence you need to be careful when positioning the cursor over a scan to obtain an accurate (X,Y) reading. It is recommended that you use the Track cursor mode for this.

Track
Use the Track cursor to exactly trace the focused (red) scan in a graph box. The cursor position is displayed in the X and Y at the bottom right-hand side of the Graphics area.

Hot Tip
If you wish to fix the cursor on a particular position on the curve, move the
Use the Axes Scale dialog to manually change the axes range. This can assist in zooming in on a particular part of the graph.

To access the Axes Scale dialog, select the Axes Scale command from the Graph menu, or select the Axes Scale button on the Graphics toolbar.

The dialog comprises the following:

### X Axis

- **Mode**: View the current units for the X axis.
- **Scale**: Use the Scale field to enter a new range for the X axis, in the format n1 - n2 (where n1 is the start of the scale and n2 is the end of the scale).

  **Hot Tip**
  The Cary Eclipse will store the ten most recently entered ranges, which you can access by selecting the down arrow adjacent to the Scale field.

### Y Axis

- **Mode**: View the current units for the X axis.
- **Scale**: Use the Scale field to enter a new range for the Y axis, in the format n1 - n2 (where n1 is the start of the scale and n2 is the end of the scale).

  **Hot Tip**
  The Cary Eclipse will store the ten most recently entered ranges, which you can access by selecting the down arrow adjacent to the Scale field.

See also:
Graph menu
Graphics toolbar
How to zoom a graph
How to track the focused trace

Axes Scales—Validate

Use the Axes Scale dialog to manually change the axes range. This can assist in zooming in on a particular part of the graph.
How to zoom a graph

How to scale your graph manually—Validate

Step 1
Click on the graph whose axes you want to change to make it the focused trace.

Step 2
Click the Axes Scales button on the toolbar, or select Axes Scales from the Graph menu. The Axes Scales dialog is displayed.

Step 3
Type in the new ranges for the X and Y axes in the format ‘n-n’, or choose an option for each from the drop-down list.

Step 4
Select OK. The axes on the selected graph will change to reflect your chosen settings.

Hot Tip
Select the Zoom Out command to resize the graph to its original settings.

How to scale your graph automatically—Validate

Step 1
Click on the graph whose axes you want to change to make it the focused trace.

Step 2
To autoscale the graph, select one of the following:

Click the Autoscale XY button on the toolbar to autoscale both X and Y axes.

Click the Autoscale X button on the toolbar to autoscale the X axis only.

Click the Autoscale Y button on the toolbar to autoscale the Y axis only.

The focused trace will be scaled according to the option selected.
User Data Form—Validate

Use the User Data Form to enter extra information about the traces in the highlighted graph box. For example, you might want to record information about the experimental conditions you used to collect the data.

To access the User Data Form, select the User Data Form command from the Graph menu, or select the User Data Form button on the Graphics toolbar. The User Data Form command is only available if you have selected a graph box.

The dialog comprises the following:

**Data Name**
This column lists the names of each visible trace in the highlighted graph box.

Use the extra columns for recording additional information about the experimental conditions you used to collect the data, eg. pH.

**Other Columns**
Use this to record the name of the parameter in which you are interested as a column heading, (for example, pH, ionic concentration, substrate concentration etc.).

**Note**
You must enter a column heading, or else the data in that column will not be saved.

See also:
Graph menu
Graphics toolbar

Peak Labels—Validate

Use the Peak Labels dialog to set up the peak information that will be included in your report.

From the Graph menu, select Peak Labels to access the dialog. Note that this option is only available when a trace is visible.

**Hot Tip**
The selections you make in the Peak Labels dialog are applied to the selected graph(s). To select multiple graphs, hold down the CTRL key and select the graphs. You can also use the SHIFT key to select a range of graphs. When the shift key is held down, all graphs in between the first and last graph chosen are selected.

The dialog comprises the following:

**Peak Style**
This parameter selects the type of peaks that will be reported by the Cary Eclipse
software. Select from the following options:

**Note**
When determining **Peaks, Valleys or Peaks Valleys** the Cary Eclipse software
nominates the highest data point of a peak as the peak position and the lowest data
point of the valley as the valley point. It does not interpolate between points.
Thus, if your data points are relatively far apart (large Data Interval), the reported
peak or valley may not be entirely accurate.

However, when you use the zero crossing points to calculate the peak and valley
positions, the data is interpolated and a more accurate peak position may be
obtained.

**Peaks**
The Cary Eclipse will report maximum points only.

**Valleys**
The Cary Eclipse will report minimum points only.

**Peaks Valleys**
The Cary Eclipse will report both maximum and minimum points.

**Zero Cross**
The Cary Eclipse will report the points at which the trace crosses over the zero ordinate
line. A zero-crossing peak type is where the continuum crosses the X axis, that is at
Y=0. This choice is useful for odd derivative scans calculated in the **Maths** window.
For example, the zero crossing points of a first derivative scan correspond to the peaks
and valleys in the trace.

**Hot Tip**
Taking a first derivative of a noisy scan will reduce the noise and assist in locating
the peaks more accurately.

**Threshold**
Enter the **Threshold** value, which is the value that defines what peaks/valleys will be
displayed. The difference in intensity between a valley, the highest peak and the
adjacent valley is measured and, if this difference is greater than the threshold it is
reported on the trace.

**Maximum**
Select **Maximum** to calculate the maximum peak in the trace/s. Select one of the **Label
options** (X and Y labels, Y label, X label) to display the maximum peak on the trace.

**All**
Select **All** to calculate all peaks above the peak threshold in the trace/s. Select one of the
**Label options** (X and Y labels, Y label, X label) to display the maximum peak on the
trace.

**X and Y Labels**
Select this if you wish the peaks on the graph to be labeled with both the X and Y co-
ordinates.

**Y Label**
Select this if you wish the peaks on the graph to be labeled with only the Y co-ordinates.

**X Label**
Select this if you wish the peaks on the graph to be labeled with only the X co-ordinates.

**No Labels**
When selected, no peaks will be labeled with co-ordinates.

**Position**
Use the slider bar underneath the graph to select the position of the label on the graph.
This option is not available if you have selected **No Labels**.
File Management—Validate

- **File menu**
- **Open dialog**
- **Save As dialog**
- **Filename extensions**
- **Auto Store page**
- **How to combine data files into a batch file**

File menu—Validate

Use the **File** menu to open or save a method or data file, print or preview a report, or exit the Cary Eclipse application.

**Hot Tip**
Highlight a method or data file in Explorer and simply drag and drop it onto the application to open it. Any data will be displayed in the Graphics area.

The following commands can be accessed from the **File** menu:

<table>
<thead>
<tr>
<th>Command</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Open…</td>
<td>Select <strong>Open</strong> to display the <strong>Open</strong> dialog where you can search for and load method, data, report, graph template and batch files, as well as other file types compatible with the Cary Eclipse software. When you load a batch file, the method which collected the data is loaded as well as the associated report and graphics layout.</td>
</tr>
<tr>
<td>Open Method…</td>
<td>Select <strong>Open Method</strong> to display the <strong>Open</strong> dialog where you can search for and load a stored method.</td>
</tr>
<tr>
<td>Save Method As…</td>
<td>Select this option to display the <strong>Save As</strong> dialog and store the current method. You can use the Save As dialog to store the method, data, report, or graphics template separately or together as a Batch file. The data can also be saved as a comma delimited ASCII Spreadsheet file.</td>
</tr>
<tr>
<td>Save As…</td>
<td>Select this option to display the <strong>Save As</strong> dialog and store the collected data. You can use the Save As dialog to store the method, data, report, or graphics template separately or together as a Batch file. The data can also be saved as a comma delimited ASCII file.</td>
</tr>
<tr>
<td>Print Setup…</td>
<td>Select this option to display the Windows Print Setup dialog where you can select the printer, printer page size and orientation.</td>
</tr>
<tr>
<td>Print Preview…</td>
<td>Select this option to display the <strong>Print Preview</strong> dialog where you can view the current report as it will be printed.</td>
</tr>
<tr>
<td><strong>Print…</strong></td>
<td>Select this option to display the Windows <strong>Print</strong> dialog where you can choose the printer and pages to print as well as the number of copies you wish to print.</td>
</tr>
<tr>
<td><strong>Explorer…</strong></td>
<td>Select this option to run Windows Explorer. This may help you locate method and data files.</td>
</tr>
<tr>
<td><strong>History List</strong></td>
<td>This displays a list of the four most recently opened files.</td>
</tr>
<tr>
<td><strong>Exit</strong></td>
<td>Select this option to exit the Validate application.</td>
</tr>
</tbody>
</table>

**See also:**
- Open
- Save As
- Print Preview
- Print
- Filename Extensions

## Open—Validate

Use this dialog to open a file previously saved by the application. To access the **Open** dialog, select **Open** or **Open Method** from the **File** menu.

### Look in

The **Look In** field lists the available folders and files. Click the down arrow on the right of this field to view the location of the current folder in your computer's hierarchy. You can also select a new location simply by selecting a different folder. The folders and files in the selected folder are displayed the list box below the Look In field.

**Note**
To open the folder in the level that is one higher than the current folder, select on the toolbar.

### Look in list box

The **Look In** list box, which is directly below the **Look In** field, displays the folders and files in the selected location. To view the contents of a folder, double-click it.

- **Click to create a new folder.** Type the name of the new folder in the highlighted text entry field that appears and press ENTER. Then double-click on the new folder to open it.
- **Click to display the directory structure of the drive.**
- **Click to display the files as a list.**
- **Click to display files as a detailed list including the file size, file type and the date the file was last modified.**
- **Click to preview the selected trace before you load it into the application.**

### File name

Enter the name of the file you want to open in this field. The easiest way to do this is to find the file in the Look in list box and then select the file. However, you can type the full path of a file and then select **Open** to open the file.
Files of type

Click on the arrow on the right of this field to view a list of the different types of application files that can be opened in the list box.

Methods (*.FMSW) Choose this file type to open a Method file. Only the method is loaded. No data is loaded.

Data (*.FDSW) Choose this file type to open a Data file. Only the data is loaded. The method that was used to collect the data is not loaded. The last used method or default method will be displayed.

Report (*.FRSW) Choose this file type to open a Report file. You can view the report in the Report area.

Batch (*.FBSW) Choose this file type to open a Batch file. A Batch file includes data, the method used to collect the data, report and graphics setup.

Spreadsheet Ascii (*.CSV) This will open ASCII files that contain single comma delimited files with two or more columns.

All files (*.*) This enables you to view all files in the directory so that you can open any compatible file.

Tell me more about Filename Extensions

Overlay Data

Select this check box to overlay individual data files in one graph box. If this is not checked the data from each file will appear in individual graph boxes. This option is only available if the Open dialog has been accessed by selecting Open... from the File menu.

Hot Tip

Use the SHIFT or CTRL key to select multiple data files.

Open

This button opens the highlighted file/s or the files listed in the File name field.

See also:

File menu

Save As—Validate

Use the Save As dialog to save the data, method, report or graphics template individually or together as a Batch file.

Note

This dialog appears when you select either the Save method as... or Save as... menu option in the File menu. When you select Save method as... this dialog appears with 'Methods' displayed in the Files of type field. When you select Save as... this dialog appears with 'Batch' displayed in the Files of type field.

Traces (or data) that are currently 'Visible' in the Graphics area are saved, regardless of how they are displayed (full screen graph box or tile).

For example, if six graph boxes are displayed in the Graphics area and each graph box has one Visible
trace then all six traces will be saved.

If you do not wish to save particular traces, make them not Visible in the **Trace Preferences** dialog. To save only the focused trace, select the **Save only focused trace** check box.

The **Save As** dialog comprises the following:

- **Look In**
  The Look In field lists the available folders and files. Click the down arrow on the right of this field to view the location of the current folder in your computer's hierarchy. You can also select a new location simply by selecting a different folder. The folders and files in the selected folder are displayed in the list box below the Look In field.

  **Note**
  To open the folder in the level that is one higher than the current folder, select on the toolbar.

- **Look in list box**
  The Look In List Box, which is directly below the Look In field, displays the folders and files in the selected location. To view the contents of a folder, double-click it.

  - Click to create a new folder. Type the name of the new folder in the highlighted text entry field that appears and press ENTER. Then double-click on the new folder to open it.
  - Click to display the directory structure of the drive.
  - Click to display the files as a list.
  - Click to display files as a detailed list including the file size, file type and the date the file was last modified.
  - Click to preview the selected trace before you load it into the application.

- **File Name**
  Enter the name of the file you want to save in this field. You can also type the full path of a file then select the **Save** button to save the file.

- **Files of Type**
  Click on the arrow on the right of this field to view a list of the different types of application files that can be opened in the list box.

  - **Methods (*.FMVO)**
    Choose this file type to only save the current Method. Any collected data is not stored.
  
  - **Data (*.FDVO)**
    Choose this file type to only save the data currently Visible in the **Trace Preferences** dialog (for any graph box). Note that the method used to collect the data is not stored with the data.
  
  - **Report (*.FRVO)**
    Choose this file type to only save the contents currently shown in the Report area.
  
  - **Batch (*.FBVO)**
    Choose this file type to save the data that is currently Visible in the **Trace Preferences** dialog (for any graph box), the method used to collect the data and the currently displayed report all together as a Batch file. The graphics setup is also stored.
  
  - **Spreadsheet Ascii (*.CSV)**
    Choose this file type to save the raw intensity data currently Visible in the **Trace Preferences** dialog (for any graph box) as.
The Cary Eclipse system uses the filename extension to distinguish between data types (e.g. method or report) and also to differentiate applications. (e.g. Concentration or Kinetics).

The Syntax for file extensions is:

<table>
<thead>
<tr>
<th>Letter</th>
<th>Represents</th>
<th>Available Options</th>
</tr>
</thead>
<tbody>
<tr>
<td>First</td>
<td>Type of analysis</td>
<td>F Fluorescence</td>
</tr>
<tr>
<td>Second</td>
<td>Type of file</td>
<td>B Batch</td>
</tr>
<tr>
<td></td>
<td></td>
<td>D Data</td>
</tr>
<tr>
<td></td>
<td></td>
<td>G Graph Template</td>
</tr>
<tr>
<td></td>
<td></td>
<td>M Method</td>
</tr>
<tr>
<td></td>
<td></td>
<td>R Report</td>
</tr>
<tr>
<td>Third</td>
<td>Application identifier</td>
<td>AB Advanced Reads</td>
</tr>
<tr>
<td></td>
<td></td>
<td>CN Concentration</td>
</tr>
<tr>
<td></td>
<td></td>
<td>KN Kinetics</td>
</tr>
<tr>
<td></td>
<td></td>
<td>LT Lifetimes</td>
</tr>
<tr>
<td></td>
<td></td>
<td>RT Ratio</td>
</tr>
<tr>
<td></td>
<td></td>
<td>SR Simple Reads</td>
</tr>
<tr>
<td></td>
<td></td>
<td>SW Scan</td>
</tr>
<tr>
<td></td>
<td></td>
<td>TM Thermal</td>
</tr>
<tr>
<td></td>
<td></td>
<td>VO Validate</td>
</tr>
</tbody>
</table>

The following files may be opened and saved in the Validate Application:

- a comma delimited ASCII Spreadsheet file (*.CSV) file. The data is saved as XY pairs delimited by a comma.

**Note**

This file type can be easily imported into a spreadsheet program. For example, Microsoft Excel. You can also double-click on the stored *.CSV file from Microsoft Explorer to load Excel automatically with the ASCII XY data displayed.

**Tell me more about Filename Extensions**

**Save**

Click this button to save the file currently present in the File name field in the directory specified in the Look in field.

**See also:**

File menu

**Filename extensions—Validate**

The Cary Eclipse system uses the filename extension to distinguish between data types (e.g. method or report) and also to differentiate applications. (e.g. Concentration or Kinetics).

The following files may be opened and saved in the Validate Application:

**Note**

Selecting *All Files* in the Windows *Open* dialog will list all available file types.
How to combine data files into a batch file—Validate

To combine a number of data files into a batch file, do the following:

Step 1

Select **Open**... from the **File** menu to display the Windows **Open** dialog.

Step 2

A list of stored batch files will appear. Select the down arrow to the right of the **Files of type** field and select 'Data' to list all the data files.

Step 3

Select the **Overlay Data** check box.
Step 4
Highlight the data files that you require. Hold down the CTRL key to select multiple files.

Step 5
Select **Open**. The highlighted data files will load into the application and appear in the same graph box.

Step 6
Select **Save As…** from the **File** menu to display the Windows **Save As** dialog.

Step 7
A list of stored data files will appear. Select the down arrow at the right of the **Files of type** field and select 'Batch' to list all the batch files. Ensure that the **Save only focused trace** check box is cleared.

Step 8
In the **File name** field, type in the file name for your new batch file.

Step 9
Select **Save** to create the new batch file. The current method will be stored with the batch file.

All the data files are now combined into the one batch file.

Software and Screen features—Validate

- **Menu line**
- **Status display**
- **Status line**
- **Buttons**
- **Function keys**
- **Right mouse menu**
- **How to remove or display the command buttons**

Menu line—Validate

- **File menu**
- **Edit menu**
- **View menu**
- **Commands menu**
- **Setup menu**
- **Graph menu**
- **Calibration menu**
- **Help menu**

Status line—Validate
The Status Line is displayed at the very bottom of the Validate Application. The line will display the following information (from left to right):

- **Hardware Status**: This field displays status messages indicating the current instrument activity.
- **Sequence Information**: This field displays status messages indicating the current system activity.
- **Current Results**: This field displays information on the last reading.

**Buttons—Validate**

The major commands in this application can be accessed both from the menu items at the top of the main window and from the application buttons. These buttons, which appear in the left side of the Validate Application window, comprise the following:

**Hot Tip**
If you wish to access these commands from the main menus, rather than from these buttons, you can remove the Command buttons from your application window. To do this, clear the **Buttons** command in the View menu. This will enlarge the Graphics area, making viewing and selection of scans easier.

<table>
<thead>
<tr>
<th>Button</th>
<th>Enables you to…</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tests</td>
<td>Select this to display the Tests dialog where you can set up performance tests. You can use the dialog to configure your instrument to check its performance based on the regulatory specifications you select.</td>
</tr>
<tr>
<td>Setup</td>
<td>Click this button to display the Setup dialog where you can set up the report and email parameters.</td>
</tr>
<tr>
<td>Zero</td>
<td>Select this to zero the current ordinate value. This command can also be accessed by pressing F5. <strong>Note</strong>: Before you select Zero, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.</td>
</tr>
<tr>
<td>Clear Report</td>
<td>Select Clear Report to remove all of the text that appears in the Report area. If you have saved the report, you can view the information again. To do this, simply open the report file using the Open… command in the File menu.</td>
</tr>
<tr>
<td>Print</td>
<td>Select Print to display the Windows Print dialog where you can choose what printer the report will be printed to. You can also select what pages to print as well as the number of copies you wish to print. This command can also be accessed by pressing CTRL+P. <strong>Note</strong>: The options on the Reports page are used to generate the contents of the Report area during a run or on a recalculation. If Auto Print has been selected the Cary Eclipse will automatically print a report at the end of a data collection.</td>
</tr>
</tbody>
</table>

See also:
- Tests
Function keys—Validate

The function keys at the top of your keyboard perform basic Cary Eclipse operations simply and quickly.

<table>
<thead>
<tr>
<th>Function key</th>
<th>What it does</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>This displays the Help for the current dialog or window.</td>
</tr>
<tr>
<td>F4</td>
<td>Press F4 to display the Go to Wavelength dialog.</td>
</tr>
<tr>
<td>F5</td>
<td>Press F5 to perform an instrument Zero. The current intensity value will be reset to zero and all subsequent results will have the value that was reset to zero subtracted from them. You can use the Zero command before selecting Start if you want your data to start at 0 intensity. This command is also accessible from the Commands menu or by selecting the Zero button.</td>
</tr>
<tr>
<td></td>
<td><strong>Note</strong> Before you select Zero, ensure that you have placed the blank solution in the cell positions that are currently in the cell path.</td>
</tr>
<tr>
<td></td>
<td>Select the Clear Zero command from the Commands menu to remove zero correction.</td>
</tr>
<tr>
<td>F6</td>
<td>Press F6 to display the Rapid Result dialog where you can read a blank and multiple sample readings without changing the application method. This command is also accessible from the Commands menu.</td>
</tr>
<tr>
<td>F9</td>
<td>If the application is online, press F9 to start a data collection. This is equivalent to selecting the Start button. If the application is not online, you will need to press F9 to first connect to the Cary Eclipse. Then press F9 to start the data collection. This command is also accessible from the Commands menu or by selecting the Start button.</td>
</tr>
<tr>
<td></td>
<td><strong>Note</strong> You will not be able to perform a Connect if another application is collecting data.</td>
</tr>
<tr>
<td>F12</td>
<td>Select this to stop the current measurement. This is equivalent to selecting the Stop button. This command is also accessible from the Commands menu or by selecting the Stop button.</td>
</tr>
</tbody>
</table>

See also:
Go to Wavelength
Zero
Rapid Result
Start/Stop buttons

Right mouse menu—Validate

The following commands are accessible by clicking the right mouse button on editable fields in the Cary
Eclipse software.

<table>
<thead>
<tr>
<th>Command</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Undo</td>
<td>If available, this reverts to the previous value before the Edit on the field.</td>
</tr>
<tr>
<td>Cut</td>
<td>If available, this cuts the highlighted text to the clipboard.</td>
</tr>
<tr>
<td>Copy</td>
<td>If available, this copies the highlighted text to the clipboard.</td>
</tr>
<tr>
<td>Paste</td>
<td>If available, this pastes the text from the clipboard to the cursor position.</td>
</tr>
<tr>
<td>Delete</td>
<td>If available, this deletes the highlighted text/number.</td>
</tr>
<tr>
<td>Select All</td>
<td>If available, this selects all entries.</td>
</tr>
</tbody>
</table>

**How to remove or display the command buttons—Validate**

The command buttons (Tests, Setup, Zero, Clear Report and Print) appear on the left side of the Validate Application window.

**To remove the command buttons**

Step 1

From the Validate Application window, select the **View** menu.

Step 2

Clear the **Buttons** menu item.

The buttons will disappear from the application screen, giving you more room to display reports.

**To show the command buttons**

Step 1

From the Validate Application window, select the **View** menu.

Step 2

Select the **Buttons** menu item.

The buttons will re-appear in the application screen.

**ADL—Validate**

- [ADL Command Line](#)
- [ADL Hooks](#)
- [How to insert ADL commands](#)

**ADL Command Line—Validate**

**Introduction to ADL**
Use the **ADL Command Line** dialog to run Single-line or Multi-line Applications Development Language (ADL) commands that enable you to perform functions over and above that of the existing program functionality.

ADL is not a general purpose programming language. It has been developed specifically by Varian to meet the needs of spectroscopists. It can be used to handle all the data collected from the Cary Eclipse—both photometric values and entire data collections. ADL programs that you develop should be stored with the extension ‘ADL’.

To access the **ADL Command Line** dialog, select the **ADL Command** item from the **Commands** menu. You can also press ALT+Y to open the dialog.

**Note**
Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.

### Software controls

#### Single Line
Selecting this lets you enter ADL commands on a single line into the Command Line entry box. You can enter more than one ADL command on a single line simply by separating the commands by a semicolon ‘;’.

#### Multi-line
The Multi-line capability enlarges the Command Line entry box. Use the Multi-line capability to create a more complex ADL program by adding a series of commands over multiple lines. When Multi-line mode is selected, the Quick Reference option becomes available.

#### ADL Quick Reference
Select Quick Reference to open the Quick Reference dialog where you can easily add a command in the correct syntax to your ADL program. This option is only available for Multi-line commands.

Show me How to insert ADL commands using the Quick Reference

A detailed description of all the commands in the Quick Reference is available by selecting the Help button on the Quick Reference dialog.

**Browse**
Select **Browse** to access the Open dialog where you can browse for and select an existing ADL program to load and run.

**Command Line (entry box)**
This entry box enables you to use ADL commands. You can enter single ADL commands into this field, or you can enter a series of ADL commands. If you have the entry type set to Single Line, you can review previous ADL entries by selecting the down arrow to the right of the list box.

The ADL commands in the Command Line are executed when you select OK.

**Note**
On entering this window, the Cary Eclipse will recall the last command or program executed from the Command Line entry box. The ADL commands...
entered in the last executed program are typically stored in the Varian\Cary Eclipse WinFLR\ADL directory on the drive on which the Cary Eclipse software is installed.

**Hot Tip**
You can rerun the ADL commands in the Command Line entry box from any screen in the Validate application by pressing ALT-X or selecting Re-run ADL from the Commands menu.

ADL Hooks—Validate

The ADL Hooks command is accessible from the View menu.

Instead of writing a full ADL program you can use ADL hooks to link into an existing Cary Eclipse application. This is particularly useful if you want to make small changes to the way an application works, e.g. you want to pop up a dialog for the user to enter some specific data before the run starts. ADL hooks are links at specific points in an application at which you can run an ADL program. For example, ADL hooks have been supplied to run the following ADL programs:

**Note**
Full documentation on ADL programming is available in the ADL Online Help. Click the button below to view this Help.

<table>
<thead>
<tr>
<th>Hook</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>VO_Startup.ADL</td>
<td>This is run when the Validate application starts. For this to to work the application must be started from a batch or method file that has ADL Hooks on.</td>
</tr>
<tr>
<td>VO_Startup.ADL</td>
<td>This is run when the application is started. This command will only work if the application has been started from a batch or method file.</td>
</tr>
<tr>
<td>VO_PreSeq.ADL</td>
<td>This runs when the Start button is selected.</td>
</tr>
<tr>
<td>VO_PostSeq.ADL</td>
<td>This runs at the end of the Start button sequence.</td>
</tr>
<tr>
<td>VO_Online.ADL</td>
<td>This runs when Cary Eclipse is detected online.</td>
</tr>
<tr>
<td>VO_OffLine.ADL</td>
<td>This runs when Cary Eclipse goes offline.</td>
</tr>
<tr>
<td>VO_Zero.ADL</td>
<td>This runs when the Zero button is selected.</td>
</tr>
<tr>
<td>VO_Recalc.ADL</td>
<td>This runs when the Recalculate button is selected.</td>
</tr>
<tr>
<td>VO_Shutdown.ADL</td>
<td>This runs when the application is shutting down.</td>
</tr>
<tr>
<td>VO_PreRead.ADL</td>
<td>This runs just before a read is taken.</td>
</tr>
<tr>
<td>VO_PostRead.ADL</td>
<td>This runs just after a read is taken (It has the reading result stored in the UserResult variable.)</td>
</tr>
</tbody>
</table>
How to insert ADL commands using the Quick Reference—Validate

The Quick Reference window is accessed from the ADL Command window. It enables you to easily add a command in the correct syntax to your ADL program. To do this:

Step 1

Press Alt-Y to open the ADL Command Line window.

Step 2

Select the Multi-line option. The Command Line entry box will expand to take multiple lines of ADL commands and the Quick Reference check box is displayed.

Step 3

Select the Quick Reference check box. The Quick Reference window is displayed.

Step 4

If necessary, use the Go To field to jump to the appropriate part of the Quick Reference listing.

Step 5

Highlight the command you require in the Quick Reference list.

Step 6

Click the Insert in ADL button. The selected ADL command will appear in the Command Line entry box.

Step 7

Where necessary, alter the defaults supplied by the inserted Quick Reference command in the Command Line entry box to suit your program.

Step 8

Repeat steps 4–7 to add more commands as required.

See also:

ADL Command Line

Help and Safety information—Validate
Help menu—Validate

Use the Help menu to view the online Help and software information for this application.

Validate Help  Select this to display the Validate Home Page. This contains a list of online Help topics specific to the Validate Application.

Help Topics  Select this to display the Cary Eclipse Windows Online Help Home Page.

Tip of the day  View useful tips about using your Cary Eclipse instrument and software.

Eclipse Tour  Select this to display the Cary Eclipse Multimedia Online Help. This will give you an animated introduction to the Cary Eclipse software and hardware.

About…  Select About to access a dialog that displays information about the application as well as Varian email and Internet contact information.
To clear this About dialog, select OK.

Safety Home Page

Click on the desired index entry to view the Help.

- About Safety
- Lamp module
- Ultraviolet radiation
- Electrical hazards — Cary Eclipse
- Panels, covers and modules
- Other precautions
- Warnings and cautions
- Warning symbols
- Color coding
- Information symbols
- FCC advisory
- CE compliant products

Graphics How To

Graphics

- How to alter the look of your graphs
- How to make a graphics template

Traces

- How to select the focused trace
- How to track the focused trace
- How to display a trace that is not appearing in a graph box
- How to remove a trace from a graph
How to remove a trace from the trace list

Sizing and scaling

- How to resize graph boxes
- How to scale your graph manually
- How to scale your graph automatically
- How to zoom a graph

Labels

- How to edit a label
- How to move a label
- How to add a label to a graph
- How to delete a label

Pictures

- How to add a picture
- How to replace a picture
- How to move a picture
- How to delete a picture

How to edit a label—Validate

Step 1

Place the cursor on the label and click the left mouse button to select the label.

**Hot Tip**

If you are having trouble selecting the label it may be that a trace is interfering with the label selection. Try zooming in and clicking in a different part of the label.

Step 2

Click the right mouse button and select **Edit Annotation** from the drop-down menu. Alternatively you can select the **Edit Annotation** command from the **Graph** menu. The **Edit Label** dialog is displayed.

Step 3

Use the **Edit Label** dialog to change the label.

For example, if you want to change the font size of the label, select the **Font** button. In the **Size** field, scroll through the available sizes for the font and select the size you require. Alternatively, highlight the currently selected size in the **Size** entry field and enter the size you require. Click the **OK** button in this dialog to return to the **Edit Label** dialog.

Step 4
Click **OK** to clear the Edit Label dialog. Your label will appear with the new font size.

See also:
- How to add a label to a graph
- How to move a label
- How to delete a label
- How to resize a label

**How to track the focused trace—Validate**

To track the focused (red) trace in the graph:

**Step 1**

Click the **Cursor Modes** button from the Graphics toolbar, or select **Cursor Mode** from the **Graph** menu. The **Cursor Modes** dialog will appear.

**Step 2**

Select **Track** and select the **OK** button.

**Step 3**

Place the blue tracking cursor in the graph box you require.

**Step 4**

Move the cursor left and right across the graph box to trace the focused (red) scan. The cursor coordinates change (in the X, Y field at the bottom left hand corner of the Graphics area) as you move along the curve.

**Hot Tip**

If you wish to fix the cursor on a particular position on the curve, move the cursor to the required position. Press **ALT** on the keyboard, then release the mouse button.

See also:
- How to select the focused trace
- How to view a particular trace

**How to make a graphics template—Validate**

A graphics template lets you set up and store the style of a graphics report for future use. You can do this by adding labels and pictures to the background of the Graphics area and altering any axes styles etc. One advantage of this over the application's built-in reporting features is that you have more flexibility as to where you can position graph boxes and any text that you may want to appear (e.g. operator name, batch number etc.) and also where you add pictures, e.g. company logos etc.

**Step 1**

Start the application and open the data file from which you want to make a graphics template.
Alternatively, collect some new data.

Step 2

Make as many graph boxes as you require in your template by using the Add Graph function.

Step 3

Adjust the graph boxes to the correct size and position them as you want them to appear in the template.

Step 4

Do any of the following, if required:

- Change the appearance of the graph boxes in the Graph Preferences dialog
- Use the Add Label function to append text to the Graphics area
- Use the Add Picture function to add pictures, such as your company's logo to the Graphics area

Step 5

From the File menu, select Print Preview. Use the Print Preview dialog displayed to check the template that you have created.

Step 6

When you are satisfied with the appearance of the graphics, select the Save As… command from the File menu. The Save As dialog is displayed.

Step 7

A list of stored data files will appear. Select the down arrow at the right of the Files of type field and select 'Graph Template' to list all the template files. Ensure that the Save only focused trace check box is cleared.

Step 8

Enter a name for the template in the File name field.

Step 9

Select OK to save the graphic template. The template will be used each time you create a new file.

Note
You can also make a graphic template in a similar manner from existing data. No data will be saved in the template.

See also:
How to add a label to a graph
How to add a picture to a graph
How to move a label
How to move a picture

**How to resize graph boxes—Validate**

Step 1

Click on the graph that you want to resize so that the highlight box appears around it.

Step 2

Move the cursor to the edge of the highlight box and place it over one of the square marks (e.g. ■) in the box until it turns into a double headed arrow (↔).

Step 3

Click and hold the left mouse button and drag the box to the desired size.

Step 4

Release the left mouse button.

See also:

How to alter the look of your graphs

**How to zoom a graph—Validate**

To zoom a graph, do the following:

Step 1

Click on the button on the toolbar, or select **Cursor Mode** from the **Graph** menu. The **Cursor Modes** dialog will appear.

Step 2

Select **Free** and select the **OK** button.

Step 3

Click and hold down left mouse button in the highlighted graph box.

Step 4

Drag the mouse sideways and up or down. You will see a zoom box made up of a dotted line appear on the screen. (See picture below.)
The dotted line is a zoom box.

Hot Tip
To move the box on the screen, hold down the left and right mouse buttons together. You can then position the box anywhere in the graph box. Releasing the right mouse button will still allow you to resize the zoom box.

Step 5
Release the left mouse button. The area defined by the zoom box will be redrawn to fill the entire graph box, and the scans within the box will be enlarged accordingly. The abscissa (X) and ordinate (Y) coordinates displayed below the graph will also be changed to reflect the positions of the corners of the zoom box.

You can repeat this process if you need to zoom in even further.

Note
The system may limit the extent to which you can Zoom.

See also:
How to scale your graph manually
How to scale your graph automatically
Accessories

If you need information on installing, aligning or using a Cary Eclipse accessory, follow the appropriate link below. (If there is not a link for the accessory you are interested in either of these two sections, then you will need to refer to the operation manual shipped with the accessory):

Note
The Cary Eclipse Help contains many video clips. Links to these clips are marked by a video camera icon. Click on the icon to run the video.

Accessories supplied as standard with the Cary Eclipse

These accessories are supplied with your Cary Eclipse instrument. Click on each link to find out what their applications are and how to use them.

- Attenuator
- Diffuser
- Single Cell Standard and Cell Base

Accessories that are optional with the Cary Eclipse

You can buy these accessories from Varian as they are not shipped as part of the standard instrument package. Many of these accessories are application specific and may not applicable for all users. Click on each link to find out what their applications are and how to use them.

Thermal Accessories

- Ambient 4 Position Multicell Holder Accessory
- Cryostat (Optistat DN)
- Peltier 4 Position Multicell Holder Accessory
- Single Cell Holder (Peltier Thermostatted)
- Single Cell Holder (Water Thermostatted)
- Temperature Probes (Series II)
- Water 4 Position Multicell Holder Accessory
- Water Circulator (PCB-150 - for use with Single Cell - Water Thermostatted)

Polarization Accessories

- Automated Polarization Accessory
- Manual Polarization Accessory

Other Accessories

- Fast Filter Accessory
- Fiber Optic Coupler
- Rapid Mix Accessory (RX.2000)
- Solid Sample Holder Accessory
- Wellplate Reader
Attenuator and Diffuser

Table of Contents

- Introduction
  - The Attenuator
  - The Diffuser
- Specifications
- Getting Started
- Unpacking
- Installation
- Parts and Supplies

Introduction

The Attenuator

![Figure 1 - The Attenuator](image)

The attenuator is a rectangular piece of mesh that has 1.5 Abs attenuation. The attenuator is inserted into the filter slides and can be inserted on both the excitation and emission sides.

The attenuator is used with highly scattering samples.

The Diffuser
Figure 2 - The Diffuser

The diffuser is a piece of silica ground on both faces held in a triangular cuvette configuration. It provides a scattered signal which is independent of wavelength from red to UV.

The diffuser is mainly used in conjunction with the Validate software:

- samples producing a lot of stray light
- emission spectral bandwidths
- emission wavelength reproducibility
- emission wavelength accuracy

The diffuser can also be used to set up corrected emission spectra, the rhodamine sample (which can be purchased separately) must be used to setup the corrected excitation spectra beforehand.

Specifications

The Attenuator

- 1.5 Abs attenuation
- Can be used with both the excitation and emission slides

The Diffuser

- Fine grounding on both faces

Getting started

Unpacking

The attenuator and the diffuser are both shipped with the Cary Eclipse as standard.

Installation

The Attenuator

The attenuator can slide into both the excitation and the emission filter slides in the Cary Eclipse sample
compartment.

The Diffuser

The diffuser fits into the single cell holder that is also shipped as standard with the Cary Eclipse.

Parts and Supplies

<table>
<thead>
<tr>
<th>Descriptions</th>
<th>Part Number</th>
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<tr>
<td>Quartz silica diffuser</td>
<td>01 106748 00</td>
</tr>
<tr>
<td>Attenuator, 1.5Abs</td>
<td>01106775 00</td>
</tr>
</tbody>
</table>

Single Cell Standard and Cell Base

Table of Contents

- Introduction
- Getting Started
  - Unpacking
- Installation
- Parts and Supplies

Introduction

The standard single cell holder is shipped with the Cary Eclipse.
Although the standard cell holder does not have temperature control, it does have a number of useful features. There are height and tilt adjustments as well as a cuvette lifting feature which will ensure that you don't get any marks on your cuvettes.

**Getting started**

**Unpacking**

Unpack the accessory, and check that you have received the following:

- Cell Holder
- Hexagonal ball driver

**Installation**

Step 1

See how to perform steps 1 to 3

Make sure that the sample compartment is empty.

Step 2

Position the single cell base on the locating pins.

Step 3
Lock the cell base by turning the lever to the right.

Step 4

See how to perform steps 4 and 5

Place the single cell holder on the cell base so that the groove points to the left wall of the sample compartment.

Step 5

Secure the cell holder by tightening the spring loaded thumb screw.

For Cuvette Height Adjustment

Step 1

See how to perform height adjustment procedure

Turning the height adjustment screw in a clockwise direction will raise the cuvette.

Step 2

Turning the height adjustment screw in an anti-clockwise direction will lower the cuvette platform.

Note
To lower the cuvette you will have to push the cuvette down because of the tension created by the cell holder.

For Cuvette Pitch Adjustment

Step 1

See how to perform pitch adjustment procedure

Turning the pitch adjustment in an anti-clockwise direction will raise the front of the cell holder.

Step 2

Turning the pitch adjustment in a clockwise direction will lower the front of the cell holder.

Note
The pitch adjustment screw requires a hex key.

Parts and Supplies
Fast Filter Accessory

Table of Contents

- Safety information
- Introduction
- Unpacking notes
- Specifications
- Installation
- Maintenance and Cleaning

Safety information

Before using this accessory, you must read the Safety Practices and Hazards in your Cary Eclipse Hardware Manual.

A link to the Safety section is also available on the main page of the Cary Eclipse online Help and Videos CD.

If the equipment is used in a manner not specified by the manufacturer, your safety may be at risk.

Introduction

The Fast Filter accessory for the Varian Cary Eclipse Fluorescence spectrophotometer provides a solution for ratiometric applications. Such measurements require rapid sequential measurement of fluorescence intensity at two (or four) different excitation or emission wavelengths. The wavelengths selected are determined by filters, which are fitted into the accessory.

The accessory consists of four metal arms extending from a central wheel. Each arm is labeled as 1, 2, 3 or 4 by holes etched into the wheel, just below the filter position, (Figure 3a).

Either two or four bandpass filters may be mounted on the filter wheel (one filter per arm). If only two filters are used, they must be mounted in opposite positions, either positions 1 and 3 or positions 2 and 4. The solid black discs must be mounted in the two remaining filter positions. This is to ensure the filter wheel is balanced.

Unpacking notes

Your accessory is packed in foam-lined cardboard boxes. Check that you have received all of the items in the accessory kit by referring to the packing list included with the shipment (Figure 1).

Please follow the filter manufacturer's recommendation with regard to which side of the filter should face the light source. This is particularly important for excitation filters.
Installation

Installation of the Fast Filter accessory can be divided into three stages:

1. Installing the filters and the black discs in the accessory
2. Installing the accessory in the instrument
3. Configuring the accessory using the software

Installing the filters in the accessory

Step 1

Using your fingers, carefully unscrew the knurled nut (Figure 2 (i)) in the center of the black plastic filter locking wheel (Figure 2 (ii)) and remove the locking wheel from the accessory.

Figure 2. (i) Knurled nut  (ii) Filter locking wheel

Figure 1. (i) The Fast Filter accessory  (ii) 2 x black solid discs
**Step 2**

Filters now need to be placed in the accessory. There are four filter positions, each labeled 1 to 4 by holes etched into the arms of the filter holder, (Figure 3a).

**Figure 3a. Filter positioning**

All four filter positions MUST be used in the accessory to ensure the filter wheel is balanced. If two filters are to be used, they must be placed in opposite positions in the filter holder. If you place a filter in position 1, the second filter must be placed in position 3 and the black discs must be placed in positions 2 and 4. If you place filters in positions 2 or 4 the black discs must be placed in positions 1 and 3.

Lay the accessory horizontally, and carefully place the filters and solid black discs into the arms of the wheel, ensuring they are pressed against the arm ends, (Figure 3b).

Please follow the filter manufacturer's recommendation with regard to which side of the filter should face the light source. This is particularly important for excitation filters.
Figure 3b. (i) & (iii) Correctly fitted black discs (ii) & (v) Correctly fitted filters (iv) Replaced Filter locking wheel

Step 3

Replace the plastic locking wheel removed in Step 1, ensuring the bandpass filters and black discs remain positioned at the arm ends, (Figure 3b).

Replace the knurled nut and rotate in a clockwise direction until finger tight. DO NOT over tighten.

Note
Ensure you record the position in which each filter is mounted. The software requires this information at a later stage.

For example use the table shown below to record this information.

<table>
<thead>
<tr>
<th>FILTER CENTRE WL (nm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Position</td>
</tr>
<tr>
<td>1</td>
</tr>
<tr>
<td>2</td>
</tr>
<tr>
<td>3</td>
</tr>
<tr>
<td>4</td>
</tr>
</tbody>
</table>

Installing the accessory in the instrument

Step 1

Turn the instrument off and remove any accessories from the sample compartment.

Step 2

Prepare to remove the outer cover of the instrument by flipping up the four black plastic clasps located on each side of the instrument. Two of these are labeled (iv) and (v) in Figure 3.

Step 3

Remove the sample compartment front door and, using your thumb, lift the circular cover plate in the base of the instrument to expose a plastic clasp, (Figure 3 (ii)). Flip the clasp up.
Figure 3. (i) Circular cover plate (removed) (ii) Front clasp (iii) Sample compartment front door (removed) (iv) Right-hand side plastic clasps. The remaining two clasps are located in the same positions as (iv) and (v) on the opposite side of the instrument.

Step 4

Carefully remove the instrument outer cover by lifting it upwards away from the instrument base.

Step 5

This step varies slightly depending on whether the accessory is to be installed into the excitation or the emission fast filter positions.

With the instrument cover removed, the black metal inner cover is now visible. You will be able to see two plates each secured by two black screws to the top of the instrument. These represent the positions for either excitation or emission measurement. The black plate labeled (i) in Figure 4 covers the chamber in which the accessory needs to be fitted to operate as an excitation fast filter. Remove the appropriate black plate by unscrewing the black screws.
Installing the Fast Filter accessory in the Excitation position

To install the accessory in the Excitation position, hold the accessory above the excitation chamber, located to the right of the sample compartment.

Orientate the accessory so the male D-plug located on the accessory is aligned with the female D-plug (Figure 4 (vii)) inside the excitation chamber.

Orientate the filter wheel as shown in Figure 4 (v) so the arms of the filter wheel form a diagonal cross.

Carefully lower the accessory into the excitation chamber, raising the edge nearest the male D-plug on a 30° angle so that the opposite edge of the accessory touches the excitation chamber first (Figure 5 below). Once the edge is positioned, lower the male D-plug so that it is sitting on top of the female D-plug in the excitation chamber.

Figure 5. The edge nearest the male D-plug is raised at a 30° angle with the opposite edge touching the excitation chamber.

To secure the Fast Filter accessory gently push down on top of the accessory base directly above the two D-plugs. Once the plugs mate, tighten the black thumbscrews. The accessory is now fitted in the excitation position.

Installing the Fast Filter accessory in the Emission Position

To install the accessory in the Emission position, hold the accessory above the emission chamber, (Figure 4 (i)). Orientate the accessory so the male D-plug located on the accessory is aligned with the female D-plug (Figure 4 (vii)) inside the emission chamber.

Orientate the filter wheel as shown in Figure 4 (v) so the arms of the filter wheel form a diagonal cross.
Carefully lower both edges of the accessory into the emission chamber ensuring the male and female D-plugs are aligned. Once the accessory is sitting in the emission chamber gently push down on top of the accessory base plate directly above the two D-plugs. Once the plugs mate, tighten the black thumbscrews. The accessory is now fitted in the emission position.

Step 6

Secure the Fast Filter accessory in the chamber by tightening the two black thumbscrews (Figure 4 (iv)). The Fast Filter accessory is now correctly fitted and you can replace the instrument cover.

Step 7

Carefully replace the instrument cover and flip down the four black plastic clamps to secure, and the front black plastic clamp to secure them. Replace the circular cover. (This is the reverse of steps 4, 3 and 2 in order).

Step 8

Install the accessory you would like to use with your Cary Eclipse, for example the singe cell holder or multicell holder.

Step 9

If required, replace the sample compartment front door and close the sample compartment lid.

Note
If the Fast Filter accessory is removed it is important that the black cover plate be replaced and secured using the two black screws.

Configuring the accessory using the software

The Fast Filter accessory is designed to be operated by the Ratio Application provided in the Cary Eclipse Bio Software.

Step 1

Open the Ratio Application.

Step 2

Turn on the instrument.

Step 3

From the main Ratio window, select the Setup button to display the Setup dialog. From the Collection Type drop-down menu on the Cary page of the Setup dialog window, select either Excitation fast filter or Emission fast filter, depending on whether you have installed the Fast Filter accessory in the excitation or emission positions respectively (refer to Figure 6).
Figure 6. Instructing the instrument to drive the Fast Filter accessory fitted to the excitation monochromator chamber.

On the Accessories page of the Setup dialog, select the Fast Filter option to display the Fast Filter page (Figure 7). In this example the software will automatically calculate the result of Filter 3/Filter 1 and the result of Filter 4/Filter 2.

Figure 7. Instructing the Ratio software to drive the Fast Filter accessory.
Select the result 1 checkbox if you inserted bandpass filters into positions 1 and 3 of the filter wheel, or result 2 for positions 2 and 4. Alternatively select both checkboxes if you inserted filters in all 4 positions of the filter wheel.

**Step 4**

You are now ready to use the Ratio application to gather data. For more information about the Ratio or other Cary software applications, select the 'Help' button.

**Note**
The text fields in the lower half of the tab are for graph labeling purposes only and have no effect on the ratio values calculated.

**Accessory Removal**

If you need to remove the Fast Filter accessory from the instrument to add or change filters, grasp the accessory base on the right end of the long edge and lift gently. Once the D-plug is disconnected, the accessory may be carefully removed.

If the accessory is not being re-inserted, be sure to replace the black metal cover over the fast filter chamber, and tighten the locking screws.

**Specifications**

For environmental and power supply requirements, refer to your instrument operating instructions. This product is suitable for indoor use only.

**Weight**

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**Dimensions (W x D x H)**

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**Maintenance and Cleaning**

Keep your accessory safely packed in its original box when not in use. Avoid unnecessary handling. For maintenance and cleaning, please refer to the manufacturer's instructions.

**Technical Assistance**

For technical assistance regarding this accessory, please contact fluorescence@varianinc.com or look for other Varian contacts.

**See also:**

Automated Polarizer Instruction sheet
Fiber Optic Coupler Accessory

Table of Contents

- Safety information
- Introduction
- Unpacking notes
- Installation
- Alignment
- Operation
- Specifications
- Maintenance and Cleaning
- Technical assistance

Safety information

Before using this accessory, you must read the Safety Practices and Hazards in your Cary Eclipse Hardware Manual.

A link to the Safety section is also available on the main page of the Cary Eclipse online Help and Videos CD.

If the equipment is used in a manner not specified by the manufacturer, your safety may be at risk and the protection provided by the equipment may be impaired.

Introduction

The Fibre Optic Coupler is an accessory that enables the use of a fibre optic probe with the Cary Eclipse. This allows the user to examine the fluorescent properties of samples at locations remote from the instrument via an optical guide.

Unpacking notes

Your accessory is packed in foam-lined cardboard boxes. Check that you have received all of the items in the accessory kit by referring to the packing list included with the shipment.

Installation

To install the Fiber Optic Coupler in the sample compartment, follow these steps:

Step 1

Turn the Cary Eclipse instrument power off using the switch on the front right side of the instrument. You may leave the PC turned on.

Step 2

Remove the sample compartment lid by sliding it all the way back. You will feel some resistance, but keep pushing to remove it completely. Remove the front panel by sliding it upwards. Ensure that the sample compartment is empty.
Step 3

Carefully remove all of the packaging foam and accessory components from the box.

CAUTION: When handling the Fiber Optic Coupler, avoid placing fingers or thumbs in the coupler entrance, as they will touch the flat mirror. Always lift using the handle (Figure 1 (i)).

Figure 1. Fiber Optic Coupler accessory

(i) Handle
(ii) Holding screw (one of two)
(iii) Aligning adjuster (one of four)
(iv) Key hole (one of two)
(v) Focus control (one of two) - DO NOT ADJUST.
(vi) Stabilizing screw
(vii) Fastening lever in open position

Insert: Close up of the stabilizing screw.

Step 4

Ensure that the fastening lever (Figure 1 (vii)) is in the open position (i.e., to the left), and place the Fiber Optic Coupler in the sample compartment.
Align the two locating holes on the base of the accessory with the locating pins on the floor at the rear of the sample compartment. Move the fastening lever from left to right to lock the accessory into position.

**Step 5**

Adjust the stabilizing screw on the accessory base (Figure 1 (vi)), to stabilize the accessory. To do this, use your fingers to screw the top screw (Figure 1, insert) clockwise until there is no side-to-side movement, then tighten the locking nut to lock the accessory into this position.

**Step 6**

Attach the special sample compartment lid for use with the accessory onto the top of the instrument, by sliding it on from the back.

**Step 7**

To connect the long Fibre Optic Probe (part number 79 100430 00, see Figure 2) to the coupler accessory:

a. Carefully remove the protective plastic caps from the two connector ends (Figure 2 (ii)) of the probe.

b. Loosen the holding screws (Figure 1 (ii)) on the Fiber Optic accessory by turning them anticlockwise.

c. Swing open the small cover on the sample compartment lid. Support the end of the probe to which a probe tip will be attached (Figure 2 (iii)). While supporting this end, carefully thread the two connector ends of the probe and the cable plug (Figure 2 (ii) and (i) respectively) down through the hole.

d. Insert the two connector ends into the two key holes (Figure 1 (iv)) in the coupler accessory, ensuring that the metal pins are pointing directly upwards. (It does not matter which connector end is inserted into which slot.) Push the ends as far as they will go so that the face on the end of the connectors presses up against a metal shop.

e. Tighten the holding screws until finger-tight. Check that the probe connectors are securely held in place.
CAUTION: Fasteners should be finger-tight only. Do not over-tighten or you may damage the connectors.

f. Connect the cable plug to the D-shaped 25 pin port on the left side of the rear sample compartment wall.

g. Secure the probe in the sample compartment lid by tightening the thumb screw up against the metal join (Figure 2 (v) and 3 (i)). Then close the small cover (Figure 3 (ii)).

Figure 3. Securing the probe in the sample compartment lid.

h. Replace the front panel that you removed in step 2 and close the sample compartment lid.

i. Screw a probe tip onto the end of the probe (Figure 2 (ii)). You may use either a solid sample probe tip (part number 79 100432 00, Figure 4), or a liquid probe tip (10 mm or 20mm - both are available as a single kit, part number 99 101045 00, see Figure 4 (ii)).

Note
If you are using a liquid probe tip and are going to align the accessory, leave the tip off, as it needs to be off for alignment.
Figure 4. (i) Connection of a solid sample probe tip to the Fibre Optic Probe (ii) Connection of a liquid probe tip to the Fibre Optic Probe

Alignment

Warning
To prevent eye damage, avoid looking at the light beam.

To ensure that the fibre optic system will operate at maximum performance, it is necessary to optimize the efficiency with which light passes through the coupling device before experimentation begins.

Note
Alignment needs to be done only once—there is no need to align the coupler each time the accessory is removed and re-installed.

To align the accessory:

Step 1
Turn the Eclipse instrument power on using the switch on the front right side of the instrument.

Step 2
Start the Align application by clicking Start in the Windows Taskbar, then Programs/Cary Eclipse/Align.

Step 3
Any fluorescent material can be used for alignment. We recommend using the supplied white alignment card.

If using the solid sample tip, clamp the probe against a white area on the card/fluorescent material. The solid sample tip should be oriented as depicted in Figure 5.

Figure 5. Close up of the solid tip as it should be positioned against the white alignment card.

If using a liquid probe tip, ensure the probe is removed. Hold the end of the probe against the card/fluorescent material at an angle of approximately 45° to avoid scattered excitation light. Use a clamp to hold the probe in this position.

Step 4
In the Align application, click the Cary tab. If using the white card, set the parameters as depicted in Figure 6 then click Apply. If using a different fluorescent material, set appropriate excitation and emission wavelengths.

**Figure 6.** Align application Cary page settings for alignment using a white card.

**Step 5**

To maximize the "Int" (intensity) reading in the top left corner of the screen (hence the amount of light reaching the detector), you may need to turn the gold adjusters (Figure 1 (iii)). To do this:

a. Slide back the lid enough to remove the sample compartment front panel.

b. Using the supplied 5.5 mm spin-tight, slowly turn the vertical excitation adjuster to the left. If the intensity reading starts to decrease, turn the adjuster to the right.

c. Keep turning the adjuster in the direction that causes the intensity value to increase. When you reach a point where the value starts to decrease, you have just passed the point of maximum intensity. Turn the adjuster very slightly back the other way to find this maximum value.

d. Repeat steps (b) and (c) for the horizontal excitation adjuster to maximize the signal. Then, repeat steps (b) through (d) until turning either adjuster in any direction results in a decrease in intensity.

**CAUTION:** Do not alter any other screws on the accessory, especially the two focus controls (Figure 1 (v)).

e. Now that you have optimized the light entering the excitation fibers, you will need to maximize the

---

**Tip**

You may find it easier to view the intensity reading graphically. To do this, click the Graph tab. Zoom in on the graph for a more precise view of the intensity.

d. Repeat steps (b) and (c) for the horizontal excitation adjuster to maximize the signal. Then, repeat steps (b) through (d) until turning either adjuster in any direction results in a decrease in intensity.
fluorescence emission reaching the detector. To do this, repeat steps (b) to (d) for the emission vertical and horizontal gold adjusters.

The instrument is over-range if the reading reaches 1000 a.u. If this occurs, it will be necessary to reduce the photo-multiplier tube (PMT) voltage on the Cary page in order to continue the alignment procedure. As a guide, a 10% change in PMT voltage causes a two-fold change in the intensity measured.

**Step 6**

Once you have maximized the intensity using the adjusters, the coupler is satisfactorily aligned. Replace the front panel, close the lid and close the Align application.

If using the liquid probe tip, you can now screw the tip onto the end of the probe (Figure 4 (ii)).

**Operation**

**To use the Fibre Optic Coupler:**

**Step 1**

Start the application you will be using and set the instrument parameters according to your analytical requirements.

**Step 2**

Present the probe to the sample and press the red Remote Read button (Figure 2 (iv)) on the probe to start the data collection.

**Specifications**

For environmental and power supply requirements, refer to your Eclipse operating instructions. This product is suitable for indoor use only.

**Weight**

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**Dimensions (W x D x H)**

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<td>350 x 210 x 50 mm</td>
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<td></td>
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<td>6.3 x 6.3 x 6.3 in</td>
<td>13.8 x 8.3 x 2.0 in</td>
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**Maintenance and Cleaning**

The outside of the Fibre Optic Coupler should be kept clean by wiping with a damp cloth (dampened with water or alcohol). Do not use abrasive cleaning agents.

Rinse the probe tips and the end of the probe with water after use and dry before storing. Avoid bending the cable sharply as this can damage the cable.
Technical Assistance

For technical assistance regarding this accessory, please contact:

Varian Optical Spectroscopy Instruments,
P.O. Box 222,
Clayton South, Victoria, Australia, 3169

or email: fluorescence@varianinc.com

Alternatively please see the contacts page for your local Varian office.

Introduction

The Rapid Mix accessory is a stop flow accessory that allows the user to start measuring reagents as soon as they have been mixed. This is an obvious advantage for reactions which are short-lived.
The Rapid Mix (RX.2000) accessory

The syringes are mounted on a rigid drive platform ensuring that the flow is stopped precisely and instantaneously. This platform and the syringe mounting blocks are extremely resistant to chemical attack.

Reagents travel in this inert sample circuit through an umbilical cord to the flow cell where they are mixed by a high efficiency T-format mixer.

Specifications

<table>
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<td>Window Sizes</td>
<td>30mm and 6mm for fluorescence detection</td>
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<td>Cell material</td>
<td>Spectrosil</td>
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<tr>
<td>Circular Dichroism</td>
<td>Triple-annealed observation cell eliminating birefringence</td>
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<tr>
<td>Beam Height</td>
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<td>Min. Vol./Shot/Reagent</td>
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<td>Prime Volume</td>
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<tr>
<td>Syringe Volume</td>
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<tr>
<td>Ratio mixing</td>
<td>Up to 25:1 by using different size syringes</td>
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<td>Temperature Range</td>
<td>0 - 60°C displayed to within 0.1°C with high and low adjustable alarms</td>
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<tr>
<td>Triggering</td>
<td>Anti-bounce, TTL, open-collector and switch-contact</td>
</tr>
<tr>
<td>Flow Circuit</td>
<td>Biocompatible and chemically inert</td>
</tr>
<tr>
<td>Construction</td>
<td>Constructed from chemically resistant materials throughout</td>
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<tr>
<td>Anaerobic Facilities</td>
<td>An anaerobic accessory (AA.1) is available to facilitate the rigorous exclusion of oxygen</td>
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<tr>
<td>Drive System</td>
<td>Manually or pneumatically driven (optional accessory DA.1)</td>
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</table>

Installation

See how to perform steps 1 to 5

Step 1

Place the Rapid Mix Accessory on top of the Cary Eclipse instrument with the syringe end of the accessory at the front.

Step 2

Insert the Rapid Mix Accessory cuvette into either the single cell standard, single cell peltier or single cell water thermostatted accessories (the single cell standard is shown in video).

Step 3

Connect the 5-pin DIN connector of the cable (supplied with the accessory) to the rapid mix accessory.

Step 4
Connect the 25-pin end of the cable to the 25-pin connection at the front of the Cary Eclipse.

Step 5

Cover the sample compartment with a black cloth to ensure that no light can enter the sample compartment when measuring samples.

**Operation**

For instructions on how to use the Rapid Mix Accessory (RX.2000), refer to the documentation supplied with the accessory.

**Parts and Supplies**

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<th>Descriptions</th>
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<td>79 100410 00</td>
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<tr>
<td>Pneumatic Drive For RX2000</td>
<td>79 100411 00</td>
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**Solid Sample Holder Accessory**

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- [Safety information](#)
- [Introduction](#)
- [Unpacking notes](#)
- [Installation](#)
- [Operation](#)
- [Specifications](#)
- [Maintenance and Cleaning](#)

**Safety information**

Before using this accessory, you must read the Safety Practices and Hazards in your Cary Eclipse Hardware Manual.

A link to the [Safety section](#) is also available on the main page of the Cary Eclipse online Help and Videos CD.

If the equipment is used in a manner not specified by the manufacturer, your safety may be at risk and the protection provided by the equipment may be impaired.

**Introduction**

The Solid Sample Holder for the Cary Eclipse is an accessory that enables you to perform fluorescence measurements on solid samples.

It consists of a vertical sample mounting "slide" supported by optical rails attached to the accessory baseplate.

Three optional kits are available for mounting specific types of solid sample. They are:

- Edge mounting kit (part number 99 101029 00)
• Powder holder and single crystal holding kit (part number 99 101030 00)
• Cuvette holder kit (part number 9 101031 00)

Unpacking notes

Your accessory is packed in foam-lined cardboard boxes. Check that you have received all of the components listed in the packing list included with the shipment.

Installation

If you ordered the Solid Sample Holder without any of the additional (optional) mounting kits, follow the standard installation procedure.

If you ordered one of the optional kits, follow the installation procedure that corresponds to your kit.

• Edge Mounting kit installation
• Powder holder and single crystal holding kit installation.
• Cuvette holder kit installation

Standard installation

Step 1

Attach the slide to the accessory base (Figure 1). To do this:

a. Completely remove the lower half of the clamping mechanism by taking out the screw that holds the two parts together (Figure 2 (ii)).
**Figure 2.** showing the screw (ii) separating the lower half of the clamping mechanism (i), and the groove (iii) where the optical rail fits.

b. Slide the lower half of the clamp under the two optical rails on the accessory base. Orient it so that the cut-out edge is facing the angular adjustment locking screw (Figure 3 (iii)).

**Figure 3.** showing (i) how to position the lower half of the clamp (outlined in white) under the optical rails (ii) on the accessory base. Locating holes (iv), Varian label (v) and fastening lever (vi) are also labeled.

c. Re-attach the upper half of the clamp mechanism to the lower half by placing the locating pin into the hole in the centre of the lower piece, and inserting and tightening the screw that you removed in step (a). The face of the slide should point towards the Varian label (Figure 3 (v)) on the base of the accessory.

**Step 2**

Use the rods, clamps and screws contained in the remaining small box (part number 99 101037 00), in combination with the tapped holes in the slide face, to mount your sample.

**Step 3**

You can now place your assembled accessory and sample into the sample compartment. To do this:

a. Ensure that the Eclipse sample chamber is empty.

b. Ensure that the Solid Sample Holder baseplate fastening lever (Figure 3 (vi)) is in the open position (that is, pointing towards the white scale marks).

c. Align the two locating holes on the base of the accessory (Figure 3 (iv)) with the locating pins on the floor at the rear of the sample compartment.

d. Move the fastening lever from left to right to lock the accessory into position.
Edge Mounting kit installation

If you ordered the optional Edge mounting kit (part number 99 101029 00), follow the installation procedure below.

**Step 1**

Follow step 1 of the standard installation procedure.

**Step 2**

Remove the standard slide by loosening the two screws at the top of the slide holder and sliding the face all the way across.

**Step 3**

Slide the edge mounting slide face onto the top of the slide holder. The edge mounting pieces attached to the slide should be facing the Varian label (Figure 3 (v)) on the accessory base, and the white grading marks should be at the top.

For larger samples, unscrew the two bottom edge mounting pieces using the supplied hexagonal ball screwdriver and remove the top edge mounting piece by unscrewing the thumb screw. Replace these with the supplied large edge mounting pieces. (The piece with the two small guiding pins is the one that is positioned in the center of the slide face. The pins allow it to slot into the tracks.

**Step 4**

Slot your sample into the "V" created by the bottom two edge mounting pieces. Then loosen the screw that holds the central edge mounting piece in place and slide this piece down to secure the sample.

**Step 5**

Follow step 3 of the standard installation procedure.

Powder holder and single crystal holding kit installation.

If you ordered the optional Powder holder and single crystal holder and single crystal holding kit (part number 99 101030 00), follow the appropriate installation procedure below.

*To install the single crystal holder:*

**Step 1**

Follow step 1 of the standard installation procedure.

**Step 2**

Attach the crystal holder (Figure 4) to the slide face using the two screws provided. The three-holder side should form a "shell". Place your crystal into the hole that most securely holds your crystal.
Figure 4. The single crystal holder:

Step 3

Follow step 3 of the standard installation procedure.

To install the powder holder:

Step 1

Follow step 1 of the standard installation procedure

Step 2

Attach the rectangular plate with a semi-circle cut out of it to the face of the slide. Do this using the four screws and the hexagonal ball screwdriver. Ensure the flat side of the plate is facing away from the slide.

Step 3

Position the powder receptacle (Figure 5 (iii)) with the small silver rod pointing upwards.

Step 4

Using gloves or lens tissue, carefully place a silica disc (Figure 5 (ii)) into the powder holder receptacle. Beware of contaminating the disc with fingerprints.

Note

The kit comes with a spare silica disc and a spare powder holder receptacle.

Step 5

Put some sample into the powder receptacle, ensuring a thin layer is covering the silica.

Step 6

Place the powder receptacle into the larger powder holder "shell" (Figure 5 (iv)). To do this:

a. Grasp the small silver rod of the powder receptacle between the thumb and forefinger.

b. Lift the powder receptacle, keeping it horizontal at all times to avoid spilling the powder.
c. Place the receptacle into the centre of the shell cavity.

Step 7

Ensuring that the protruding black metal column (Figure 5 (ν)) is on the inside, (in order to securely press the sample up against the silica disc), screw the lid (Figure 5 (ι)) of the powder holder onto the shell.

Figure 5. Exploded view of the powder holder. The sample sits between the silica disc (ιι) and the column of the lid (ν).

Step 8

Take the assembled powder holder and slide the rim (Figure 5 (νι)) into the semi-circle groove on the accessory slide face created in step 2.

Step 9

Follow step 3 of the standard installation procedure.

Note

When disassembling the powder holder, ensure that the silica disc is facing down when you unscrew the lid, to prevent spillage of powder.

Cuvette holder kit installation

If you ordered the optional Cuvette holder kit (part number 99 101031 00), follow the installation procedure below.

Step 1
Follow step 1 of the standard installation procedure.

Step 2

Attach the cell holder to the slide by aligning the two holes on the cell holder with the two holes at the bottom of the slide and using the hexagonal ball screwdriver to screw in two screws. Ensure that the open side of the cell holder is facing outwards.

Step 3

Insert the cuvette containing the sample

Step 4

Follow step 5 of the standard installation procedure.

**Operation**

The solid sample holder accessory provides both rotational and translational adjustment of the sample. The angle of incidence of the excitation may be varied from 20–35°. This is the angle between the exciting light and a line perpendicular to the surface of the sample mounting slide (Figures 6 and 7). The angle of incidence is indicated by the position of the corner of the "stage" relative to the graduated markings on the accessory baseplate (Figure 6). The markings are at intervals of one degree. The largest mark indicates an angle of incidence of 30°.

**Figure 6.** a) Angular adjustment locking screw b) Corner of stage. Markings indicate the angle of incidence of the exciting light in the sample mounting slide.

**Figure 7.** Angle of incidence of excitation. a) Normal b) Angle of incidence c) Excitation d) Slide
To alter the angle of incidence:

**Step 1**

Loosen the angular adjustment thumbscrew (large thumbscrew).

**Step 2**

Rotate the stage to the desired orientation.

**Step 3**

Lock the stage into position by rotating the thumbscrew until finger-tight.

It is recommended that you use the Kinetics application to perform this procedure. Alternatively, you can use the Advanced Reads application.

To optimize measurement of a particular sample with the solid sample holder accessory, it is recommended that you initially monitor both the fluorescence intensity and the signal intensity at the excitation wavelength. Rotate the accessory "stage" (Figure 6 (b)) to alter the angle of incidence of the exciting light.

Samples of varying depth may be moved to the focal position by loosening the thumbscrew that tightens the clamps around the optical rails, and sliding the entire mount forward or backward as required. This ensures the exciting light interacts with the sample directly in front of the emission window thus enhancing the collection of emission.

The contribution to the total signal from scattered exciting light is inherently greater when measuring opaque solids as compared to clear (non-scattering) liquids measured at the traditional right angle orientation. For this reason, it is highly recommended that excitation and emission filters be used. This can be achieved using the Eclipse software by setting both the "Excitation filter" and "Emission filter" selections on the Options tab of Setup to AUTO. If required, further reduction of scattered excitation could be achieved by placing appropriate cut-off and/or bandpass filters in the filter holders provided in the instrument sample chamber.

### Specifications

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</table>
Wellplate Reader

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- Introduction
- Installation
- Alignment
- Operation
- Parts and Supplies

Introduction

The Cary Eclipse Wellplate reader is an extremely versatile accessory and performs as efficiently as part of the Cary Eclipse instrument as many independent wellplate readers do on their own.

The Cary Eclipse Wellplate reader is capable of handling up to 384 well plates but can also handle custom well plates. The Wellplate Reader is computer controlled in the X-Y planes with mechanical adjustment in the Z plane.

Figure 1 - The Wellplate Reader

Installation

Step 1

See how to perform steps 1 to 4
Make sure that the sample compartment is empty.

Step 2

Position the wellplate reader on the locating pins at the rear of the sample compartment.

Step 3

Turn the fastening lever to the left.

Step 4

Connect the accessory cable to the 15-pin port which is located on the left wall of the sample compartment.

Step 5

See how to perform steps 5 to 8

Install the top cover for the Wellplate Reader by sliding it along the rails at the top of the sample compartment and tighten the knob to secure the cover. Leave the sample compartment open.

Step 6

Loosen the extended section of the sample compartment (which is attached to the Wellplate Reader). This is achieved by loosening the thumbscrews underneath the front of the accessory.

Step 7

Close the hinged section of the extended cover and lift the section attached to the accessory so that it fits neatly into the hinged section.

Step 8

Tighten the thumbscrews underneath the front of the accessory to hold the extended section of the sample compartment permanently in place.

Alignment

The top left, top right and bottom right well positions of a wellplate are used for the purposes of alignment. For the purposes of this example, a 96 wellplate and a 384 wellplate will be used. Thus the A1, A12 and H12 positions (96) or A1, A24 and P24 positions (384) are used for alignment.

The Wellplate Reader should be aligned whenever you change brands or types of wellplates. Manufacturers of wellplates have varying dimensions between wells and the dimensions also change when you swap from a 96 well to a 384 well plate. You cannot move the Wellplate Reader out of alignment by manually pushing the wellplate platform—it remembers alignment coordinates and will reset itself at the start of a run.

To align a wellplate:

Step 1
From the Align Application window, select the Wellplate (Wellplate) tab to access the Wellplate (Wellplate) page.

Step 2

From the Plate type field, select the Plate type you require. Alternatively, you can create or modify a plate using the Plate format dialog.

How to create or modify a wellplate format

Step 3

Place a fluorescing solution in Wells A1, A12 and H12. Fill the well to at least three-quarters (3/4) full.

Step 4

Select the A1 well position on the Wellplate Reader diagram to drive the Wellplate reader to this well. To do this, simply click in the well. The well will change color, to indicate that it is selected (as shown below).

Step 5

Select the Cary tab to display the Cary page. On this page, do the following:

a. Set excitation and emission wavelength parameters that are relevant for the solution you are using.

   Tip: To minimize collecting scattered excitation light, you should ensure that the excitation and emission wavelengths are at least the sum of the excitation and emission slit widths. For example, if you are an Excitation slit of 5 nm and an Emission slit of 10 nm, set the emission wavelength at least 15 nm greater than the Excitation wavelength.

b. Set the Emission filter option to Auto.

c. Set the Excitation filter option to Auto.

d. Adjust the PMT Voltage as required, ensuring that the intensity signal does not go over-range.

e. Select Apply to update the instrument parameters.

Step 6

Select the Wellplate (Wellplate) tab to display the Wellplate (Wellplate) page. On this page, do the following:

a. Select Auto-align to commence the auto-align procedure that will align the wellplate.

   Note: If the signal over-ranges during the auto-align procedure, decrease the PMT Voltage value on the page and then re-perform the Auto-align.
b. Once the alignment procedure is completed correctly, select the A1 well position on the Wellplate Reader diagram to drive the Wellplate reader to this well.

Step 7

Select the Cary tab to display the Cary page. On this page, do the following:

a. Set the excitation wavelength to visible light (for example, 540 nm).

b. Select Apply.

Step 8

Visually determine whether the image of the beam is within the well.

**If the beam is within the well**, do the following:

a. Select the Wellplate (Wellplate) tab to display the Wellplate (Wellplate) page.

b. Select Save Plate to save the current settings.

The wellplate is now aligned and ready for use. However, if you would like to obtain the optimum performance from your wellplate reader, go to **Step 9**. Otherwise, go to **Step 11**.

**If the beam is NOT within the well**, (this may occur for 384 wellplates) do the following:

a. Select the Wellplate (Wellplate) tab to display the Wellplate (Wellplate) page.

b. Use the Arrow buttons to move the plate so that the beam image is within the well and does not clip the sides.

c. When you are satisfied with the visual alignment, select Save A1.

d. Select the A12 (96) or A24 (384) well position on the Wellplate Reader diagram to drive the Wellplate reader to this well. Confirm that the image is within this well. If the image is not within the well, drive back to well A1, re-adjust the beam image and re-save the A1 setting.

e. Select H12 (96) or P24 (384) well position on the Wellplate Reader diagram to drive the Wellplate reader to this well. Confirm that the image is within this well. If the image is not within the well, drive back to well A1, re-adjust the beam image and re-save the A1 setting.

f. Once you are satisfied that the beam is image is within all well positions, select Save Plate to save the current settings.

The wellplate is now aligned and ready for use. However, if you would like to obtain the optimum performance from your wellplate reader, go to **Step 9**. Otherwise, go to **Step 11**.

Step 9

**Note:**

To obtain the optimum performance from your wellplate reader, you can perform the following steps (9 and 10). However, these steps are OPTIONAL. If you do not wish to perform these steps, go to **Step 11**.

a. Select the Cary tab to display the Cary page. On this page, set the Ave. time to 0.1 s.

2. Select the Wellplate (Wellplate) tab to display the Wellplate (Wellplate) page. On this page, select the A1 well position on the Wellplate Reader diagram to drive the Wellplate reader to this well.

3. Select the Graph tab to display the Graph page. You will need to monitor the signal intensity on the
graph while you are performing the adjustment procedure outlined below. Your aim is to maximize the intensity.

**Hot Tip**
You may need to change the graph scale when performing the alignment. To do this, select from the Graph toolbar and enter the required scale. Alternatively, select to automatically scale the axes.

**Step 10**

You need to adjust the emission optics controls, which are located on the right side of the Wellplate Accessory (when you are looking at it). Using the spanner (part number 72 100153 00) provided in the Wellplate Accessory kit, adjust the vertical and horizontal alignment screws (refer to Figure 2, below).

Monitor the intensity signal on the Graph page, and continue to adjust the alignment screws until you can reach a maximum intensity.

**Caution**
Do not adjust any other screws as this may change the focal point of the accessories. Only adjust the vertical and horizontal emission alignment screws.

The Wellplate Reader alignment is now fully optimized.

![Figure 2 - The emission alignment screws](image)
1. Vertical alignment screw  2. Horizontal alignment screw

**Step 11**

From the File menu, select Exit to close the Align application. You can now select the aligned plate in all Cary Eclipse applications that use a Wellplate accessory.

You can use this alignment configuration to read all plates of the same type and volume. The plate will not have to be auto-aligned again as long as the same plate type/brand is used. However, if the volume is
varied and the height of the plate changed, by either adding or removing the spacer, the signal should be maximized for optimum performance.

**Operation**

Refer to the Kinetics, Scan and Advanced Reads software help sections for specific operational details.

**Parts and Supplies**

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<th>Descriptions</th>
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**Ambient 4 Position Multicell Holder Accessory**

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- Safety information
- Introduction
- Unpacking notes
- Installation
- Operation
- Specifications
- Maintenance and Cleaning

**Safety information**

Before using this accessory, you must read the Safety Practices and Hazards in your Cary Eclipse Hardware Manual.

A link to the Safety section is also available on the main page of the Cary Eclipse online Help and Videos CD.

If the equipment is used in a manner not specified by the manufacturer, your safety may be at risk and the protection provided by the equipment may be impaired.

**Introduction**
The Ambient 4 Position Multicell holder for the Cary Eclipse is an accessory that permits fluorescence measurements on up to four samples at a time.

**Unpacking notes**

Make sure that all of the adhesive tape, plastic and cardboard is removed before operating the Ambient Multicell Holder.

**Installation**

**Step 1**

Turn the Cary Eclipse instrument power off using the switch on the front right side of the instrument. You may leave the PC turned on.

**Step 2**

Slide the top panel of the sample compartment all the way back and completely remove the front panel by sliding it upwards. Ensure that the sample compartment is empty.

**Step 3**

Lift the Multicell Holder by the handle, holding the end of the cable in your other hand. Reset the accessory at the front of the sample compartment in order to allow access to the D-shaped, 25 pin port at the left side of the rear sample compartment wall. Connect the cable plug to the 25 pin port.

**Step 4**

Ensure that the fastening lever is in the open position (i.e., to the left). Align the two holes on the base of the accessory with the locating pins on the floor sample compartment. Move the fastening lever from left to right to lock the accessory into position.

**Step 5**

Replace the front panel ensuring the accessory cable is tucked neatly into the sample compartment. The accessory is now ready for use.

**Note**

To remove the accessory, turn the instrument off and then follow the above procedure in reverse.

**Operation**

Operation of the accessory is through the Cary Eclipse software.

**To use the multicell holder:**

**Step 1**

From the Setup dialog, select the Accessories tab to access the accessories page.
Select the Multicell holder option and from the cell diagram, select the number of cells to use.

Step 3

To display the temperature of items in the Status Display window, select these from the Temperature display option.

Step 4

Select OK to exit the Setup window.

Step 5

Select Start to begin data collection.

Specifications

For environmental and power supply requirements, refer to your Cary Eclipse Hardware Operation manual (part number 85 101758 00). This product is suitable for indoor use only.

Connections

- 25-pin D-range connector on flying lead

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Maintenance and Cleaning

All cleaning should be done with a soft cloth. If necessary, this cloth can be dampened with water or a mild detergent to clean black painted surfaces. Do not use organic solvents or abrasive cleaning agents.

See also:

Cryostat - Variable Temperature Liquid Nitrogen
Optistat DN

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**Introduction**

The Oxford Instruments "Optistat DN", a variable temperature liquid nitrogen cryostat, is designed for variable temperature (77-320°K, +/- 0.1°K) optical spectroscopy measurements and electrical measurements requiring optical access. Varian provides a mount and covers for the Optistat DN so that it may be used in the Cary Eclipse instrument.

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**Optistat**<sup>DN</sup>

**Figure 1** - The Optistat DN Cryostat and its measurements

---

**Specifications**
Further specifications for the Optistat DN Cryostat can be found in the manual supplied with the cryostat.

**Installation**

Step 1

Make sure that the sample compartment is empty.

Step 2

See how to perform steps 2 to 4

Position the cryostat holder on the locating pins in the Cary Eclipse sample compartment (the fastening lever should be positioned closest to the front of the instrument).

Step 3

Turn the fastening level of the cryostat holder to the right. This will lock the base plate into position.

Step 4

Place the O-ring (supplied with the cryostat holder) around the cryostat.

Step 5

See how to perform steps 5 to 8

Insert the rear section of of the cryostat sample compartment lid from the back of the instrument by sliding it along the rails at the top of the sample compartment.

Step 6

Position the Cryostat in the cryostat holder and tighten the cryostat holder thumb screws so that they are finger tight. Do not over-tighten.

---

**Sample space (mm)**

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**High temperature with OPTIHT option**

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**Temperature stability (K)**

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**Hold time at 77 K (hr)**

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**Sample change time (min)**

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**Weight (kg)**

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*Note*: For operation between 320 and 500 K, high temperature sapphire windows are required on the inner window ports.
Step 7

Make adjustments to the O-ring so that it fits into the groove of the rear section of the sample compartment lid.

Step 8

Slide the rear section of the lid so that it sits right against the cryostat and tighten the thumbscrew.

Step 9

*See how to perform steps 9 and 10*

Insert the front section of the sample compartment lid. Position the O-ring so that it fits neatly into the groove of the front section of the sample compartment lid.

Step 10

Insert the front sample compartment panel.

**Operation**

Operation instructions for the Optistat DN Cryostat can be found in the manual supplied with the instrument.

**Parts and Supplies**

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**Peltier 4 Position Multicell Holder Accessory**

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**Safety information**

Before using this accessory, you must read the Safety Practices and Hazards in your Cary Eclipse Hardware Manual.
A link to the Safety section is also available on the main page of the Cary Eclipse online Help and Videos CD.

If the equipment is used in a manner not specified by the manufacturer, your safety may be at risk and the protection provided by the equipment may be impaired.

**Introduction**

The Peltier-thermostatted 4 Position Multicell Holder for the Cary Eclipse is an accessory that permits the fluorescence measurements on up to four samples, while precisely controlling the temperature and rate of stirring.

**Unpacking notes**

Make sure that all of the adhesive tape, plastic and cardboard is removed before operating the Peltier Multicell Holder.

**Installation**

**Step 1**

Turn the Cary Eclipse instrument power off using the switch on the front right side of the instrument. You may leave the PC turned on.

**Step 2**

Slide the top panel of the sample compartment all the way back and completely remove the front panel by sliding it upwards. Ensure that the sample compartment is empty.

**Step 3**

Lift the Multicell Holder by the handle and hold the end of the hose in your other hand. Reset the accessory at the front of the sample compartment in order to allow access to the D-shaped, 25 pin port at the left side of the rear sample compartment wall. Connect the cable plug to the 25 pin port.

**Step 4**

Ensure that the fastening lever is in the open position (i.e., to the left). Hold the extended nose (the black metal flange which contains the stirring controls, see Figure 1) and align the two holes on the base of the accessory with the locating pins on the floor at the rear of the sample compartment. Move the fastening lever from left to right to lock the accessory into position.
Figure 1. Peltier 4 Position Multicell Holder accessory

Step 5

Ensure the two black locking latches at the back of the extended nose are pointing upwards (unlocked, see Figure 1). Position the extended nose into place by aligning its locating pins with the holes in the front of the sample compartment floor. Lock the extended nose into position by pushing the locking latches downwards.

Step 6

Slide the special door for thermostatted accessories (part number 99 101032 00) into position ensuring the electrical cable is tucked neatly into the sample compartment.

Step 7

Connect the hose ends to the Temperature Controller inlet and outlet connections.

Note
The Peltier Multicell Holder must be used only with a Varian Temperature Controller (part number 00 100386 00).

Step 8

Connect the circular communications plug to the 14 pin socket on the Temperature Controller.

Note
You may need to rotate the communications plug before it will slot into the communications socket, as it has been designed to be inserted in one position only.

Step 9
Ensure the Temperature Controller has sufficient liquid for optimal operation.

![Note]

The temperature probe holder (see Figure 1) that spans the two black spots on the multicell holder should be removed when inserting or removing cuvettes.

**Operation**

Operation of the accessory is through the Cary Eclipse software, with the exception of the stirrer control which is located on the front of the extended nose.

**To use the multicell holder:**

**Step 1**

From the setup dialog, select the Accessories tab to access the accessories page.

**Step 2**

Select the Multicell holder option and from the cell diagram, select the number of cells to use.

**Step 3**

**To enable temperature control (optional)**

a. Select the Temperature Control check box. From the Device option, select Temperature Controller.

b. Enter the required temperature and select the monitor. This monitor will allow data collection to begin when the temperature of the monitoring device is within ± 0.5°C.

**Step 4**

To display the temperature of other items in the Status Display window, select these items from the Temperature display option. Figure 2 (below) shows a typical setup of the multicell peltier accessory.

**Step 5**

Select OK to exit the Setup window.

**Step 6**

Select Start to begin the data collection.
Figure 2. Accessories tab showing a typical setup of the multicell holder accessory

Specifications

For environmental and power supply requirements, refer to your Cary operating instructions. This product is suitable for indoor use only.

Connections

- 25-pin D-range connector on flying lead.
- 14 pin circular lead (connect to Varian Temperature Controller)
- 2 water circulation tubes (connect to Varian Temperature Controller)

Weight

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Dimensions (W x D x H)

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<th>Condition</th>
<th>Dimensions (mm)</th>
<th>Dimensions (in)</th>
</tr>
</thead>
<tbody>
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<td>Packed</td>
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<td>30.1 x 14.2 x 12.8</td>
</tr>
<tr>
<td>Unpacked</td>
<td>180 x 320 x 180</td>
<td>7.1 x 12.6 x 7.1</td>
</tr>
</tbody>
</table>

Maintenance and Cleaning
All cleaning should be done with a soft cloth. If necessary, this cloth can be dampened with water or a mild detergent to clean black painted surfaces. Do not use organic solvents or abrasive cleaning agents.

See also:

Single Cell Holder (Peltier Thermostatted)

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- Introduction
- Specifications
- Getting Started
  - Unpacking
- Installation
  - Cuvette pitch adjustment
  - Alignment
  - Setting the temperature
- Operation
- Maintenance
- Trouble Shooting

Introduction

The single cell peltier is an electronically controlled thermostatting single cell accessory capable of controlling the temperature to within +/- 0.1°C. It provides excellent temperature reproducibility, stability and accuracy.

The single cell peltier has a temperature range of 0-100°C, but this range can be extended below 0°C with the aid of a refrigerated water bath. Accurate temperature setting and temperature feedback is provided through the Cary Eclipse Software interface.

Other accessories that can be used in conjunction with the Peltier Single Cell Accessory are:

- Cell Base
- Temperature Probe

Specifications

The performance specifications of the Peltier Single Cell accessory are:

- Temperature reproducibility ± 0.1 °C
- Temperature stability ± 0.1 °C
- Temperature accuracy ± 0.1 °C
- Temperature range 0-100 °C

The temperature display range is between 0 and 100 °C

Getting started
Unpacking

Unpack the accessory, and check that you have received the following:

- Peltier Cell Holder (with ribbon cable attached)
- Temperature control module
- Hexagonal ball driver
- Tubing and connectors
- Euroflux 600 water pump

Installation

You will need:

- The standard Cary Eclipse cell holder base
- The length of silicone rubber tubing provided
- Alternate sample compartment front panel (P/N 01 106483 90)
- A circulating water bath or tap water

To install the Peltier Cell Holder:

Step 1

See how to perform steps 1 to 4

Position the peltier accessory on the bench, next to your Cary Eclipse.

Step 2

Remove the top and front panels from the Cary Eclipse sample compartment and make sure that the sample compartment is empty.

Step 3

Note

Placing the cell holder on the cell holder base and attaching the tubing and ribbon cable to the cell holder should occur outside the sample compartment.

Place the cell holder on the cell holder base, aligning the notches in the cell holder with the raised black knobs in the cell holder base.

Step 4

Tighten the thumbscrew to secure the cell holder to the cell holder base.

Step 5

See how to perform steps 5

Take the small diameter tubing provided and cut it in half. Attach the tubing to the connectors on the cell holder (item 4 in Figure 1).
Step 6

Attach the two large rubber tubes to the ends of the small tubes using the two white connectors provided.

Step 7

Note
The out-flow of the cell holder must flow back into the water reserve or the water reserve will empty.

Attach one of the large tubes to the water pump outlet.

Step 8

Place the other large tube in the water reserve with the pump.

Note
The water flows from the water pump in the water reserve to the cell holder, the from the cell holder, back into the water reserve.

Step 9

See how to perform step 9

Connect the cell holder to the peltier accessory using the ribbon cable (item 5 in Figure 1).

Step 10

If available, fit the alternate sample compartment front panel to the instrument. When you place the cell holder and base into the sample compartment, ensure that the tubing and the ribbon cable from the cell holder protrude through the cutout at the bottom of the sample compartment front panel.

Step 11

If you intend to use the remote read switch (supplied with the alternate sample compartment front panel) to take readings:

(a) Plug the cable attached to the alternate sample compartment front panel into the port in the left hand wall of the sample compartment.

(b) Plug the remote read cable into the port on the front of the alternate sample compartment front panel.

Step 12

Install the cell holder and cell holder base in the sample compartment as follows:

See how to perform steps 12 to 14

(a) Place the cell holder base in the sample compartment, aligning the two locating holes over the two locating pins in the floor of the sample compartment.
(b) Move the cell base lever to the right.

![Diagram of cell holder components]

**Figure 1: Cell Holder components:**
1. Alignment hole and notch  
2. Adjustment screw  
3. Thumbscrew  
4. Tubing connectors  
5. Ribbon cable attachment

Step 13

Connect the RS-232C cable to the back of the peltier accessory. Connect the other end of the cable to a COM port at the back of your PC (they are usually marked |0|0|).

Step 14

Replace the power cable at the back of the PC.

Step 15

If you connected the cable to a COM port other than COM port 1 on your PC you will need to change the software settings in the Cary Eclipse software.

a. Startup the application you are going to use the Single Cell Peltier Accessory with and click the **Setup** button.
   b. Select the **Accessories** tab and check the **Temperature Control** group check box.
   c. Select **Automatic Temperature Setting**.
   d. Select **Single Cell Peltier**
   e. Click on the **Port** drop-down list
   f. Select the port that you have connected the cable to.
   g. Click **OK** to close the Setup dialog.

You should now align the cell holder.

For Cuvette Pitch Adjustment

Step 1

 ![See how to perform pitch adjustment procedure]
Turning the pitch adjustment in an anti-clockwise direction will raise the front of the cell holder.

Step 2

Turning the pitch adjustment in a clockwise direction will lower the front of the cell holder.

>Note
The pitch adjustment screw requires a hex key.

Alignment

Once the cell holder is installed in the sample compartment you need to align it in the light path for maximum light throughput.

To align the cell holder:

Step 1
Start the Align application by clicking the Start button in the Windows Taskbar and selecting Programs/Cary Eclipse /Align.

Step 2
Select the Cary tab.

Step 3
In the Instrument Parameters group, set the excitation wavelength to 0 nm (white light) by checking the Zero Order check box.

Step 4
Click Apply. (The green power indicator on the instrument should start flashing to indicate that the instrument is active.)

Step 5
Place a cell in the cell holder.

Step 6
Select the Graph tab and adjust the pitch screw to find the maximum intensity.

You should now set the required temperature.

Setting the temperature

To set the temperature:

The temperature setting for the peltier accessory is controlled from the software.

Step 1
Ensure that the voltage switch on the rear of the peltier accessory is set to the correct value for your location (120 or 230 V), connect the supplied power cord to the peltier accessory and the mains power supply, and turn the mains power on.

Step 2

Click on the **Setup** button in the Cary Eclipse application that is open.

Step 3

Click on the **Accessories** tab.

![Figure 1 - The Accessories tab (highlighting temperature control)](image)

1. **Accessories tab**
2. **Multicell/Temperature button**
3. **Temperature control group box**
4. **Temperature display group box**

Step 4

Click on the **Multicell/Temperature** button (the number of buttons in the **Accessories** tab will vary depending upon the application that is open).

Step 5
Check the Temperature control check box and select Single Cell Peltier from the drop-down list. Make sure that the COM port that is shown in the Port drop-down list matches the port that you have the peltier accessory connected to.

Step 6

The temperature of the peltier accessory can be set by clicking on the up or the down arrow (as shown in Figure 1). The up and down arrows can be found next to the Temperature (°C) entry field. Alternatively, you can enter a temperature directly into the Temperature (°C) entry field.

Step 7

You can get a constant read out of the Block temperature - both the temperature it is set to and its actual temperature along with the temperature of up to four probes. This is controlled in the Temperature display group box (also shown in Figure 1).

When the temperature has stabilized the cell holder is ready for use.

You may monitor the temperature of the cell contents by using the Cary Eclipse Temperature Probe accessory (part number 00 100408 00). Refer to the Temperature Probe instructions for further details.

Note

The time taken for the temperature to stabilize will depend upon several factors, including the ambient temperature, the viscosity of the solution and the required temperature.

Stirring

Switch on the STIR button and set the agitation speed (from 0-9). An agitation speed of 0 activates the stirrer.

Operation

To use the cell holder:

Step 1

See how to perform step 1

Place a cell in the cell holder.

Step 2

Set up your water circulating system to provide a flow rate of 0.5 litres per minute.

Step 3

Allow sufficient time for the cell and sample to warm to the specified temperature. This may take up to thirty minutes, depending on the nature of the solution.

Step 4

When all the components of the system are at the required temperature you may commence the measurement.
Maintenance

Regularly inspect the accessory to check for water leaks or degradation of the tubing. Replace when necessary.

Troubleshooting

<table>
<thead>
<tr>
<th>Problem</th>
<th>Solution</th>
</tr>
</thead>
</table>
| Water leaks inside the sample compartment. | Circulate hot water through the system and try to determine the source of the leak.  
Inspect the tubing and check that it is securely fitted to the Cell Holder. You may wish to use a sealant around the connections, but ensure that it does not absorb light at the wavelengths you are working at. |

Spare parts

The following spare parts are available from Varian. Only Varian supplied parts should be used, unless otherwise specified.

<table>
<thead>
<tr>
<th>Description</th>
<th>Part number</th>
</tr>
</thead>
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<tr>
<td>Single Cell Holder—Peltier Thermostatted</td>
<td>00 100764 00</td>
</tr>
<tr>
<td>Cary Eclipse Alternate sample compartment front panel</td>
<td>01 106790 90</td>
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</table>

Single Cell Holder (Water Thermostatted)

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- Introduction
- Specifications
- Installation
- Alignment
  - Pitch and Height Adjustment
    - Cuvette Pitch Adjustment
    - Cuvette Height Adjustment
  - Alignment procedure
- Setting the temperature
- Operation

Introduction

The Water Thermostatted Single Cell Accessory enables users to efficiently maintain cells at a specific temperature. This is achieved with the use of a constant flow of water to the accessory from a water bath. The PCB-150 can be used with the Water Thermostatted Single Cell Accessory or a third party water bath can be used.

The accessory is not only capable of handling regular cuvettes but can also handle micro-cells as well.

Specifications
The performance characteristics of the accessory are dependant on the specifications of the water bath that is used with the accessory. These performance characteristics can be achieved with the use of the PCB-150.

- Temperature range 20-60 °C
- Temperature accuracy ± 0.1 °C
- Reproducibility ± 0.05 °C
- Program ramp

**Installation**

Step 1

Make sure that the sample compartment is empty.

Step 2

Position the cell holder base on the two locating pins at the rear of the Cary Eclipse sample compartment.

Step 3

Turn the fastening lever to the right.

Step 4

Position the water thermostatted cell holder on the sample compartment base so that the groove in the cell holder points to the left wall of the sample compartment.

Step 5

Tighten the thumb screw of the water thermostatted cell holder

Step 6

Attach the PCB-150 to the Water Thermostatted Cell Holder and the Cary Eclipse according to the PCB-150 instructions.

**Alignment**

**Pitch and Height Adjustment**

Once the Water Thermostatted Cell Holder is installed in the sample compartment you need to align it in the light path for maximum light throughput.

For Cuvette Pitch Adjustment

Step 1

*See how to perform pitch adjustment procedure*
Turning the pitch adjustment in an anti-clockwise direction will raise the front of the cell holder.

Step 2

Turning the pitch adjustment in a clockwise direction will lower the front of the cell holder.

**Note**
The pitch adjustment screw requires a hex key.

**For Cuvette Height Adjustment**

Step 1

See how to perform height adjustment procedure

Turning the height adjustment screw in a clockwise direction will raise the cuvette.

Step 2

Turning the height adjustment screw in an anti-clockwise direction will lower the cuvette platform.

**Note**
To lower the cuvette you will have to push the cuvette down because of the tension created by the cell holder.

**Alignment procedure**

Step 1

Start the Align application by clicking the 'Start' button in the Windows Taskbar and selecting Programs > Cary Eclipse > Align.

Step 2

Select the Cary Tab

Step 3

In the Instrument Parameters group, set the excitation wavelength to 0 nm (white light) by enabling the Zero Order check box.

Step 4

Click 'Apply'. (the green power indicator on the instrument should start flashing to indicate that the instrument is active.)

Step 5

Place a cell in the Water Thermostatted Cell Holder.
Step 6

Select the Graph Tab and adjust the pitch screw on the cell holder to find the maximum intensity.

**Setting the temperature**

See the PCB-150 instructions for how to set the temperature.

**Operation**

To use the Water Thermostatted Cell Holder:

Step 1

Place a cell in the Water Thermostatted Cell Holder.

Step 2

Allow sufficient time for the cell and sample to warm to the specified temperature. This may take up to thirty minutes, depending on the nature of the solution.

Step 3

When all the components of the system are at the required temperature you may commence the measurement.

**Water 4 Position Multicell Holder Accessory**

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- Safety information
- Introduction
- Unpacking notes
- Installation
- Operation
- Specifications
- Maintenance and Cleaning

**Safety information**

Before using this accessory, you must read the Safety Practices and Hazards in your Cary Eclipse Hardware Manual.

A link to the Safety section is also available on the main page of the Cary Eclipse online Help and Videos CD.

If the equipment is used in a manner not specified by the manufacturer, your safety may be at risk and the protection provided by the equipment may be impaired.

**Introduction**
The Water-thermostatted 4 Position Multicell holder for the Cary Eclipse is an accessory that permits fluorescence measurements on up to four samples, while controlling the temperature and rate of stirring.

**Unpacking notes**

Make sure that all of the adhesive tape, plastic and cardboard is removed before operating the Peltier Multicell Holder.

**Installation**

**Step 1**

Turn the Cary Eclipse instrument power off using the switch on the front right side of the instrument. You may leave the PC turned on.

**Step 2**

Slide the top panel of the sample compartment all the way back and completely remove the front panel by sliding it upwards. Ensure that the sample compartment is empty.

**Step 3**

Lift the Multicell Holder by the handle and hold the end of the hose in your other hand. Reset the accessory at the front of the sample compartment in order to allow access to the D-shaped, 25 pin port at the left side of the rear sample compartment wall. Connect the cable plug to the 25 pin port.

**Step 4**

Ensure that the fastening lever is in the open position (i.e., to the left). Hold the extended nose (the black metal flange which contains the stirring controls, see Figure 1) and align the two holes on the base of the accessory with the locating pins on the floor at the rear of the sample compartment. Move the fastening lever from left to right to lock the accessory into position.
Figure 1. Water 4 Position Multicell Holder accessory

Step 5

Ensure the two black locking latches at the back of the extended nose are pointing upwards (unlocked, see Figure 1). Position the extended nose into place by aligning its locating pins with the holes in the front of the sample compartment floor. Lock the extended nose into position by pushing the locking latches downwards.

Step 6

Slide the special door for thermostatted accessories (part number 99 101032 00) into position ensuring the electrical cable is tucked neatly into the sample compartment.

Step 7

Before the accessory can be thermostatted you need to connect the hoses to the Varian PCB-150 Circulating Water Bath or other appropriate circulating water bath. To do this, connect the two black hose ends to the bath inlet and outlet connections. Ensure the water bath has sufficient liquid for circulation before you turn the bath on.

Step 8

Connect the nine pin end of the communication cable to a free serial port at the rear of the PC (e.g., COM 1). Connect the other end of this cable to the plug at the rear of the PCB-150.

Note

The temperature probe holder (see Figure 1) that spans the two black posts on the multicell holder should be removed when inserting or removing cuvettes.
Operation

Operation of the accessory is through the Cary Eclipse software, with the exception of the stirrer control which is located on the front of the extended nose.

To use the multicell holder:

Step 1

From the Setup dialog, select the Accessories tab to access the accessories page. (See Figure 2).

Figure 2. Accessories tab showing a typical setup of the multicell holder accessory.

Step 2

Select the Multicell holder option and from the cell diagram, select the number of cells to use.

Step 3

To enable the temperature control (optional)

   a. If using the PCB-150 water bath:
i. Select the Temperature Control check box.
ii. In the Device list, click the PCB-150 Water bath.
iii. In the Port list, click the appropriate COM port. This is the port you connected the communication cable to at the back of the PC.
iv. Enter the required temperature and select the monitor. This monitor will only allow data collection to begin when the temperature of the monitoring device is within ± 0.5°C of the temperature specified.

b. If using a non-Varian water bath:
   i. Select the Temperature Control check box
   ii. In the Device list, click None.
   iii. Enter the required temperature and select the monitor. This monitor will only allow data collection to begin when the temperature of the monitoring device is within ± 0.5°C of the temperature specified.

Step 4

To display the temperature of other items in the Status Display window, select these items from the Temperature display option. Figure 2 shows a typical setup of the multicell water accessory.

Step 5

Click OK to exit the Setup window.

Step 6

Select Start to begin data collection.

Specifications

For environmental and power supply requirements, refer to your Cary Eclipse Hardware Operation manual (part number 85 101758 00). This product is suitable for indoor use only.

Connections

- 25-pin D-range connector on flying lead
- 2 water circulation tubes (connect to Varian PCB-150 Circulating Water Bath or other appropriate circulating water bath)

Weight

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Dimensions (W x D x H)

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<tr>
<th></th>
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<tr>
<td></td>
<td>30.1 x 14.2 x 12.8 in</td>
<td>7.1 x 12.6 x 7.1 in</td>
</tr>
</tbody>
</table>

Maintenance and Cleaning
All cleaning should be done with a soft cloth. If necessary, this cloth can be dampened with water or a mild detergent to clean black painted surfaces. Do not use organic solvents or abrasive cleaning agents.

See also:

Water Circulator (PCB-150)

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- Getting started
  - Unpacking
- Installation
- Setting the temperature
- Operation
- Maintenance

Introduction

The PCB-150 (Peltier Controlled Cryobath, part number 00-100740-00) is a water circulator suitable for use with thermostatted cell holders such as the water-thermostatted single cell holder.

Specifications

- Temperature range: 20-60 °C
- Temperature accuracy: ± 0.1 °C
- Reproducibility: ± 0.05 °C
- RS232C interface: (communication protocol available on request to fluorescence@varianinc.com)

Getting started

Unpacking

Unpack the accessory, and check that you have received the following:

- Water circulator unit
- Power cord
- Water tubes
- Connectors.

Caution
Inspect all parts for damage during transit. Any damage should be reported immediately. Please refer to the accessory by model (PCB-150), part number (79 100296 00) and serial number (printed on the side of the packing box).

Installation

You will need the following to install the PCB-150 water circulator:

- A star headed screwdriver
- Syringe or other device to fill the unit with water
- De-ionized water
- Thermostatted cell holder

**Note**

This section assumes you have already installed the thermostatted cell holder you will be using in the sample compartment of your Cary Eclipse instrument, as per the instructions provided with the accessory.

To install the PCB-150 unit:

Step 1

Position the unit on the laboratory bench, next to your Cary Eclipse.

Step 2

See how to perform steps 2 and 3.

Unscrew the cap screw present in the ‘Water Fill’ inlet on the rear panel of the unit using a star head screwdriver.

Step 3

Using a syringe or a similar hand pump, inject approximately 50 mL of de-ionized water into the ‘Water Fill’ inlet.

Step 4

Replace the cap screw.

Step 5

See how to perform step 5

Connect the water tubes to the ‘Water In’ and ‘Water Out’ connectors on the rear panel of the unit.

Step 6

See how to perform step 6
Ensure that the voltage switch is set to the correct value for your location (120 or 230V), and connect the power cord to the unit and to the mains power supply.

Step 7

See how to perform step 7

Connect the free ends of the water tubes to the tubing leading from the Cary Eclipse sample compartment.

Step 8

Switch the unit on by setting the power button (on the front of the unit) to ‘I’.

This will start the pump on the water circulator, and water will start circulating through the tubing and the cell holder.

If the water level alarm comes on at this stage (the button lights up and an alarm sounds) you may need to top up the water level in the unit. Press the Water Level Alarm button to stop the alarm sounding. The light will remain on until the unit is filled with sufficient water.

You should now set the required temperature.

**Setting the temperature**

The temperature setting for the PCB-150 is controlled from the software.

To set the temperature:

Step 1

Click on the **Setup** button in the

Step 2

Click on the **Accessories** tab.
Step 3

Click on the **Multicell/Temperature** button (the number of buttons in the **Accessories** tab will vary depending upon the application that is open).

Step 4

Check the **Temperature control** check box and select PCB-150 from the drop-down list. Make sure that the Com port that is shown in the **Port** drop-down list matches the port that you have the PCB-150 connected to, at the back of the computer.

Step 5

The temperature of the PCB-150 can be set by clicking on the up or the down arrow (as shown in Figure 1). The up and down arrows can be found next to the **Temperature (°C)** entry field. Alternatively, you can enter a temperature directly into the **Temperature (°C)** entry field.

Step 6
Can can get a constant read out of the PCB-150 temperature - both the temperature it is set to and its actual temperature along with the temperature of up to four probes. This is controlled in the Temperature display group box (also shown in Figure 1).

Operation

To use the PCB-150 water circulator:

Step 1

Load your sample(s) in the cell holder as described in the instructions provided with the cell holder.

Step 2

Allow sufficient time for the cell(s) and solution(s) to warm to the specified temperature. This may take up to thirty minutes, depending on the nature of the solution(s).

Step 3

When all the components of the system are at the required temperature, close the sample compartment lid (optional for the Cary 50) and commence the measurement.

You may monitor the temperature of the cell contents by using the Cary Temperature Probe accessory (part number 00 100408 00). Refer to the Temperature Probe manual for further details.

Maintenance

Regularly inspect the accessory to check for water leaks or degradation of the tubing. Replace when necessary.

Automated Polarizer

Part number 00 100775 00

- Installation category 1
- Pollution degree 2
- Safety Class 3 (EN61010-1)

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Safety Information

Before using this accessory, you must read the Safety Practices and Hazards section in your Cary Eclipse Hardware Manual.
A link to the Safety section can also be found on the main page of your instrument Help and Videos CD.

If the equipment is used in a manner not specified by the manufacturer, your safety may be at risk.

**Introduction**

The Automated Polarization accessory for the Varian Cary Eclipse Fluorescence spectrophotometer provides a solution for polarization applications. Such measurements require exciting the sample with vertically polarized light and sequentially measuring the intensity of fluorescence through an analysing polarizer, oriented vertically and then horizontally.

The accessory consists of four metal arms extending from a central wheel (*Figure 1 (ii)*). Each arm is labeled as 1, 2, 3 or 4 by holes etched into the wheel, just below the filter position, (*Figure 3a*).

Two polarizers are mounted on the polarizer wheel (one polarizer per arm). Polarizers must be mounted in either positions 1 and 3 or positions 2 and 4. Solid black discs must be mounted in the remaining empty positions. This is to ensure the polarization wheel is balanced.

**Unpacking Notes**

Your accessory is packed in a foam-lined cardboard box. Inside the box you will find a wooden case that contains 2 vertical and 2 horizontal polarizers (*Figure 1 (iii)*), a plastic bag containing 4 black plastic discs (*Figure 1 (iv)*), and 2 cardboard boxes, each containing a motorized wheel, into which the polarizers must be mounted. Check that you have received all of the items in the accessory kit by referring to the packaging list included with the shipment (*Figure 1*).

![Figure 1. i) & iii) Two automated polarizers ii) Vertical and horizontal polarizers iv) Four black discs](image)

**Specifications**

For environmental and power supply requirements, refer to your instrument operating instructions. This product is suitable for indoor use only.
Installation

Installation of the Automated Polarization accessory can be divided into three stages:

1. Installing the polarizers and black discs in the two motorized polarizer wheels.
2. Installing the motorized wheels in the instrument.
3. Configuring the accessory using the software.

Installing the polarizers in the two automated polarizer accessories

Step 1

Using your fingers, carefully unscrew the knurled nut (Figure 2 (i)) in the centre of the plastic polarizer locking wheel (Figure 2 (ii)) and remove the locking wheel from the accessory.

![Figure 2](image-url) i) Knurled nut  ii) Polarizer locking wheel

Step 2

Polarizers now need to be placed in the accessories. There are four spaces, each labeled 1 to 4 by holes etched into the arms of the polarizer holder, (Figure 3a).
All four polarizer positions must be filled, using 2 polarizers and 2 black discs. By default the software will require you to place the polarizers in positions 1 and 3 in the polarizer holder. Each polarizer end is labeled with a "V" or "H" to identify if they are "vertical" or "horizontal" polarizers, (Figure 3b). Place the vertical polarizer in position 1 and the horizontal polarizer in position 3. The remaining two positions are filled with the black discs.

**Note**

The polarizers must be mounted in the motorized holding wheel so that the "V" or "H" label is facing away from the metal mounting, arm facing towards the user, (Figure 3b (vi)).

Lay one of the motorized wheels horizontally so that the metal arm is facing upwards. Carefully place the polarizer marked "V", vertical, into position 1 and the horizontal, "H" polarizer into position 3 so that the tip of the polarizer that contains the label is pointing out from the centre of the arm. Place a black disc in each of the remaining two positions, (position 2 and 4) (Figure 3b). Repeat this for the second polarization accessory.

**Figure 3b.** (i) & (iv) Fitted black discs (ii) Replaced Polarizer locking wheel (iii) & (v) Correctly fitted
polarizers (vi) Polarizer labeling. In this example the accessory is fitted with two polarizers. They are placed with "Vertical" in position 1 and "Horizontal" in position 3, as shown. The solid black discs are placed in positions 2 and 4.

Step 3

Replace the plastic locking wheel(s) removed in Step 1, ensuring the polarizers remain positioned at the arm ends (Figure 3b).

Replace the knurled nut(s) and rotate in a clockwise direction until finger tight. DO NOT over tighten.

Installing the accessory in the instrument

Step 1

Turn the instrument off and remove any existing accessories from the sample compartment.

Step 2

Prepare to remove the outer cover of the instrument by flipping up the four black plastic clasps located at each side of the instrument. Two of these are labeled (iv) and (v) in Figure 4.

Step 3

Remove the sample compartment front door and, using your thumb, lift the circular cover plate in the base of the instrument to expose a black plastic clasp, (Figure 4 (ii)). Flip the clasp up.

Step 4

Carefully remove the instrument outer cover by lifting it upwards away from the instrument base.

Step 5
With the instrument cover removed, the black metal inner covers are now visible (Figure 5). You will be able to see two plates, each secured by two black screws to the top of the instrument. Remove both the black plates by unscrewing the two black screws. These represent the positions for either excitation or emission measurement. An Automated Polarization accessory must be placed in both the excitation and emission chambers. The black plate labeled (i) in Figure 5 covers the emission chamber. The second black plate, closest to the front of the instrument, covers the excitation chamber.

![Figure 5. (i) Plate covering accessory chamber (emission) (ii) & (iii) Black screws (iv) Black thumb screw (v) Polarizer wheel orientation (vi) Accessory chamber (excitation) with cover removed (vii) & (viii) Alignment of the male and female D-plugs](image)

**Instilling an Automated Polarization accessory in the Excitation position**

To install an Automated Polarization accessory in the Excitation position, hold the accessory above the excitation chamber, located to the right of the sample compartment.

Orientate the accessory so that male D-plug located on the accessory is aligned with the female D-plug (Figure 5 (vii) & (viii)) inside the excitation chamber.

Orientate the polarizer wheel as shown in Figure 5 (v) so the arms of the wheel form a diagonal cross.

Carefully lower the accessory into the excitation chamber, raising the edge nearest the male D-plug on a 30° angle so the opposite edge of the accessory touches the excitation chamber first (Figure 6). Once the edge is positioned, lower the male D-plug so that it is sitting on top of the female D-plug in the excitation chamber.
**Figure 6.** The edge nearest the male D-plug is raised at a 30° angle with the opposite edge touching the excitation chamber.

To secure the Automated Polarization accessory gently push down on top of the accessory base directly above the two D-plugs. Once the plugs mate, tighten the black thumbscrews. The accessory is now fitted in the excitation position.

**Installing an Automated Polarization accessory in the Emission Position**

To install an Automated Polarization accessory in the Emission position, hold the accessory above the emission chamber, (*Figure 5 (i)*). Orientate the accessory so the male D-plug located on the accessory is aligned with the female D-plug (*Figure 5 (vii)*) inside the emission chamber.

Orientate the polarizer wheel as shown in Figure 5 (v) so the arms of the polarizer wheel form a diagonal cross.

Carefully lower both the edges of the accessory into the emission chamber ensuring the male and female D-plugs are aligned. Once the accessory is sitting in the emission position chamber gently push down on top of the accessory base plate directly above the two D-plugs. Once the plugs mate, tighten the black thumbscrews. The accessory is now fitted in the emission position.

**Step 6**

Once the two Automated Polarization accessories are secured by tightening the two black thumbscrews you can replace the instrument cover.

**Step 7**

Once the instrument cover is replaced flip down the four black plastic clamps and the front black plastic clamp to secure the cover. Replace the circular cover. (This is the reverse of steps 4, 3 and 2 in order).

**Step 8**

Install the accessory you would like to use with your Cary Eclipse, for example the single cell holder or multicell holder.
Step 9

If required, replace the sample compartment front door and close the sample compartment lid.

**Note**
If the Automated Polarization accessories are removed it is important that the black cover plates be replaced and secured using the black screws.

Configuring the accessory using the software

The Automated Polarization accessory is to be operated by the Thermal Application provided in the Cary Eclipse Bio Software.

**Note**
An ADL is required to operate the Automated Polarization accessory in the Scan and Advanced Reads applications. The ADL can be downloaded from http://www.varianinc.com/osi/fluoro/adl/

Step 1

Open the Thermal Application

Step 2

Turn on the instrument

Step 3

From the main Thermal window, select the Setup button to display the Setup dialog. On the Cary tab, select Go To in the Collect Temperatures group.
Figure 7. Software setup to allow the use of the automated polarizers, which are fitted in the excitation and emission monochromator chambers.

On the Accessories page of the Setup dialog, select the Polarizer check box and select the Auto radio button. Under Accessory Setup, select the Excitation polarizer and set position 1 to "Vertical" and position 3 to "Horizontal" on the polarization position diagram—that is, set position 1 to "V" and position 3 to "H". Select the Emission polarizer and set position 1 to the "Vertical" ("V") position and set position 3 to the "Horizontal" ("H") position.
Figure 8. Configuring the Thermal Application for use with the Automated Polarizers in the excitation position.

Step 4

You are now ready to use the automated polarizer with the Thermal Application. For more information about the Thermal or other Cary software applications, click the 'Help' button.

Accessory Removal

If you need to remove the Automated Polarizer accessories from the instrument to add or change polarizers, grasp the accessory base on the right of the end of the long edge and lift gently. Once the D-plug is disconnected, the accessory may be carefully removed.

If the accessories are not being re-inserted, be sure to replace the black metal covers over the polarization chambers, and tighten the locking screws.

Maintenance and Cleaning

Keep your accessory safely packed in its original box when not in use. Avoid unnecessary handling. If dust or fingerprints are observed on either surface of the polarizer, it should be cleaned using the following procedure.

Remove the polarizer from the motorized mounting wheel.
Moisten a piece of lint free tissue (such as a lens tissue) with 1-2 drops of spectroscopic grade methanol (NEVER drop solvent directly onto the polarizer).

Gently wipe the upper surface of the polarizer clean (be careful not to use excessive force).

Repeat the cleaning procedure on the lower surface using a fresh piece of lens tissue.

**Technical Assistance**

For technical assistance regarding this accessory, please contact fluorescence@varianinc.com

See also:
- **Accessories**
- **Information on the Automated Polarizer**
- **The Manual Polarizer**

**Manual Polarizer**

Part number 00 10761 00

- Installation category : 1
- Pollution degree : 2
- Safety Class : 3 (EN61010-1)

**Table of Contents**

- Safety information
- Introduction
- Unpacking notes
- Specifications
- Installation
- Maintenance and Cleaning

**Safety Information**

Before using this accessory, you must read the Safety Practices and Hazards section in your Cary Eclipse Hardware Manual.

A link to the Safety section can also be found on the main page of the Cary Eclipse online help system.

If the equipment is used in a manner not specified, the protection provided by the equipment may be impaired.

**Introduction**

One of the most effective ways of studying the rotational motion of molecules in solution is by using fluorescence polarization. With the Manual Polarizer Accessory, you can calculate the polarization and anisotropy of a sample.

**Unpacking notes**
Specifications

This product is suitable for indoor use only.

The polarizers are made of thin film polarizing material.

**Wavelength range**

275-750nm

**Weight**

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<tr>
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**Dimensions (W x D x H)**

Packed 200 x 115 x 113 mm (7.9 x 4.5 x 4.5 in)

**Installation**

**Step 1**

Clear the sample compartment

**Note**

The polarizers are delicate pieces of equipment and clearing the sample compartment is a safety precaution to ensure that the polarizers can be inserted easily.

**Step 2**

To fit the polarizer to the right-hand corner of the sample compartment, locate the accessory in the tow locating pin holes.

**Step 3**

Tighten the thumb screw to secure the polarizer frame.
Polarizer Angles

The two polarizers can be set to four possible angles.

Vertical (V)

The V located at the top of the polarizer indicates vertical or 0°.

35°

The first notch in a clockwise direction is 35°.

55°

The second notch in an anti-clockwise direction is 55° (magic angle 54.7°).

Horizontal (H)

The third notch in an anti-clockwise direction is 90° or horizontal.

Operation

For operational instructions, refer to the Cary Eclipse Online Help System.

Maintenance and Cleaning

All cleaning should be done with a soft cloth. If necessary, this cloth can be dampened with water or a mild detergent to clean black painted surfaces. Do not use organic solvents or abrasive cleaning agents.

Note

If you do come into contact with the silica lens, it can be cleaned with spec grade methanol and lens tissue.

See also:

Accessories
Information on the Manual Polarizer
The Automated Polarizer
Tips and Tricks

The list below shows some useful techniques that can be used with the Cary Eclipse software.

Right-click for menus

If you right-click in the graphics or report area you can quickly access the appropriate menu commands for that area.

Can't find a file you know that you stored?

If you can't find a file in the directory that you stored it in it may be because the wrong file type is selected in the File, Open dialog box. Just type .* in the file name field and press Enter. There are two file types in which data can be stored—Data files and Batch files. The Data file type stores only the traces that are visible when you select Save. The Batch file type stores the traces as well as the current method so that the method will be loaded when you retrieve the data in the batch file.

Creating your own screen saver

The Cary software installs a screen saver called VarianMM. You can use this screen saver to display a video or a message on the PC when it is not in use. To use it as a screen saver follow these steps:

1. Click the Windows Start button.
2. Select Settings, Control panel, Display.
3. Select the Settings tab.
4. Check that the Color Palette is set to High color (16 bit) or greater. Click Apply. Select "Apply settings without restarting". Click OK to keep the settings.
5. Select the Screen saver tab.
6. Select VarianMM from the drop down list in the Screen Saver field.
7. Click Settings.
8. Right-click on the Play list area.
9. Select Add...
10. Find and select the .AVI or .MPG file that you would like to use as a screen saver. Click OK.
11. Select Double size from the drop down list in the Video Size field. Click OK.
12. Click Preview. The screen saver will run.
13. If you want to use this screen saver select OK.

Note:
You can play any series of .AVI or .MPEG files using the VarianMM screen saver.

Retrieving files to overlay

The Batch vs Data file system also has implications for retrieving traces to overlay on the screen. If you want to overlay two traces which have been stored in different files one of them needs to be a Data file i.e they can't both be Batch files. As a Batch file loads a method as well as the traces you can only load one batch file at a time. If you have traces in a Batch file that you want to overlay on other traces in another batch file, open up one of the files and save the traces as a Data file. You will then be able to overlay the Data file's traces on the Batch file traces.

Another trick: Start off with Batch file as the file type selected in the File, Open dialog box. Select the file to use. Then change the file type selection to Data. Press OK and the system will extract the Data files and load them without the Batch information.
Previewing a report on screen before printing

After you have used the Recalculate button to generate a Report select the File, Print preview menu command to see how the report will look when it is printed. Use the arrow keys at the bottom to scroll through the pages.

Cut and paste a report to a word processor for extra formatting control

If you want to control where page breaks happen or you want the extra formatting capabilities that a word processor offers you can cut and paste a Cary Eclipse report into a word processor. First, use the Recalculate button to generate the report. Then right-click on the report area in the Cary Eclipse software. Choose 'Select all' from the menu. Right-click again and select 'Copy'. Switch to your word processor and paste. To get graphics in the report switch back to the Cary Eclipse software, right-click on the graphics area and select Copy graph from the menu. Switch to your word processor, position the cursor where you want the graph and select paste.

Sending an electronic copy of the report to others

If you want to send (by email for example) a report to someone else you have several options:

- You can use the File | Save Data As option to save a report file, and then send them this file. However, they will need to have the application that you used to generate the report files loaded on their PC.
- You can export the report to a Word processor as described above.
- You can print the report to an Acrobat .pdf file. The recipient only needs to have a copy of the Acrobat reader which can be downloaded for free from the web at www.adobe.com.

To print to a .PDF file from an application you will need to purchase the Adobe Acrobat program. When you have Acrobat Acrobat installed you will be able to select Acrobat PDFWriter as the printer in the Print dialog. This will automatically create a .PDF file for you.

Importing sample names with weight and volume information

In the Advanced Reads and Concentration applications, you can import sample names into the application with weight and volume corrections already applied. Simply separate the sample name, volume and weight by a comma. (e.g. My Sample in Methanol, 1.0001, 101.) Duplicates and triplicate information can be similarly imported. See the online Help for Samples tab of either Advanced Reads and Concentration for more information.

Autoscaling graphs

You can Autoscale the focused traces of multiple graph boxes simultaneously. To do this, first select the graph boxes you require by:

- holding down the Ctrl key and selecting each required graph box;
- holding down the Shift key and selecting any graph box to select all graph boxes; or,
- holding down the Shift key and selecting any graph box to select all graph boxes, then deselecting those not required by selecting them whilst holding down the Ctrl key.

Once you have selected the graph boxes to be Autoscaled, select the appropriate Autoscale icon or Autoscale menu entry.

Another trick: The three methods of selecting graph boxes mentioned above can also be used in other ways in addition to Autoscaling. For example, if you wanted to delete all but one graph box, you could hold down the Shift key and click on any graph to select all graph boxes, then hold down the Ctrl key and deselect the graph you wish to keep. Clicking on the Remove Graph toolbar icon (or Graph menu item) would delete all selected graph boxes.

Keeping Graph Preferences settings
To store the Settings from the Graph Preferences dialog, such as axes color, and width, so that the system remembers these settings every time an application is opened you will need to do the following:

1. Open the application of interest
2. Add a graph to the graphics area
3. Open the graph preferences dialog and setup all your graph settings
4. From the file menu select the Save Data As menu and save your settings as a batch file. For example 'Kinetics Default settings.fbkn'.
5. Create a shortcut on the desktop for this batch file.
6. Next time you want to open this application, rather than selecting the icon in the Cary Eclipse folder on the desktop, select the shortcut (e.g. 'Kinetics Default settings.fbkn') icon instead and you will get your graph preferences load automatically.

⚠️ Note:
This needs to be done separately for each application.

**Setting up to use a specific brand of wellplate (wellplate)**

If you consistently use a specific brand of wellplate you may wish to save time by setting up a profile for that plate. Each brand has slightly different dimensions and well positions. To create a profile, refer to How to create or modify a wellplate format. Once you have created your own profile you will be able to use it in all appropriate Cary Eclipse applications.
Maintenance

**Warning**  This instrument contains electrical circuits, devices, and components operating at dangerous voltages. Contact with these circuits, devices and components can cause death, serious injury, or painful electrical shock.

Operators and other unauthorized personnel must never remove the main cover. This must be opened only by Varian-trained, Varian-qualified, or Varian-approved service engineers.

This Help deals with the maintenance procedures for the Cary Eclipse Fluorescence Instrument. You should refer to your PC and printer/plotter manuals for their maintenance practices.

Any maintenance procedures not specifically mentioned here should be carried out only by Varian-trained, Varian-qualified or Varian-authorized service engineers.

- Cleaning your instrument
- How to install and remove the lamp module
- How to align the lamp module
- How to replace/check a fuse
- How to care for flowcells

See also:
- **Warning Symbols**


Cleaning your instrument

Any spills in the sample compartment should be wiped up immediately as well as any deposits on the sample compartment windows. The exterior surfaces of the Cary Eclipse spectrophotometer should be kept clean.

All cleaning should be done with a soft cloth. If necessary, this cloth can be dampened with water or a mild detergent. Do not use organic solvents or abrasive cleaning agents.
How to care for flow cells

General care of flow cells

It is important to inspect the flow cell regularly and ensure that it is kept clean and free from bubbles. If the cell is not clean, or if air bubbles are allowed to remain in the cell, the analytical results may be wrong, and the zero position may drift.

Cleaning instructions for flowcells

Step 1

Prepare a general wetting solution of Tween* 80 in water (5 to 10% v/v) and run this solution through the cell at regular intervals. If the routine sampler is not to be used for an extended period, this wetting solution should be pumped into the flowcell to keep all tubes and cell walls coated. The system should be thoroughly rinsed with distilled water immediately before use.

Step 2

If a stronger treatment is required, a solution of KOH in methanol can be used (about 1N KOH). Rinse the system briefly with this solution and flush thoroughly with distilled water. Do not leave the KOH/methanol in contact with the cell for long periods.

Step 3

To remove air bubbles, proceed as follows:

a. Purge the cell completely with air several times.

b. Flush the cell thoroughly with Tween* 80 solution or KOH/methanol solution. Rinse thoroughly with distilled water.

c. Flush the cell thoroughly as in step (a) before using the cell.

Tween* 80 is a registered Trademark of J.T. Baker Chemical Co.
How to install and remove the lamp module

This section describes how to replace the lamp module on the Cary Eclipse. Before changing the lamp module, ALWAYS disconnect the Cary Eclipse from the PC.

Removing the lamp module

Warning

When operating, the lamp module emits high intensity light which can damage eyes. To avoid eye damage, never operate the lamp module outside the instrument.

The lamp module contains components operating at high voltages. To avoid electric shock, NEVER disassemble the lamp module.

MIRRORS
Even though the mirrors are guarded, be careful removing and installing the lamp module so they (and other instrument components) are not damaged.

Lamp module removal

To remove the lamp module:

Step 1

Disconnect the Cary Eclipse from the mains power supply.

See how to perform steps 2 to 5.

Step 2

Undo the five clips - two on either side of the instrument, and the clip at the front of the instrument, under the circular plug.

Note:
Make sure that the sample compartment is empty

Step 3

Remove the instrument cover.

Step 4

Facing the front of the Eclipse—turn the instrument onto its right side to give access to the base.

Step 5

On the base of the instrument, undo and remove the four thumb screws of the lamp module cover.

See how to perform steps 6 and 7.
Step 6

Through the front opening you will see three bolts, each marked with a black dot . Undo the three bolts until they are free to move.

Step 7

Move the lamp module towards the front of the instrument to disengage two locating pins at the rear of the lamp module.

See how to perform steps 8 to 10.

Step 8

Partially withdraw the module through the opening in the base.

Step 9

Squeeze the clip on the plug and remove the plug from its socket.

Step 10

Withdraw the lamp module completely.

Note:
The lamp module must not be disassembled by an operator.

Lamp module installation

See how to perform steps 1 to 3.

Step 1

Plug the connector of the new lamp module into the socket in the instrument.

Step 2

Fit the lamp module in the base of the Cary Eclipse, ensuring that the two guide pins are correctly located and that the wiring is kept clear of the other parts of the instrument.

Step 3

Tighten the three bolts through the front opening.

See how to perform step 4.

Step 4

Replace the bottom panel and tighten the four thumb screws.
See how to perform step 5.

Step 5
Return the instrument to its upright position, put the cover back on the instrument and latch all five clips.

Step 6
Replace the power cable at the rear.

Step 7
Turn the PC and the instrument back on.

After replacing the lamp module you must align the source mirror.
How to replace/check a fuse

When replacing fuses in the Cary Eclipse spectrophotometer, you must observe the following warning:

**Warning**

For continued protection against the risk of fire, replace only with fuses of the specified type and current rating.

The spectrophotometer contains two fuses that are located at the back of the instrument.

To replace/check a fuse:

**Step 1**

Disconnect the fluorescence spectrophotometer from the power supply.

**Step 2**

Undo the fuse cover - which is located under the instrument's power cable at the rear of the instrument - by levering it open with a blade headed screw driver.

**Step 3**

Lift the cover while removing the red fuse housing. The red fuse housing contains the two fuses.

**Note:**

For the specifications of replacement fuses, see the back of the instrument.

**Step 4**

Remove the damaged fuse(s) from and replace it with one that matches the specifications.

**Step 5**

Replace the red fuse housing back into the housing slot and close the fuse housing cover.

If a fuse blows repeatedly, it may indicate other problems with the Cary Eclipse instrument. You should contact your local Varian sales office and arrange a service call.

See also:

*Warning Symbols*
How to align the lamp module

**Warning**

When operating, the lamp module emits high intensity light which can damage eyes. To avoid eye damage, never operate the lamp module outside the instrument.

The lamp module contains components operating at high voltages. To avoid electric shock, NEVER disassemble the lamp module.

**Caution**

**Mirrors:**
Even though the mirrors are guarded, be careful removing and installing the lamp module so they (and other instrument components) are not damaged.

Lamp module alignment

To align the lamp module:

**Note**

The instrument power must be on for the alignment procedure.

**Step 1**

See how to perform steps 1 and 2

Open the Align application. Select **Cary Eclipse** then **Align**.

**Step 2**

Undo the five clips, two on either side of the instrument and the clip at the front of the instrument under the circular plug. Remove the instrument cover.

**Step 3**

See how to perform steps 3 to 6

In the Align application, select the **Cary** tab and set up the instrument parameters as follows:

**Note**
Emission parameters are not important for this procedure.

- Ex. Wavelength (nm) 540
- Ex. Slit (nm) 1.5
- Ave. time (s) 0.1
- PMT Voltage Medium

Press **Apply**.
Step 4

Select the **Graph** tab, select the **Reference** button.

Step 5

Click on the **Vertical Rescale** button to find the signal range.

Step 6

Click on the **Scale** button and enter an upper limit that is 20% higher than the current signal and a lower limit that is 20% lower than the current signal. This will make the lamp module alignment easier when observing the graph.

Step 7

> See how to perform steps 7 to 11

**Note**

The brass adjustment screws for vertical and horizontal adjustment are clearly marked.

---

**Figure 1**

*Lamp module adjustment screws*

Adjust the bottom left screw (vertical adjustment) for maximum signal with the 5.5 mm hex driver supplied (you may need to adjust the y-axis scaling).
Step 8

Adjust the top right screw (horizontal adjustment) for maximum signal with the 5.5 mm hex driver supplied (you may need to adjust the y-axis scaling).

Figure 2
1. Maximum signal level

Step 9

Select the Lamp tab.

Step 10

Click on the Save conditions button to store the instrument parameters.

Step 11

Replace instrument cover and fasten the five clips.
How to Contact Varian

If you need to contact your local Varian office or Dealer then select from the list below. Please note that these contact details are updated frequently, you can find the latest version on our website at: http://www.varianinc.com

Varian Offices And Dealers—World Regions

- North America
- Asia/Pacific
- Europe
- Middle East
- Africa
- Central and South America

Varian Optical Spectroscopy Instruments

Your Cary instrument was designed and manufactured in Australia at Varian's plant in Melbourne. You can contact us about anything you like. Here are the contact details:

Email: fluorescence@varianinc.com

You can also contact the Fluorescence Product Manager on:

fax: +61 3 9560 7950
telephone: +61 3 9560 7133
mail: Varian Australia
      679 Springvale Road
      Mulgrave, Victoria, 3170
      Australia

Varian Offices and Dealers—Africa

Note that the phone/fax numbers are in the format: + AA BB number, where the '+' refers to the international dialing code for your country, 'AA' is the country code and 'BB' is the area code (without any leading zeros).

- Egypt
- Morocco
- South Africa

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<tr>
<td>c/o Akhnaton Trading &amp; Representation</td>
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**Varian Offices and Dealers—Asia and Pacific**

Note that the phone/fax numbers are in the format: + AA BB number, where the '+' refers to the international dialing code for your country, 'AA' is the country code and 'BB' is the area code (without any leading zeros).

- [Australia and Head Office](#)
- [Burma](#)
- [China](#)
- [Hong Kong](#)
- [India](#)
### Australia and Head Office

<table>
<thead>
<tr>
<th>Varian Australia Pty Ltd, 679 Springvale Road</th>
<th>Phone: + 61 1300 368 274</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mulgrave Victoria 3170, P.O. Box 222, Clayton South, VIC. 3169.</td>
<td>Fax: + 61 1300 658 274</td>
</tr>
<tr>
<td>Australian sales enquires: <a href="mailto:sales@osi.varian.com">sales@osi.varian.com</a></td>
<td></td>
</tr>
<tr>
<td>Australian service enquires: <a href="mailto:service@osi.varian.com">service@osi.varian.com</a></td>
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<table>
<thead>
<tr>
<th>Varian Australia Pty Ltd, 6/81 Frenchs Forest Road (East)</th>
<th>Phone: + 61 1300 368 274</th>
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</thead>
<tbody>
<tr>
<td>Frenchs Forest, N.S.W. 2086,</td>
<td>Fax: + 61 1300 658 274</td>
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<table>
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<tr>
<th>Varian Australia Pty Ltd, 14/43 Lang Parade</th>
<th>Phone: + 61 1300 368 274</th>
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<tbody>
<tr>
<td>Milton, Q’LD. 4064,</td>
<td>Fax: + 61 1300 658 274</td>
</tr>
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<table>
<thead>
<tr>
<th>Varian Australia Pty Ltd, Unit 3, 63 Walters Drive</th>
<th>Phone: + 61 1300 368 274</th>
</tr>
</thead>
<tbody>
<tr>
<td>Herdsman Business Park, Osborne Park, W.A. 6017.</td>
<td>Fax: + 61 1300 658 274</td>
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<tr>
<th>Varian Australia Pty Ltd, Unit 3, 422-424 Torrens Road</th>
<th>Phone: + 61 1300 368 274</th>
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<tr>
<td>Kilkenny, S.A. 5009,</td>
<td>Fax: + 61 1300 658 274</td>
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### Burma

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<tr>
<th>U Tin Hla, 24 Tank Road</th>
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*Indonesia  
Japan  
Korea  
Malaysia  
New Zealand  
Pakistan  
Philippines  
Singapore  
Sri Lanka  
Taiwan  
Thailand  
Vietnam*
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<tr>
<td>Myanmar</td>
<td>Ywama, Insein</td>
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<tr>
<td></td>
<td>Yangon, Myanmar</td>
<td></td>
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</tr>
<tr>
<td>China</td>
<td>Varian Technologies China Limited.</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Rm.1201, Jinyu Mansion</td>
<td>Phone+86 10 66081530, 66081531, 66081532, 66081533</td>
<td></td>
</tr>
<tr>
<td></td>
<td>129A# Xuanwumen Xidajie,</td>
<td>Fax+86 10 66411534</td>
<td><a href="mailto:varianbj@varianinc.com.cn">varianbj@varianinc.com.cn</a></td>
</tr>
<tr>
<td></td>
<td>Xicheng District</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Beijing 100031, P.R.China</td>
<td>Email: <a href="mailto:varianbj@varianinc.com.cn">varianbj@varianinc.com.cn</a></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Varian Technologies China Limited.</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Rm 2005 Shanghai Square</td>
<td>Tel +86-21-63756969, 63756930/31/32/33/34/35/36</td>
<td></td>
</tr>
<tr>
<td></td>
<td>138# Huaihai Zhong Road</td>
<td>Fax+ 86-21-63756848</td>
<td><a href="mailto:variansh@varianinc.com.cn">variansh@varianinc.com.cn</a></td>
</tr>
<tr>
<td></td>
<td>Shanghai 200021</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Varian Technologies China Limited.</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Rm 3-122 City Square</td>
<td>Tel +86-24-23341248/49/50</td>
<td></td>
</tr>
<tr>
<td></td>
<td>206# North Nanjing St.</td>
<td>Fax+ 86-24-23341095</td>
<td><a href="mailto:variansy@varianinc.com.cn">variansy@varianinc.com.cn</a></td>
</tr>
<tr>
<td></td>
<td>Heping District</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Shenyang 110001, P.R.China</td>
<td>Email: <a href="mailto:variansy@varianinc.com.cn">variansy@varianinc.com.cn</a></td>
<td></td>
</tr>
<tr>
<td>Hong Kong</td>
<td>South East Chemicals &amp; Instruments Ltd</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>1210 Chai Wan Industrial City Phas 1</td>
<td>Phone: + 852 2802 6333</td>
<td></td>
</tr>
<tr>
<td></td>
<td>60 Wing Tai Road</td>
<td>Fax: + 852 2807 0542</td>
<td><a href="mailto:sechk@netvigator.com">sechk@netvigator.com</a></td>
</tr>
<tr>
<td></td>
<td>Chai Wan</td>
<td></td>
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<tr>
<td>India</td>
<td>Varian India Pvt Ltd</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Second Floor, ICC Chambers</td>
<td>Phone: +91 22 8570181 / 8570787 / 8570788 / 8575790 / 8576791</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Saki Vihar Road, Powai</td>
<td>Fax: 91 22 8572183 / 8570792</td>
<td><a href="mailto:varian.mum@rma.sprintrpg.ems.vsnl.net.in">varian.mum@rma.sprintrpg.ems.vsnl.net.in</a></td>
</tr>
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<td>Mumbai 400 072</td>
<td>Email: <a href="mailto:varian.mum@rma.sprintrpg.ems.vsnl.net.in">varian.mum@rma.sprintrpg.ems.vsnl.net.in</a></td>
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<tr>
<td></td>
<td>Varian India Pvt Ltd</td>
<td>Ph: 91 44 499 5828/499 2172/499 2918</td>
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<tr>
<td></td>
<td>Varian House</td>
<td>Fax: 91 44 499 3381</td>
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<tr>
<td></td>
<td>12 Bawa Road</td>
<td>Email: <a href="mailto:varian.mad@rma.sprintrpg.ems.vsnl.net.in">varian.mad@rma.sprintrpg.ems.vsnl.net.in</a></td>
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</tr>
<tr>
<td>Chennai 600018</td>
<td>Varian India Pvt Ltd, 101 Competent House, 7, Nangal Raya Business Centre</td>
<td>+91 11 554844</td>
<td>+91 11 554 8445</td>
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<tr>
<td>New Delhi 110 046</td>
<td>PT. Unitama Analitaka Perkasa, Jl Garuda No. 31C</td>
<td>+62 21 426 8478, 426 8479, 426 8480</td>
<td>+62 21 426 8481</td>
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<tr>
<td>Jakarta 10610</td>
<td>Varian Technologies Japan, Sumitomo Shibaura Building, 8F, 4-16-36, Shibaura, Minato-Ku</td>
<td>+81 3 52321239</td>
<td>+81 3 52321264</td>
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<tr>
<td>Tokyo 108, Japan</td>
<td>(Branch office) Varian Technologies Japan, Shin-osaka Doi Building, 7-5-25, Nishinakajima</td>
<td>+81 6 305 6552</td>
<td>+81 6 305 6556</td>
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<tr>
<td>Korea</td>
<td>Varian Technologies Korea Ltd, Shinsa 2nd Bldg., 2F, 966-5 Daechi Dong</td>
<td>+82 2 345 22452</td>
<td>+82 2 34522451</td>
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<tr>
<td>Seoul, Korea, 135-280.</td>
<td>Varian Korea Ltd, 433-1 Mogok Dong, Songtan-Shi, Kyungki-Do, Korea, 459040</td>
<td>+82 333 665 5171</td>
<td>+82 333 665 0117</td>
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<tr>
<td></td>
<td>Daejeon office, Hanjin Officetel 1006</td>
<td>+82 42 823 4022</td>
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<tr>
<td>551-7 Bongmyoung Dong</td>
<td>+82 42 825 8645</td>
<td>Yusung-Ku, Daejeon, Korea.</td>
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<tr>
<td>Varian Technologies Korea Ltd.</td>
<td></td>
<td>Sojun Bldg., SF876-6 Dal-Dong</td>
<td>+82 522 654518/9</td>
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<td>Namgu, Ulsan</td>
<td>+82 522 614469</td>
<td>Jyungnam, Korea. 680-030</td>
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<tr>
<td>Malaysia</td>
<td></td>
<td>Siber Hegner (M) Sdn Bhd</td>
<td>+603 460 9501</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Unit F601, Phileo Damansara 1</td>
<td>+603 758 4325</td>
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<td>No. 9 Jalan 16/11 off Jalan Damasara</td>
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<td>46350 Petaling Jaya</td>
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<tr>
<td>New Zealand</td>
<td></td>
<td>A I Scientific</td>
<td>+64 9 478 1351</td>
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<td>P.O.Box 35579</td>
<td>+64 9 478 1360</td>
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<tr>
<td></td>
<td></td>
<td>Browns Bay</td>
<td>Mobile: +64 21 633 940</td>
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<td>Auckland</td>
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<tr>
<td></td>
<td></td>
<td>Unit J, 46 Constellation Drive</td>
<td>Email: <a href="mailto:aissinz@ihug.co.nz">aissinz@ihug.co.nz</a></td>
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<td>Mairangi Bay, Auckland</td>
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<tr>
<td>Pakistan</td>
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<td>Head Office</td>
<td>+92 42 7320309, 7237392, 7323368</td>
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<td>We Brothers Scientific Work</td>
<td>Telex: 44518</td>
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<td>Rana Chambers 18 Lake Road</td>
<td>+92 42 7235630</td>
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<tr>
<td></td>
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<td>Lahore 54000. Pakistan</td>
<td>Email: <a href="mailto:brothers@paknet1.pct.pk">brothers@paknet1.pct.pk</a></td>
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<td>2nd Floor, 65C</td>
<td>Phone: +92 21 5899541, 5899542, 5888507</td>
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<td>Commercial Area-A</td>
<td>Telex: 24649</td>
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<td>Defence II</td>
<td>Fax: +92 21 588 8507</td>
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<td>Karachi 75500. Pakistan.</td>
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<td>We Brothers Scientific Works</td>
<td>Phone: +92 51 277216, 277244</td>
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<tr>
<td></td>
<td></td>
<td>House No.6 Street No. 37</td>
<td>Fax: +92 51 277244</td>
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<tr>
<td></td>
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<td>F-6 Near Farooqia Market</td>
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<td>Islamabad, Pakistan.</td>
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<td>Philippines</td>
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<tr>
<td><strong>Molave Trading, Inc.</strong></td>
<td><strong>Phone:</strong> +63 2 4110360</td>
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<td>P.O. Box No. 590</td>
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<td>Greenhills Post Office</td>
<td><strong>Fax:</strong> +63 2 924 2094</td>
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<td>Greenhills, San Juan</td>
<td><strong>Email:</strong> <a href="mailto:aceroent@mnl.sequel.net">aceroent@mnl.sequel.net</a></td>
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**Sri Lanka**

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<tr>
<th>Analytical Instruments Put Ltd</th>
<th><strong>Phone:</strong> +941 575 025</th>
</tr>
</thead>
<tbody>
<tr>
<td>2nd floor, Vogue building</td>
<td><strong>Fax:</strong> +941 573 945</td>
</tr>
<tr>
<td>528 Galle Road</td>
<td></td>
</tr>
<tr>
<td>Colombo</td>
<td>e-mail: <a href="mailto:anains@dynanet.lk">anains@dynanet.lk</a></td>
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**Singapore**

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<thead>
<tr>
<th>SiberHegner (South-East-Asia) Private Limited</th>
<th><strong>Ph:</strong> +65 274 1222</th>
</tr>
</thead>
<tbody>
<tr>
<td>102E Pasir Panjang Road, #04-05/12</td>
<td><strong>Fax:</strong> +65 273 1503</td>
</tr>
<tr>
<td>Citilink Warehouse Complex, Singapore 118529</td>
<td>Email: <a href="mailto:kelvin.low@siberhegner.com.sg">kelvin.low@siberhegner.com.sg</a></td>
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**Taiwan**

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<tr>
<th>Varian Technologies Asia, Ltd.</th>
<th><strong>Phone:</strong> +886 22 698 9555</th>
</tr>
</thead>
<tbody>
<tr>
<td>18F - 13, No. 79 Hsin Tai Wu Rd., Sec. 1,</td>
<td><strong>Fax:</strong> +886 22 698 9678</td>
</tr>
<tr>
<td></td>
<td>email: <a href="mailto:contact@tw.varian.com">contact@tw.varian.com</a></td>
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**Thailand**

<table>
<thead>
<tr>
<th>Thai Unique Co. Ltd.</th>
<th><strong>Phone:</strong> +66 2 282 9749</th>
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</thead>
<tbody>
<tr>
<td>80-82 Pracharthipatai Road</td>
<td><strong>Telex:</strong> 20775</td>
</tr>
<tr>
<td>Bangkok 10200, Thailand</td>
<td><strong>Fax:</strong> +66 2 280 1788</td>
</tr>
<tr>
<td></td>
<td>Email: <a href="mailto:thawatt@mozart.inet.co.th">thawatt@mozart.inet.co.th</a></td>
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**Vietnam**

<table>
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<tr>
<th>Siber Hegner Machinery</th>
<th><strong>Phone:</strong> +84 4 8244 912 or : +84 4 244 913</th>
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<tr>
<td>Suite 3 26/3.28, Binh Minh Hotel</td>
<td><strong>Fax:</strong> + 844 8244915</td>
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<tr>
<td>27 Ly Thai To St</td>
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<td>Hanoi Vietnam</td>
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<tr>
<th>Siber Hegner</th>
<th><strong>Phone:</strong> +848 8223 079</th>
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<tbody>
<tr>
<td>Suite 103 City view building</td>
<td><strong>Fax:</strong> +848 8223 084</td>
</tr>
<tr>
<td>12 Mae Dinh Chi Street, Dist. 1</td>
<td></td>
</tr>
<tr>
<td>HO CHI MINH CITY</td>
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## Varian Offices and Dealers—Europe

Note that the phone/fax numbers are in the format: + AA BB number, where the '+' refers to the international dialing code for your country, 'AA' is the country code and 'BB' is the area code (without any leading zeros).

- **Austria**
  - Varian GesmbH,
  - Phone: +43 1 699 9669
  - Eisgrubengasse 2
  - Fax: +43 1 699 40 18
  - A-2334 Vosendorf, Austria
  - Email: varian.vienna@at.varian.com

- **Belgium**
  - NV Varian Belgium SA
  - Tel: +32 (0) 15-55 64 60
  - Mechelsesteenweg 362
  - Fax: +32(0) 15-55 61 86
  - 2860 Sint-Katelijne-Waver
  - Email: varian.benelux@be.varian.com

- **Bulgaria**
  - Spectratech
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  - 5, Rilski Ezera str
  - Fax: + 35 92 962 4501
  - BG-1407 Sofia
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- **Croatia**
  - AnAs - Analytical Instruments
  - Phone: +385 1 6600 785
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  - Fax: +385 1 6600 785
  - HR-10010 Zagreb, Croatia
  - Mobile phone: +385 98 27 8851
  - Email: vladimir.svob@zg.tel.hr

- **Czech Republic**
  - Amedis spol. S.R.O.
  - Phone: + 420 2 740 753

### Table

<table>
<thead>
<tr>
<th>Country</th>
<th>Contact Information</th>
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<tbody>
<tr>
<td>Austria</td>
<td>Varian GesmbH, A-2334 Vosendorf, Austria</td>
</tr>
<tr>
<td>Belgium</td>
<td>NV Varian Belgium SA, 2860 Sint-Katelijne-Waver</td>
</tr>
<tr>
<td>Bulgaria</td>
<td>Spectratech, BG-1407 Sofia</td>
</tr>
<tr>
<td>Croatia</td>
<td>AnAs - Analytical Instruments, HR-10010 Zagreb, Croatia</td>
</tr>
<tr>
<td>Czech Republic</td>
<td>Amedis spol. S.R.O., HR-10010 Zagreb, Croatia</td>
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<th>Country</th>
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</thead>
<tbody>
<tr>
<td><strong>Czech Republic</strong></td>
<td>Charkovska 14</td>
<td>+420 2 748 191</td>
<td><a href="mailto:amedis@mbox.vol.cz">amedis@mbox.vol.cz</a></td>
</tr>
<tr>
<td><strong>Czech Republic</strong></td>
<td>CZ-10100 Praha 10</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Denmark</strong></td>
<td>Analytical Instruments AS</td>
<td>+45 44 35 02 02</td>
<td><a href="mailto:ai@analytical.dk">ai@analytical.dk</a></td>
</tr>
<tr>
<td><strong>Denmark</strong></td>
<td>Kirke Värlösevej 14</td>
<td>+45 44 47 10 32</td>
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<tr>
<td><strong>Denmark</strong></td>
<td>DK-3500 Värlöse</td>
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<tr>
<td><strong>Finland</strong></td>
<td>Tamro MedLab Oy.</td>
<td>+358 2044 5120</td>
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<td><strong>Finland</strong></td>
<td>P.O. Box 11</td>
<td>+358 2044 54970</td>
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<td><strong>Finland</strong></td>
<td>FIN-01641 VANTAA</td>
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<tr>
<td><strong>France</strong></td>
<td>Varian S.A.,</td>
<td>+33 16 986 38 38</td>
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<td><strong>France</strong></td>
<td>Quartier de Courtaboeuf,</td>
<td>+33 16 928 2308</td>
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<td>B.P. No. 12</td>
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<td><strong>France</strong></td>
<td>F-91941, Les Ulis, Cedex,Orsay</td>
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<tr>
<td><strong>France</strong></td>
<td>FRANCE.</td>
<td>+33 78 908212</td>
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<tr>
<td><strong>France</strong></td>
<td>Societe Varian</td>
<td>Telex: 380898</td>
<td></td>
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<tr>
<td><strong>France</strong></td>
<td>2 rue de Savoie, B.P. 607</td>
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<tr>
<td><strong>France</strong></td>
<td>69804 Saint Priest, Cedex,..</td>
<td></td>
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<tr>
<td><strong>Germany</strong></td>
<td>Varian Deutschland GmbH,</td>
<td>+ 49 6151 7030</td>
<td><a href="mailto:Andreas.Stroh@de.varian.com">Andreas.Stroh@de.varian.com</a></td>
</tr>
<tr>
<td><strong>Germany</strong></td>
<td>Alsfelder Strasse 6, Postfach ll 14 35,</td>
<td>+49 6151 703 237</td>
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<tr>
<td><strong>Germany</strong></td>
<td>D64289 Darmstadt</td>
<td>Telex: 419429</td>
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<td><strong>Greece</strong></td>
<td>FAMAR S.A.,</td>
<td>+30 1 988 9200</td>
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</tr>
<tr>
<td><strong>Greece</strong></td>
<td>7, P. Marinopoulou Str</td>
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<td>Alimos -17456</td>
<td>Telex: 224946</td>
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<tr>
<td><strong>Greece</strong></td>
<td>Athens</td>
<td>Fax: +30 1 988 9222</td>
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<td><strong>Hungary</strong></td>
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<tr>
<td><strong>LAB-EX</strong></td>
<td>Phone: +36 1 206 2455</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hadak utja 41, H-1119 Budapest</td>
<td>Fax: +36 1 206 2451</td>
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</tr>
<tr>
<td>Email: <a href="mailto:labex@mail.matav.hu">labex@mail.matav.hu</a></td>
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</tr>
<tr>
<td>P.O.Box 909</td>
<td>Fax: +354 562 8435</td>
</tr>
<tr>
<td>121 Reykjavik</td>
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<tr>
<td><strong>JVA Analytical Ltd</strong></td>
<td></td>
</tr>
<tr>
<td>Unit 1, Longmile Business Centre</td>
<td>Phone: +353-1-4569822</td>
</tr>
<tr>
<td>Longmile Road</td>
<td>Fax: +353-1-4569825</td>
</tr>
<tr>
<td>Dublin 12</td>
<td>E-mail: <a href="mailto:jva@iol.ie">jva@iol.ie</a></td>
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<tr>
<td><strong>Varian S.p.A.</strong></td>
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<tr>
<td>Via Fratelli Varian 54</td>
<td>Fax: +39 11 997 9344</td>
</tr>
<tr>
<td>10040 Leini</td>
<td>Telex: 310099</td>
</tr>
<tr>
<td>Torino Italy</td>
<td>Email: <a href="mailto:gianni.guida@it.varian.com">gianni.guida@it.varian.com</a></td>
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<td>Phone: +31 0164 282800</td>
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<tr>
<td>Analytical Instruments</td>
<td>Fax: +31 0164 282828</td>
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<tr>
<td>Boerhaaveplein 7, 4624 VT Bergen op Zoom</td>
<td>Email: <a href="mailto:varian.benelux@nl.varian.com">varian.benelux@nl.varian.com</a></td>
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<td>Liakollveien 1</td>
<td>Fax: +47 22 61 10 30</td>
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<tr>
<td>Postboks 122 Holmlia</td>
<td>E-mail: <a href="mailto:post@holger.no">post@holger.no</a></td>
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<td>Yetimhane Sokak 12/5</td>
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<td>28, Manor Road,</td>
</tr>
<tr>
<td></td>
<td>Walton-on-Thames,</td>
</tr>
<tr>
<td></td>
<td>Surrey KT 12 2Qf</td>
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<td>England</td>
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<tr>
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<td>The Genesis Centre,</td>
</tr>
<tr>
<td></td>
<td>Birchwood Science Park South</td>
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<td></td>
<td>Warrington (Cheshire), England</td>
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<tr>
<td>Yugoslavia</td>
<td>Global Technology Assistance</td>
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Note that the phone/fax numbers are in the format: + AA BB number, where the '+' refers to the international dialing code for your country, 'AA' is the country code and 'BB' is the area code (without any leading zeros).

- **Bahrain**
- **Iran**
- **Israel**
- **Jordan**
- **Kuwait**
- **Saudi Arabia**
- **Syria**
- **United Arab Emirates**

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<tr>
<td><strong>Applied Science Co., Ltd</strong></td>
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</tr>
<tr>
<td>Office 4, Bldg. 26</td>
<td>Phone: +98 21 878 7710</td>
</tr>
<tr>
<td>Didar Street, Africa Ave.</td>
<td>Telex: 88222341</td>
</tr>
<tr>
<td>Tehran - 15188, (P.O. Box 15175-161)</td>
<td>Fax: +98 21 888 8675</td>
</tr>
<tr>
<td>Email: <a href="mailto:asc@vessal.com">asc@vessal.com</a></td>
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<tr>
<td><strong>Israel Scientific Instruments</strong></td>
<td>Phone: +972 3 923 2202</td>
</tr>
<tr>
<td>32 Shacham Street</td>
<td>Fax: +972 3 922 9750</td>
</tr>
<tr>
<td>Amargad Building</td>
<td>Email: <a href="mailto:isirg@netvision.net.il">isirg@netvision.net.il</a></td>
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<tr>
<td>Kiryat Matalon</td>
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<tr>
<td>Petah-Tikva 49170</td>
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<td><strong>The Arab Drug Store</strong></td>
<td>Phone: +962 6 553 8981</td>
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<tr>
<td>Amin Shocair PH. C.</td>
<td>Fax: +962 6 553 2358</td>
</tr>
<tr>
<td>P.O. Box No. 208</td>
<td>Email: <a href="mailto:ads@ads.com.jo">ads@ads.com.jo</a></td>
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### Kuwait

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<tr>
<td>Gulf Scientific Corporation</td>
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<tr>
<td>c/o Al-Arfaj Engineering Co.</td>
<td>Telex: 22841</td>
</tr>
<tr>
<td>P.O. Box No 391</td>
<td>Fax: +965 481 7442/3</td>
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<tr>
<td>Salmiyah 22004</td>
<td>Email: <a href="mailto:gsckwt@ncc.moc.kw">gsckwt@ncc.moc.kw</a></td>
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### Saudi Arabia

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<tbody>
<tr>
<td>Gulf Scientific Corporation</td>
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<tr>
<td>c/o Dar Establishment</td>
<td>Fax: +966 3 858 1479</td>
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<tr>
<td>P.O. Box 5230</td>
<td>Email: <a href="mailto:gscsaudi@sol.net.sa">gscsaudi@sol.net.sa</a></td>
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### Syria

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<tr>
<td>Mimosa, 18, Eid Bldg.</td>
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<tr>
<td>Kasr Al-Dyaja Place</td>
<td>Fax: +963 11 333 0512</td>
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<tr>
<td>Abou Roumaneh</td>
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<tr>
<td>P.O. Box No. 5098,</td>
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### United Arab Emirates

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<td>Gulf Scientific Corporation</td>
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<td>Fax: +971 4 816 778</td>
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<tr>
<td>Jebel Ali</td>
<td>Email: <a href="mailto:gsc@emirates.net.ae">gsc@emirates.net.ae</a></td>
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<tr>
<td></td>
<td>Telex: 48150 GSC JA</td>
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### Varian Offices and Dealers—North America

Note that the phone/fax numbers are in the format: + AA BB number, where the '+' refers to the international dialing code for your country, 'AA' is the country code and 'BB' is the area code (without any leading zeros).

- **Canada**
- **Mexico**
- **Trinidad/Tobago**
- **U.S.A.**

### Canada

<table>
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<tr>
<td>Varian Canada Inc.</td>
<td>Toll free phone: 1 800 387 2216</td>
</tr>
<tr>
<td>6705 Millcreek Dr, Unit 5</td>
<td>Phone: +1 905 819 8181</td>
</tr>
<tr>
<td>Mississauga, L5N 5R9</td>
<td>Fax: +1 905 819 8348</td>
</tr>
<tr>
<td>Ontario.</td>
<td>Email: <a href="mailto:dan.williams@varianinc.com">dan.williams@varianinc.com</a></td>
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### Varian Offices and Dealers—Central and South America

Note that the phone/fax numbers are in the format: +AA BB number, where the '+' refers to the international dialing code for your country, 'AA' is the country code and 'BB' is the area code (without any leading zeros).

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<td>+52 5 5145383 or +52 5 523 9472</td>
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<td>Int'l ph: +1.925.939.2400</td>
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<tr>
<td>Varian Argentina LTD (from June 16th, 1997)</td>
<td>Sucursal Argentina, Av.Ricardo Balbin 2316 1428 Buenos Aires</td>
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<td>Phone: +54 11 4783 5306</td>
<td>Fax: +54 11 4786 5172</td>
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<tr>
<td>Email: <a href="mailto:varian@tournet.com.ar">varian@tournet.com.ar</a></td>
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<tr>
<td>Arellano Limitada</td>
<td>Avenida 6 de Agosto No. 2240 Edificio Maria Cecilia Mezzanine No. 4</td>
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<tr>
<td>Phone: +591-2-352502</td>
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<td>email: <a href="mailto:arellano@latinwide.com">arellano@latinwide.com</a></td>
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<tr>
<td>Varian Industria e Comercio Ltda.,</td>
<td>Avenida Dr. Cardoso de Mello, 1644</td>
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<tr>
<td>Phone: +55 11 820 0444</td>
<td>CEP 04548005</td>
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<tr>
<td>Telex: 55 11 30405</td>
<td>SAO PAULO, S.P., BRAZIL</td>
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<tr>
<td>Fax: +55 11 820 9350 or +55 11 820 3449</td>
<td>Email: <a href="mailto:vlopes@amcham.com.br">vlopes@amcham.com.br</a></td>
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<td>Phone: +56 2 340 0000</td>
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<td><a href="mailto:p.labarrera@merck.cl">p.labarrera@merck.cl</a></td>
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<td>Walter Rothlisberger Co., LTDA</td>
<td>Apartado Aereo 06803</td>
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<tr>
<td>Phone: +57 1 283 3132</td>
<td>Calle 26-A No. 13-A-27</td>
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<td>Telex: 43409</td>
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<td>Phone: + 506 226 9502 / 226 9402</td>
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<td>P.O. Box 434</td>
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<td>SANTO DOMINGO, DOMINICAN REPUBLIC.</td>
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<tr>
<td>Científicos</td>
<td>Phone: +593 2 453178</td>
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<td>P.O. Box No. 17-08-8583 Sucursal 8</td>
<td>Fax: +593 2 442051</td>
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<tr>
<td>(Servicio y Equipo Medico</td>
<td>Phone: +507 251247 or + 507 270537</td>
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<td>Eberhard Lewkowitz S.R.L.</td>
<td>Phone: +595 21 444400/1/2</td>
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<tr>
<td>Casilla De Correo 523</td>
<td>Telex: 378 PY</td>
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<tr>
<td>ASUNCION, PARAGUAY</td>
<td>Fax: +595 21 444437</td>
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<tr>
<td>Av. Los Frutales nbr 220</td>
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<td>LIMA 3, PERU.</td>
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<td>Varian Technologies C.A. (VTCA)</td>
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<tr>
<td>Multicentro Paseo El Parral, Piso 3</td>
<td>Phone: +58 41 257 608</td>
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<td>Oficinas 3.1 Y 3.2 - El Parral</td>
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<tr>
<td>Avenida Rio Orinoco</td>
<td>email: <a href="mailto:manez@telcel.net.ve">manez@telcel.net.ve</a></td>
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Troubleshooting

The links below display various lists of frequently asked questions from Cary Eclipse users. Hopefully the answer to your problem is here.

- Hardware Frequently Asked Questions
- Software Frequently Asked Questions
Troubleshooting—Hardware

The links below display various lists of frequently asked questions from Cary users. Hopefully the answer to your problem is here.

Hardware Frequently Asked Questions

- Instrument error messages
- Accessories

If you cannot find the answer in any of the lists above, you still have plenty of options available to you to solve the problem.

- You can call your local Varian office or Dealer and arrange a service call.
- If you purchased the Telediagnostics capability from Varian then you can arrange for a Telediagnosis by Varian. You can find out more about the Telediagnostics package on our Website.
- You can visit Varian's website.
- You can contact the factory where your Cary instrument was made.
**Instrument error messages**

**Q.** What should I do if I get an error message that doesn't recommend a course of action?

**A.**
1. Note the error number.
2. Close the Cary Eclipse software.
3. Turn the Cary Eclipse instrument off.
4. Reboot the computer.
5. Turn the Cary Eclipse instrument on.
6. Restart the Cary Eclipse software.

If the error reoccurs, you may need to contact Varian and notify us of the error number and the circumstances that it occurs under. Ideally, you should:

Save the current method, data and report as a Batch file and send an email to fluorescence@varianinc.com which includes the following information:

- The error number
- Details of any accessories you may have installed in the instrument
- The steps you took to produce the error (if known)

You should also attach a copy of the batch file to the email.
General

Q. I cannot see an OK or Cancel button on a dialog so I cannot set up my method. What do I do?
A. You have your screen resolution set too low. Right-click on the Windows Desktop, select Properties and select the Settings Tab. Change the Desktop Area setting to 800 x 600. Make sure that the Color Palette is set to High colour (16 bit).

Q. I cannot find how to set the correct operating (abscissa) mode for my run. How do I set this?
A. Check that you are using the correct application. For example, time-based measurements are performed in the various kinetics applications.

Q. How do I generate a report from an existing file?
A. Press the Recalculate button and make changes as required in the dialog that appears. When you have finished, use the Print button or the Print command in the File menu to print your new report. You can view the report on screen before printing by using the Print Preview command in the File menu.

Q. I want to export my data to a spreadsheet file. When I press Recalculate, the Autoconvert options are grayed out. How do I do this?
A. You need to use the Save Data As option in the File menu. In the dialog that appears, set the Files of type field to Spreadsheet ASCII (csv) and your data will be exported. You can also refer to ‘How to export collected data’ in your application’s Help.

Q. I have saved my sample file, but now I cannot find it in the directory. What has happened?
A. You may have saved the file as a different file type, for example as a Data file rather than a Batch file. To see all the files in the directory, select Open in the File menu and set the Files of type field to All Files.

Q. How do I include details of the method when I export data to a spreadsheet file?
A. Select the option Select for ASCII (csv) with Log in the Autoconvert group on the Reports tab of the Setup dialog.

Q. The Cary Eclipse software is running slowly, how do I get it back to normal?
A. You may have too many traces available in the Trace Preferences dialog. Delete any unwanted traces, and the speed of your application will increase. See ‘How to remove a trace from the trace list’ in your application’s Help.

Q. I have typed in an ADL program, but it is not carried out when I press the Start button. How can I apply my ADL program to a data collection?
A. You need to use the User Collect option that is available in some applications on the Cary tab of the Setup dialog. Enter your ADL commands in the User Result line that appears when you activate User Collect.

Q. I need to store information about a trace and print it in my report e.g. sample preparation details such as pH or the reagents used. How do I do this?
A. Use the User Data Form feature. This can be accessed from the Graphics menu or from the toolbar.
Starting a run

Q. I have a ‘Connect’ button instead of a ‘Start’ button. What does this mean?
A. Only one Cary Eclipse application can communicate with the instrument at a time. You have another Cary Eclipse application open that is online with the Cary Eclipse instrument. Provided that the other application is not collecting data, you can press Connect to make this application go online and the Start button appear.

Q. I only have one application running, yet I am still seeing a Connect button instead of a Start button. Why?
A. The Cary Eclipse instrument is still initializing and the System Information application has control over it. When the initialization has finished, your chosen application will have control and the Start button will become active.

Q. I have set up a method and want to press Start, but the Start button is grayed out. Why is this?
A. You may be using a thermostattable accessory, and the Cary Eclipse is waiting for this to reach the required temperature before it allows you to commence the run.

Q. Why do I sometimes get asked twice for a sample name when I press Start?
A. Storage on (Prompt at start) may be selected in the Autostore page of the Setup dialog. The first prompt by the Cary is for the batch file name into which your data will be stored. The second prompt is for the name of the trace within the batch file. This appears in the Trace Preferences dialog and in your report. You cannot change this name once it is entered.

Q. I need to set up the Cary Eclipse to perform a simple calculation during the run. How do I do this?
A. You need to use the User Collect option that is available in most applications on the Cary page of the Setup dialog. Enter your ADL commands in the User Result line that appears when you activate User Collect.

Q. How do I store information about a trace and print it in my report e.g. sample preparation details such as pH or the reagents used?
A. Use the User Data Form feature. This can be accessed from the Graphics menu or from the Graphics toolbar.
Graphics

General

Q. I accidentally deleted all my traces from my graph box by pressing Clear All Traces. Can I get them back?
A. Yes, open the Trace Preferences dialog (via the Graph menu or the toolbar button) and then mark the traces you require in your graph as Visible.

Q. I can find a Zoom Out button, but not a Zoom In button. How do I enlarge an area of the screen?
A. You need to use the Free Cursor (accessed from the Cursor Mode option in the Graph menu or by selecting the toolbar button) and then click and hold down the left mouse to create a box around the area of interest. See ‘How to zoom a graph’ in your application’s Help.

Q. Why does choosing to display my traces in different line formats have no effect?
A. The different line formats will only alternate when the Trace Width has been selected at 1 pixel. Style cycling and Trace Widths are selected in the Graph Preferences dialog accessed from the Graph menu or by selecting the toolbar button.

Q. When I autoscale a graph with several traces in it some of the traces are drawn off the edge of the graph box. How do I autoscale them all?
A. The autoscale function uses the currently focused trace to autoscale. Click on the trace which has the most variation in terms of Y-scale and then use the autoscale function.

Q. It is frustrating to set up axes colors and widths etc in the Graph Preferences dialog box, only to find that next time the instrument is turned on they are lost. Is there a way to save these settings?
A. To store the Settings from the Graph Preferences dialog, such as axes color, and width, so that the system remembers these settings every time an application is opened you will need to do the following:

1. Open the application of interest
2. Add a graph to the graphics area
3. Open the graph preferences dialog and setup all your graph settings
4. From the file menu select the Save Data As menu and save your settings as a batch file. For example ‘Kinetics Default settings.bkn’.
5. Create a shortcut on the desktop for this batch file.
6. Next time you want to open this application, rather than selecting the icon in the Cary Eclipse folder on the desktop, select the shortcut (e.g. ‘Kinetics Default settings.bkn’) icon instead and you will get your graph preferences load automatically.

Note: This needs to be done separately for each application.

Graph Labeling

Q. Why can’t I add a label to my graph?
A. You may not have selected the graph box. You need to first click on the graph box you require to select it.
A black line with eight 'handles' will appear around the box. There must also be at least one trace Visible in the graph box before you can add a label to it.

**Q.** How can I store information with my graph so I can print it every time I run a sample? i.e. I do not want to add labels to every sample trace.

**A.** The solution is to add the sample information to the graphics background and then save this as a graphics template. See ‘How to add a label to a graph’ in your application’s Help.

**Q.** When I use the peak label function every peak in my trace is labeled. I only want the larger peaks to be tagged. How do I do this?

**A.** You need to increase the Peak Threshold setting. To do this for an existing graph, press the Recalculate button and select the button Peak Information in the Reports tab. The Peak Labels dialog will appear. Click in the Peak Threshold field and increase the value that appears there. Click OK to close the dialog and then OK to start the recalculation. Your graph will now appear with different peaks labeled.

**Q.** After I have used the autoscale function some of my peak labels are off the screen. How do I see them?

**A.** You need to manually rescale the Y axis of your graph. See ‘How to scale your graph manually’ in your application’s Help.

**Q.** One of the labels is in the wrong spot. How do I move it?

**A.** Simply click the left mouse button on the label to select it (a box will appear around the label), then click and hold down the left mouse button within the box. Move the mouse up or down or left or right to move the label.

**Q.** I can’t select a label, why not?

**A.** Sometimes, a label may be hard to select as it is partially obscured by a picture or another label. In this case, you may need to enlarge the area in question by zooming in. Alternatively, try right-clicking on the label, especially if it is over a trace.

**Q.** I can find a Zoom Out button, but not a Zoom In button. How do I enlarge an area of the screen so I can select a label?

**A.** You need to use the Free Cursor mode and then click and hold down the left mouse to create a box around the area of interest. See ‘How to zoom a graph’ in your application’s Help.

**Q.** I need to store additional information about each trace in my batch file and print it in my report. Is there some way to do this without using Add Label?

**A.** Yes, use the User Data Form feature. This can be accessed from the Graphics menu or from the toolbar.

**Q.** I would like to change the names of my sample traces. Can I do this?

**A.** No, the name you entered as the sample name when you collected the data is the permanent name.
File Handling

Q. I have saved my sample file, but now I cannot find it in the directory. What has happened?
A. You may have saved the file as a different format, for example as a Data file rather than a Batch file. To see all the files in the directory, select Open in the File menu and set the Files of Type field to All Files.

Q. What is the difference between a data file and a batch file?
A. A data file contains only collected traces. A batch file contains collected traces, the method by which the traces were collected, a graphics template, and a report on the data.

Q. How can I overlay data from two batch files?
A. You need to open the first batch file, make all the traces Visible (in one or more graph boxes), and then save the traces as a data file (*.Dxx) rather than a batch file (*.Bxx). Next, open the second batch file and then open the data file you stored in the first step.

Q. I have multiple graph boxes with multiple traces in each. However, I only want to save the traces in one graph box. How do I do this?
A. Use the Remove Graph button to delete all the other graph boxes from the screen, and then use the Save Data As option to save the files in the remaining box. Make sure that the Save Only Focused Trace option is not selected or you will only save the red trace.

Q. I use the same User Collect statement in a number of different methods. Can I store this?
A. Yes, you can store User Collect statements on the Edit tab of the System Information application.

Q. I need to store information about a trace and print it in my report e.g. sample preparation details such as pH or the reagents used.
A. Yes, use the User Data Form feature. This can be accessed from the Graphics menu or from the toolbar.

Q. I had several traces on the screen and thought that I saved them all. When I retrieve the file I only get one trace—where are the others?
A. When you saved the file, Save only focused trace at the bottom of the Save as dialog must have been selected. Make sure that this option is not selected if you want to save all the displayed traces. You can not retrieve the traces that you did not save.
Exporting Data

Q. I want to export my data to a spreadsheet file. When I press Recalculate, the Autoconvert options are grayed out. How do I export the data?
A. You need to use the Save Data As option in the File menu. In the dialog that appears, set the Files of Type field to Spreadsheet ASCII (csv) and your data will be exported. You can also refer to ‘How to export collected data’ in your application’s Help.

Q. I want to export method data with my ASCII file when I collect the data. How do I do this?
A. Check the option Select for ASCII (csv) with Log in the Autoconvert group on the Reports tab before pressing Start.

Q. Why don't I see my graphics in the Report area?
A. The Report area normally only shows the text generated by the reporting options. If you want to view a copy of your report with graphics before you print it, make sure that you have the Graph option selected in the Reports tab, and then use the Print Preview option on the File menu. Alternatively you can use the Copy Graph command to copy the Graphics area, and then use Edit report, then Paste, to Paste the graph into the Report area.

Q. The Cary Eclipse software refuses to open a CSV file, in fact it stops working.
A. Check that the CSV file is using a period '.' to indicate a decimal point and not a comma ','. The numbers in the XY pairs should be separated by a comma ','. 


Maths Calculator

Q. My Maths Calculator just does not work properly. What is wrong with it?
A. Your number format in your Window's country settings may be causing problems. To check this, double-click on the Regional Settings option in your Windows Control Panel. Click the Number tab. Make sure that the Decimal Symbol field is set to '.' and the Digit Grouping Symbol is set to ' ' (space).

Q. I have created a calculation in the Maths Calculator and selected Apply, but my graph doesn't change. Why not?
A. You need to press the '=' button to perform the calculations on your graph. The Apply button adds the displayed mathematical expression in the Operation field to the calculation that will be performed on your graph. (The list box at the top of the Maths window displays all the operations that will be performed.)

Q. I have accidentally selected Apply, can I undo this?
A. Yes, select the CE button. This undoes the last 'Apply'.

Q. I want to delete the current mathematical expression in the Maths window and do a new calculation. How do I do this?
A. Click the Clear (C) button.

Q. How do I subtract a trace in one data file from another trace in a different data file? I seem to only be able to open one data file at a time.
A. If you can only open one data file at a time, it is likely that you are trying to open two different batch files. You need to first save the two traces that you are trying to subtract as data files from the Save As Data command in the File menu. Set the Files as Type option to Data (*.Dxx) (xx is the two-letter designation used by your application.). Once both files are saved as data files you will be able to display them both on screen. The subtraction procedure is described in 'How to perform a spectral subtraction' in the maths module.'
User Data Form

Q. I entered lots of information into the User Data Form and saved the file. But when I re-open the file, the information is gone. What have I done wrong?

A. You have forgotten to add a heading to the column(s) in the User Data Form. Entries in columns without a heading are not saved. Use this form like a spreadsheet rather than like a note pad.
Recalculation and Reports

Q. I just want to change my report, do I still press the Recalculate button?
A. Yes, but if more than one tab appears in the dialog, just make changes on the Reports tab. When you have finished, use the Print button or the Print command in the File menu to print your new report.

Q. I need to print additional information on each sample in my report. Is there some way to do this?
A. Yes, use the User Data Form feature. This can be accessed from the Graphics menu or from the toolbar.

Q. I can edit a report and change the data—surely this should not be possible?
A. If you do not want to allow any editing of reports, you need to set up GLP Administration of the application. From the Home page of the Help, select Software applications > GLP Admin.

Q. How do I put page breaks in a report?
A. The easiest way is to use the Edit report option and cut and paste the report into a Word processor.

Q. Why is the option Edit Report in the Edit menu not available?
A. The application is under GLP Administration control, and editing of reports is not allowed.

Q. Why isn't the logo I selected included in my report?
A. If you are running the Cary Eclipse software under Windows NT you need to be logged on as an Administrator in order to change the logo in the System information application. Once this is done you can use the system under your normal login ID.

Q. I thought that I had generated a report but I can't see it on the screen, where is it?
A. Double-click on the graph area to display the report beneath the graph. You can double-click again to remove the report.

Q. Why can't I see a picture of my graphs in the Reports section? I opted to include the graph in the report.
A. The Reports section displays only text information, not graphics. Use the Print Preview option on the File menu to view the whole report on screen before printing it.
Glossary

Absorbance (also Optical Density)

The amount of light that is absorbed by a sample can be expressed as absorbance.

\[ A = -\log_{10} \left( \frac{I}{I_0} \right) \]

Absorptivity (also Extinction coefficient or Molar absorptivity)

The proportionality factor in the Beer-Lambert law, which states that the absorbance of a solute is directly proportional to its concentration and the length of the light path through the cell, is called Extinction coefficient.

Actinometry

The measurement of intensity of radiant energy. Chemical actinometers are substances that respond to irradiation in some measurable way that can be related to the intensity of the irradiating source. For example, potassium ferrioxalate, when complexed with 1:10 phenanthroline, increases linearly in absorbance at 510nm as a function of irradiating time.

Activation energy (Ea)

The minimum energy needed to be overcome in a reaction to make it occur.

Analyte

An analyte is a component of a sample that has a measurable characteristic. Normally, an analyte is a chemical component, whose concentration is calculated by comparing a measured response (e.g. absorbance) with a calibrated response curve for the analyte.

Arrhenius plot

A graph to determine the activation energy of a system. The natural log of the rate constant, \( \ln(k) \), is plotted against the reciprocal of the temperature in Kelvin, \( 1/T \), with the slope being equal to the \( \text{Ea}/R \), where \( R \) is the gas constant (8.314 JK-1mol-1).

Assay

The determination of the concentration of an analyte in a sample.
Bioluminescence

Refer to Chemiluminescence

Calibration

Calibration is the determination of the instrument response (usually absorbance) to various concentrations of the analyte. A calibration is usually performed by measuring a series of standards (samples with known concentrations or path lengths) and then fitting a calibration curve.

Calibration curve

The calibration curve is the fitted model (e.g. a straight line) to the function results (e.g. absorbance at a specific wavelength) at different concentrations of the standard. The graph showing the fitted curve and the used data points is also called calibration curve. Note that the theoretically best calibration curve is a straight line, according to the Beer-Lambert law.

Chemiluminescence

Chemiluminescence occurs when a chemical reaction produces an electronically excited species which emits a photon in order to reach the ground state. These sort of reactions can be encountered in biological systems; the effect is then known as bioluminescence.

Color

Color can be defined as the perception or subjective response by an observer to the objective stimulus of radiant energy in the visible range of the spectrum (400 nm to 700 nm). The perceived color depends on the absorptive and reflective properties of the object, the energy distribution of the source of illumination and the visual sensitivity of the observer. For the measurement of color, see Colorimetry.

Colorimetry

Colorimetry has two different meanings:
  i) It is the determination of the concentration of a colored solution by comparing it visually with the color of a standard solution. By the use of a photometric system the comparison can improved and extended to solutions that are apparently colorless.
  ii) It is the characterization of the color by a spectrophotometric measurement and calculation of, for example, tristimulus values (X, Y and Z) for the color.

Combinatorial chemistry

A technique of creating vast number of compounds by reacting a set of starting chemicals in all possible combinations.

Consecutive reactions

these are reactions that proceed one after each in sequence, eg A ® B® C® D.
**Content uniformity**

Content uniformity is the uniformity of the active ingredient in a preparation. A certain number of units are individually assayed and the statistics of the results must meet criteria of an acceptance table.

**Correlation coefficient**

The correlation coefficient ($R^2$) is calculated for the calibration curve. The correlation coefficient is a common measure of the exactness of the fit; a perfect calibration curve has a value of 1.

**Data interval**

The setting which fixes the interval between wavelength/ wavenumber readings when performing a wavelength or wavenumber scan.

**Derivative spectrum**

A derivative spectrum is obtained by the calculation of the first or higher derivatives of the absorbance with respect to the wavelength. The first derivative is the rate of change of absorbance against the wavelength.

**DNA melt**

Many biological molecules (proteins, DNA) are very large and have a native three-dimensional state. This three-dimensional state is maintained partly by the stereochemistry of chemical bonding and partly by weak intramolecular hydrogen bonding. On heating, the weak bonds can break and the molecule loses some or all of its three-dimensional structure. This is the process of thermal denaturation.

**Emission**

The release of light from a fluorochrome when an electron falls from an excited state to a lower energy state of the molecule.

**Emission spectrum**

A plot of emitted light wavelength versus the relative intensity of emitted light. Essentially a plot of wavelength versus the probability that the emitted light will have the energy of that wavelength.

**Energy of a photon of light**

\[ E = \frac{hc}{l} \]

where "h" is Planck's constant, "c" is the speed of light, and "l" is the wavelength of the light.
Energy transfer

The transfer of energy between molecules in an excited state.

Enzyme activity

In most preparations, the actual molar concentration of enzymes is unknown. Consequently, the amount of enzyme present can be expressed only in terms of its activity. To standardize the reporting of enzyme activities, the Commission on Enzymes of the International Union of Biochemistry has defined a standard unit:

One International Unit (1 U) of enzyme is that amount which catalyzes the formation of 1 µmole of product per minute under defined conditions.

The concentration of enzyme in an impure preparation can be expressed in terms of units/ml and the specific activity of the preparation as units/mg protein. As an enzyme is purified, the specific activity will increase to a limit (that of the pure enzyme).

Enzyme assay

Enzyme assays are an indirect method of analyzing one compound or group of compounds in a complex matrix. By careful selection of the enzyme, it will be known that whatever change occurs to the sample after the addition of the enzyme is due only to reaction of the specific compound or compounds. This selectivity is the basis of enzyme assays.

Enzyme assays can be divided broadly into two types: rate assays and end-point assays.

With rate assays all other parameters that have an influence on the rate are controlled at a constant level, the rate of reaction will then be directly proportional only to the substrate concentration.

With end-point assays the conditions are selected so that the conversion of substrate to product is complete within a reasonable period of time (5 to 20 minutes). The difference between initial absorbance and final absorbance is directly proportional to the amount of substrate.

Enzyme mechanisms

Enzyme mechanistic studies use rate information obtained from kinetic measurements to determine the mechanism by which an enzymatic reaction proceeds. Mechanisms are determined by studying the effect of variations in substrate concentration and inhibitors on the initial rate of reaction.

Excitation

The absorption of light energy by a fluorochrome, during which an electron in the fluorochrome molecule is boosted to a higher energy level.

Excitation spectrum

A plot of incident-light wavelength versus the total fluorescence emitted. Essentially a plot of incident wavelength versus the probability of excitation.

Excited state

The electronic state reached by a molecule upon the absorption of energy. This state is usually more reactive than the ground state and so the molecule undergoes some process to get rid of this excess energy, such as a photochemical reaction.

Exponential decay
A mathematical expression describing the decay of an unknown or variable, such as concentration, as a function of the exponent, e.

**Extinction coefficient (e)**

The amount of light absorbed. The molar extinction coefficient is the optical density of a one-molar solution of the compound through a one-cm light path. The value usually quoted is the molar extinction coefficient at the wavelength of maximum absorption.

**First-order reaction**

A chemical process whose rate of reaction depends on the concentration of only one reactant raised to the power of one.

**Fluorescence**

The emission of light (or other electromagnetic radiation of longer wavelength) by a substance as a result of absorption of other radiation. The emission continues only as long as the stimulus producing it continues and persists with a half-life of less than about 10^-8 second

**Fluorochrome**

A fluorescent dye

**Ground state**

A most stable state in which a molecule can exist.

**Half-life**

The time required to affect 50% of the reactant

**Homogeneous**

A system that consists of a single phase, uniform mixture
- I -

**Intensity of light**

The flow of energy per unit area. Intensity is a function of the number of photons per unit area and their energy (energy of a photon is inversely proportional to wavelength).

**Irradiating light source**

The source used to irradiate a molecule to initiate a photochemical or photophysical process.

**Irradiating wavelength**

The wavelength at which a molecule is irradiated to produce a photochemical or photophysical process. This wavelength is specific to each molecule and depends on the absorption spectrum of the molecule.

**Isosbestic point**

The point in which absorbance vs wavelength spectra intersecting each other, produces no change in absorbance value. All spectra cross at this one point.

- J -

(EMPTY)

- K -

**Kinetics**

Kinetics is concerned with the rate at which chemical or physical processes occur. The goal of kinetic studies is usually to determine an equation which describes the rate in terms of component concentrations and, optionally, other factors such as temperature, surface area, etc.

- L -

**Luminescence**
An emission of light occurring at a temperature below that of incandescent bodies. Luminescence includes phosphorescence and fluorescence. Luminescence comes about through the ability of certain substances to absorb light of relatively high frequency and re-emit it in installments of discrete lower frequencies.

- M -

Maximum

Refers to the maximum peak in an absorbance spectrum. The derivative at this point equal to zero.

Mechanism

The process, or series of processes, leading to a chemical reaction.

Melting curve

A melting curve is the change in absorbance with temperature when a DNA or other protein sample melts or undergoes denaturation.

Molar

A unit of concentration which is defined as the amount of substance, measured in moles, in 1 litre of solution.

Molar absorptivity

see Absorptivity

Mole

The fundamental unit in chemistry used to defined the amount of a substance.

Monochromatic light

Light consisting of only one wavelength or color.

Multicomponent analysis (also MCA)

Multicomponent analysis is the quantification of more than one component in a sample by UV-Visible spectroscopy and a mathematical deconvolution of the spectrum.

- N -

Natural bandwidth
Natural bandwidth is the width in nm at the half height (50% peak height) of an absorbance peak of a sample.

**Noise**

The Noise level is an indication of how stable a reading is. It determines the precision of the measurement and the detection limit of the instrument. Noise is measured at a particular wavelength and absorbance level. It is specified either as the peak To-peak value, (Maximum deviation on either side of the Abs value) or the RMS (root mean square) which is approximately one fifth of the peak-to-peak value. A poor signal to noise performance makes it very difficult to tell what the real Abs value is as it fluctuates. It introduces errors in to both quantitative and qualitative spectroscopy.

**Non-equilibrium**

The stage in which there is a continuous change in concentrations in a reacting system.

**Non-Measurement-Phase-Stepping**

Method of measurement used in the Cary instrument which allows measurement of the samples and reference beams at the same grating position (wavelength).

**Optical density**

see Absorbance.

**Order of reaction**

A classification of reactions according to their concentration relationships on the rate of reaction.

**Peak**

The maxima of an absorbance band are called peaks. Local peaks within an absorbance band are also called absorbance shoulders.

**Peltier effect**

The heating or cooling effect produced by passing an electric current through the junction of two dissimilar metals, the heating or cooling depending on the direction of the current. This is much more effective than using a water bath because the rate of change in temperature is faster and the stability is greater over a period of time.

**Phosphorescence**
The emission of light, occurring from the lowest triplet excited state of a molecule, as the result of absorbing radiant energy. This is emission is on a slower time scale than fluorescence, in the order of minutes to milliseconds.

**Photochemical reaction**

A chemical reaction that results from the absorption of light.

**Photo-degradation**

The depletion of a molecule of compound as the result of the absorption of light.

**Photometric accuracy**

How accurately the instrument measures. Measurement of photometric accuracy has traditionally been measured by the following methods: Chemical standards, such as K2Cr2O7 (accuracy ±0.005 Abs) Calibrated neutral density filters (accuracy is within 0.5 - 1% of their stated values or 0.002-0.004Abs). The double aperture method is by far the most accurate method. It is used by major national standards laboratories to measure the absolute accuracy of their reference spectrophotometers. This method has no limitations on SBW, wavelength or temperature and can yield precisions about 2 orders better than the previously mentioned uncertainties. The effects of poor photometric accuracy will be to produce incorrect results.

**Photometric linearity**

How accurately the instrument measures absorbance with increasing concentration. The measurement of photometric linearity is achieved by measuring the absorbance of successive K2Cr2O7 solutions of increasing concentrations, or by measuring the additive absorption of a series of filters. Poor photometric linearity will produce incorrect results.

**Photometric reproducibility**

The photometric reproducibility is a measure of the variability of the readings over multiple measurements of the same stable sample.

**Photomultiplier tube (PMT)**

A photoelectric cell that converts light into electric current and amplifies the current.

**Photon of light**

A quantum of light. This concept is based on Planck's quantum theory of light, which says that the energy of an oscillating system can have only discrete (quantized) values. These values are proportional to Planck's constant, h.

**Photophysical processes**

The events leading to the deactivation of a molecule in the excited state, with the exception of chemical reactions.

**Pseudo first-order**

A higher order reaction that has one or more reactants in excess, so that the rate of the reaction now depends on the concentration of only one reactant. The reaction now mimics first-order kinetics.
- Q -

Quantify

see Assay.

Quantum efficiency (quantum yield, $f$)

The efficiency at which a fluorochrome converts absorbed light to emitted light (the ratio of the number of photons emitted to the number of photons absorbed).

Quantum yield

A measure of the efficiency of a process. For a photochemical reaction, the quantum yield of photoproduct formation is the amount of product formed divided by the amount of light absorbed by the reactant.

- R -

Rate calculation

The rate of a reaction is the velocity of at which a chemical or physical process occurs. Usually an equation has to be determined which describes the rate in terms of changes in component concentrations. The simplest equation is the one for a zero order reaction, also called a slope, where the rate of the reaction is independent of the concentration of the reactants.

$$V = k$$

where $k$ is the rate constant.

Another common reaction equation is the first order reaction, where the rate of the reaction is directly proportional to the concentration of the reactant:

$$V = k \times [A]$$

The rate is calculated by a mathematical fit of the rate equation itself, or an integrated form of the equation, to the measured data.

Rate constant

The constant of proportionality in the expression relating the rate of a reaction to the concentration of the reactants.

Rate law

The empirical equation that describes the relationship between the concentration of reactants and the rate of a reaction.
Residual

The residual of a data point in a calibration curve is the difference between the actual analyte value and the calculated value when the point is fitted to the calibration curve.

Resolution

The spectrophotometer's ability to distinguish between two absorbance bands which are close together.

Second-order reaction

A chemical process whose rate of reaction depends on the concentration of two reactants. These reactants need not be different.

Side-reactions

Any unwanted reactions that occur during the investigation of a specific chemical process.

Signal averaging time

The amount of time the signal is averaged over for any particular data point.

Smooth

Smooth performs an n-point smoothing of a spectrum using Savitsky-Golay algorithm. For each data point in the spectrum, the calculation takes a set of data points (the filter length) around the current data point and fits a curve of a specified polynomial degree. The fitted curve is then used to calculate the new value for the current data point on the smoothed curve.

Spectral band width (SBW)

The physical slit width which is used to allow a narrow spectrum into the monochromator. SBW choice will affect the resolution of the spectrum obtained.

Stability, long term

The drift of the reading per unit time. Measurement is made by recording the Abs value of a sample at one wavelength for several hours. The specification indicates the maximum deviation from the correct Abs value per unit time. Poor stability introduces errors in the time base measurements. The instrument cannot distinguish between slope due to reaction or slope introduced by the instrument. baseline corrections can be invalid.

Steady state assumption (SSA)

The assumption that the change in concentration of a species with time is effectively zero. This occurs when the reagent is in large excess, such as in the case of pseudo first-order reactions.

Stoichiometric relationship
The relationship between the reactants and products in terms of the amount of compound, usually in moles, needed for reaction.

**Stokes shift**

The difference in wavelength between the apex of the excitation spectrum (shorter wavelength, higher energy) and the apex of the emission spectrum (longer wavelength, lower energy).

**Stopped-flow kinetics**

The investigation into rapid reactions occurring on a sub-second time scale using the stopped-flow technique. This requires specialized apparatus and a spectrophotometer that can acquire data at a rate of milliseconds to tens-of-milliseconds.

**Stray Light**

The amount of light reaching the detector which is of a wavelength other than that selected. Also called Stray radiant energy (RE). To measure stray light solutions are used which are non transmitting at the selected wavelength. (No light of the selected wavelength can reach the detector). Any observed transmittance is due only to stray light. The stray light level determines the maximum absorbance the instrument is able to measure and changes in band shape. Stray Light also causes deviations form the Beer-Lambert Law.

**Temperature probe**

A device used for measuring the temperature of the solution inside the cuvette.

**Thermal equilibration**

When the temperature of a solution remains constant to within the specifications of the heating accessory and measuring device. For the peltier systems, this is to within 0.05°C of the desired temperature.

**Thermal denaturation**

see DNA melt.

**Uniform reaction**

A reaction involving the clean inter-conversion of reactants to products. Only one reaction is occurring, with no consecutive or side reactions taking place.
Valley

The minima of an absorbance band are called valleys.

Wavelength accuracy

How close the indicated wavelength is to the actual wavelength. Measurement is made by scanning the D2 emission lines which are known to occur at 485.0 nm and 656.1 nm and observing the wavelengths at which these lines actually appear. Poor wavelength accuracy can produce low quantitative results as the Abs measurement may be on the side of the peak instead of at the maximum Abs.

Wavelength of light (l)

The distance in nanometers between nodes in a wave of light. The wavelength is inversely proportional to the energy of the light. \( l \propto \frac{1}{E} \).

Wavelength repeatability

The ability of the instrument to correctly return to the set wavelength repeatedly. Measurement is made by repeatedly scanning a filter peak. If the wavelength repeatability is good then the same trace should be obtained each time. Poor wavelength repeatability will result in errors in quantitative analysis due to wavelength shifts.