

### 3 Quick Start Guide

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This chapter provides information on running an Agilent 1260 Infinity II LC System.



## Best Practices

**NOTE**

For best practices, refer to the *Agilent Information Center* on Agilent InfinityLab LC Series User Documentation (G4800-64600) or the 1290 Infinity Pump Quick Reference Sheet (01200-90091).

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## Prepare a Run

This procedure exemplarily shows how to prepare a run. Parameters as shown in the screenshots may vary, depending on the system installed.

### WARNING

**Toxic, flammable and hazardous solvents, samples and reagents**

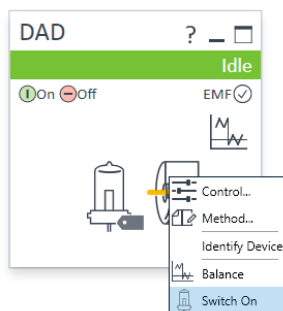
**The handling of solvents, samples and reagents can hold health and safety risks.**

- When working with these substances observe appropriate safety procedures (for example by wearing goggles, safety gloves and protective clothing) as described in the material handling and safety data sheet supplied by the vendor, and follow good laboratory practice.
  - Do not use solvents with an auto-ignition temperature below 200 °C (392 °F). Do not use solvents with a boiling point below 56 °C (133 °F).
  - Avoid high vapor concentrations. Always keep the temperature in the sample compartment at least 25 K below the boiling point of the solvent used.
  - Do not operate the instrument in an explosive atmosphere.
  - Reduce the volume of substances to the minimum required for the analysis.
  - Never exceed the maximum permissible volume of solvents (8 L) in the solvent cabinet. Do not use bottles that exceed the maximum permissible volume as specified in the usage guideline for solvent cabinet.
  - Ground the waste container.
  - Regularly check the filling level of the waste container. The residual free volume in the waste container must be large enough to collect the waste liquid.
  - To achieve maximal safety, regularly check the tubing for correct installation.
-

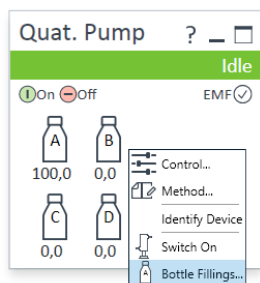
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#### Prepare a Run

- 1 Switch on the detector.



- 2 Fill the solvent bottles with adequate solvents for your application.
- 3 Place solvent tubings with bottle head assemblies into the solvent bottles.
- 4 Place solvent bottles into the solvent cabinet.
- 5 Solvent bottle filling dialog (in the software).



Solvent Bottle

Fillings

	Actual Volume		Total Volume	
A:	<input type="text" value="0.80"/>	liter	<input type="text" value="1.00"/>	liter
B:	<input type="text" value="0.92"/>	liter	<input type="text" value="1.00"/>	liter
C:	<input type="text" value="0.78"/>	liter	<input type="text" value="1.00"/>	liter
D:	<input type="text" value="0.81"/>	liter	<input type="text" value="1.00"/>	liter

Actions

Prevent analysis if level falls below  liter

Turn pump off if running out of solvent

Ok Cancel Help

**6** Purge the pump (in normal usage scenario).

OR

Prime the pump (after installation of the system).

**NOTE**

For details on priming and purging, refer to the technical note *Best Practices for Using an Agilent LC System*.

**7** Change solvent (if necessary).

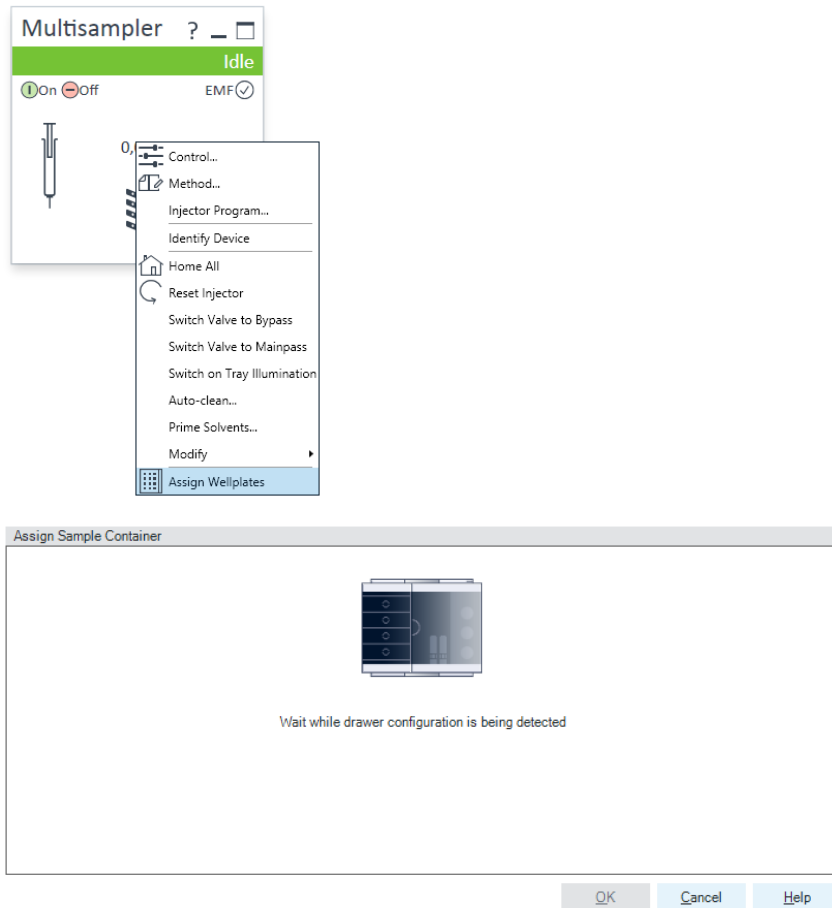
The screenshot displays the software interface for the Quat. Pump (G7111B). The interface is divided into several sections:

- Flow:** A text box shows a flow rate of 0.800 mL/min.
- Solvents:** A list of four solvent channels (A, B, C, D) with checkboxes and percentage/amount input fields. Channel A is set to 60.0% Water, and Channel B is checked and set to 40.0% Acetonitrile.
- Pressure Limits:** Min: 0.00 bar, Max: 600.00 bar.
- Stoptime/Posttime:** Radio buttons for 'As Injector/No Limit' and 'Off'. 'As Injector/No Limit' is selected with a value of 10.00 min. 'Off' is also selected with a value of 1.00 min.
- Advanced Settings:**
  - Minimum Stroke:** Radio buttons for 'Automatic' (selected) and a value of 20 µL.
  - Compressibility:** Radio buttons for '95 \*10<sup>-6</sup>/bar' (selected) and 'No compensation'.
  - Maximum Flow Gradient:** A text box showing 100.000 mL/min<sup>2</sup>.
  - Primary Channel:** A dropdown menu set to 'Automatic'.
- Bottom:** A blue button labeled 'Import Timetable...' and a status bar showing 'Timetable (1/100 events)'.

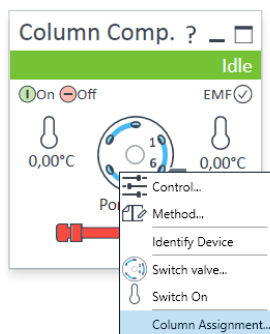
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8 Choose the tray format of the sampler.



**9** Add a new column.



**10** Enter the column information.

**Plumbing**

Valve Position	Location
1	Left 1
2	Left 2

**Visualization**

Valve Type: 2-pos/6-port valve 600 bar (5067-4137)

---

**Column Tag Information** >>

Location	Color Code	Description	Length [mm]	Diameter [mm]	Particle Size [µm]	Max. Pressure [bar]	Injections
Left 1	Red		0	0,0	0,0	0	0
▶ Left 2	Blue		0	0,0	0,0	0	0
Left 3	None		0	0,0	0,0	0	0
Left 4	None		0	0,0	0,0	0	0
Right 1	None		0	0,0	0,0	0	0
Right 2	None		0	0,0	0,0	0	0
Right 3	None		0	0,0	0,0	0	0
Right 4	None		0	0,0	0,0	0	0

Ok/Write Tag    Cancel    Help

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## 11 Select the column position.

**Temperature**


Left:  Not Controlled  40.0 °C  As Detector Cell  Unchanged

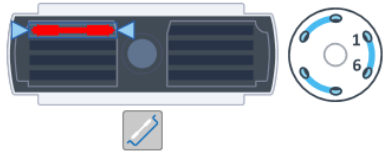
Right:  Not Controlled  25.0 °C  As Detector Cell  Unchanged  Combined

**Valve Position/Column**

Use Current Column / Position

Use Selected Column / Position

Position 1 



Enforce column for run

**Stoptime** **Posttime**

As Pump/Injector  1.00 min

Off  1.00 min

**Advanced**

**Enable Analysis**

when front door open

Left:  With any temperature  When temperature is within ± 0.8 °C for 0.0 min

Right:  With any temperature  When temperature is within ± 0.8 °C for 0.0 min

**Valve Position/Column After Run**

Do not switch

Switch to position / column at beginning of run

Increase valve position / column

Use valve position / column

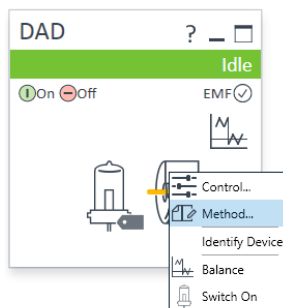
Position 1

Timetable (empty)

Ok Apply Cancel



**12** Set the detector according to the needs of your method.



Quat. Pump | VWD | Sampler | **DAD** | Column Comp.

**DAD (G7115A)**

Signals	Acquire	Wave length	Band width	Reference Wavelength	Reference Bandwidth	
Signal A	<input checked="" type="checkbox"/>	254	4	<input checked="" type="checkbox"/> 360	100	nm
Signal B	<input type="checkbox"/>	254	4	<input type="checkbox"/> 360	100	nm
Signal C	<input type="checkbox"/>	210	4	<input type="checkbox"/> 360	100	nm
Signal D	<input type="checkbox"/>	230	4	<input type="checkbox"/> 360	100	nm
Signal E	<input type="checkbox"/>	280	4	<input type="checkbox"/> 360	100	nm
Signal F	<input type="checkbox"/>	260	4	<input type="checkbox"/> 360	100	nm
Signal G	<input type="checkbox"/>	270	4	<input type="checkbox"/> 360	100	nm
Signal H	<input type="checkbox"/>	290	4	<input type="checkbox"/> 360	100	nm

Peakwidth  
> 0.0063 min (0.13 s response time) (40 Hz)

Stoptime | Posttime

As Pump/Injector       Off  
 1.00 min       1.00 min

**Advanced**

Spectrum  
Store: None  
Range from: 190 to 400 nm  
Step: 2.0 nm

Analog Output  
Zero Offset: 5 %  
Attenuation: 1000 mAU

Margin for negative Absorbance      Slit  
100 mAU      4 nm

Autobalance      Lamps on required for acquisition  
 Prerun       UV Lamp  
 Postrun       Vis Lamp

▶ Timetable (empty)

**NOTE**

For details on running a method, see “Setup the Checkout Method” on page 61 as an example.

## Check Out the System

### Checkout Method

This is an exemplary method for the Agilent InfinityLab LC Series.

Exemplary configuration:

- Quaternary Pump G7111B
- Multisampler G7167A
- Multicolumn Thermostat G7116A
- Diode Array Detector WR G7115A

The RRLC checkout sample (5188-6529) serves as standard for systems with 600 bar and contains 100 ng/ $\mu$ L each of nine components dissolved in water / acetonitrile (65/35). The nine components are:

- Acetanilide
- Acetophenone
- Propiophenone
- Butyrophenone
- Benzophenone
- Valerophenone
- Hexanophenone
- Heptanophenone
- Octanophenone

The Agilent isocratic checkout sample (01080-68704) serves as standard for systems with 400 bar and contains each of four components dissolved in methanol.

- Dimethylphthalate
- Diethylphthalate
- Biphenyl
- o-Terphenyl

**NOTE**

Find the correct settings for the individual modules here:

- Checkout method parameter settings Isocratic Pump (G7110B) ([Table 3](#) on page 58)
- Checkout method parameter settings Quaternary Pump VL (G7111A) ([Table 4](#) on page 58)
- Checkout method parameter settings Quaternary Pump (G7111B) ([Table 5](#) on page 59)
- Checkout method parameter settings Multisampler (G7167A) or Vialsampler (G7129A) ([Table 6](#) on page 59)
- Checkout method parameter settings Multicolumn Thermostat (G7116A) ([Table 7](#) on page 59)
- Checkout method parameter settings Diode Array Detector HS (G7117C) or Diode Array Detector WR (G7115A) ([Table 8](#) on page 60)
- Checkout method parameter settings Variable Wavelength Detector (G7114A) ([Table 9](#) on page 60)

**Table 2** Overview of column options for different pumps

Pump	Option 1	Option 2	Option 3
1260 Infinity II Quaternary and Binary Pump (G7111B/G7112B)	Poroshell 120 EC-C18, 4.6 x 100 mm, 2.7 µm	Poroshell 120 EC-C18, 3.0 x 150, 2.7 µm	Poroshell 120 EC-C18, 3.0 x 50 mm, 2.7 µm
	695975-902T	693975-302T	699975-302T
1260 Infinity II Quaternary Pump VL (G7111A)	Poroshell 120 EC-C18, 4.6 x 100 mm, 4 µm	Poroshell 120 EC-C18, 4.6 x 50 mm, 2.7 µm	Poroshell 120 EC-C18, 4.6 x 150 mm, 4 µm
	695970-902T	699975-902T	693970-902T

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**Table 3** Checkout method parameter settings Isocratic Pump (G7110B)

Parameter	Value
Flow	1.5 mL/min
Solvents	65 % ACN in water
Compressibility	75
Stoptime	10 min
Pressure Limit	600 bar
Minimum Stroke	Automatic

**Table 4** Checkout method parameter settings Quaternary Pump VL (G7111A)

Parameter	Value
Flow	1 mL/min
Solvent A	Water
Solvent B	ACN
Compressibility	75
Composition	65 % B (ACN)
Composition	35 % A (Water)
Stoptime	10 min
Pressure Limit	400 bar
Minimum Stroke	Automatic

**Table 5** Checkout method parameter settings Quaternary Pump (G7111B)

Parameter	Value
Flow	0.8 mL/min
Solvent A	Water
Solvent B	ACN
Compressibility	95
Composition	40 %B (ACN)
Composition	60 %A (Water)
Stoptime	10 min
Pressure Limit	600 bar
Minimum Stroke	Automatic
Timetable	2.5 min 80 %B

**Table 6** Checkout method parameter settings Vialsampler/Multisampler (G7129A/G7167A)

Parameter	Value
Injection	1 µL
Stoptime	as pump
Draw speed	100 µL/min

**Table 7** Checkout method parameter settings Multicolumn Thermostat (G7116A)

Parameter	Value
Temperature (left)	40 °C
Temperature (right)	combined
Stoptime	as pump

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**Table 8** Checkout method parameter settings Diode Array Detectors (G7115A/G7117C)

Parameter	Value
Signal A	254/4 nm
Ref A	360/100 nm
Peakwidth	40 Hz
Stoptime	as pump
Spectrum	None
Autobalance	Prerun

**Table 9** Checkout method parameter settings Variable Wavelength Detector (G7114A)

Parameter	Value
Wavelength	254 nm
Peakwidth	40 Hz
Stoptime	as pump
Autobalance	Prerun

## Setup the Checkout Method

### NOTE

The setup of the checkout method in this procedure is an example. For the individual module parameters, see “[Checkout Method](#)” on page 56.

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- 1 Turn on the lamp.
- 2 Load the the default method DEF\_LC.M

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### 3 Change the method and timetable settings for the 1260 Infinity II Quaternary Pump (G7111B)


The screenshot displays the software interface for the Quat. Pump (G7111B). The top navigation bar includes tabs for 'Quat. Pump', 'VWD', 'Sampler', 'DAD', and 'Column Comp.'. The main window is titled 'Quat. Pump (G7111B)' and features a small pump icon. The interface is divided into several sections:

- Flow:** A text box shows a flow rate of 0.800 mL/min.
- Solvents:** A list of four solvent channels (A, B, C, D) with percentage and name input fields. Channel A is set to 60.0% Water, and Channel B is checked and set to 40.0% Acetonitrile. Channels C and D are set to 0.0%.
- Pressure Limits:** Minimum pressure is set to 0.00 bar and maximum pressure is set to 600.00 bar.
- Stoptime/Posttime:** Two radio button options: 'As Injector/No Limit' (selected) with a 10.00 min timer, and 'Off' with a 1.00 min timer.
- Advanced:** A sidebar containing:
  - Minimum Stroke:** Radio buttons for 'Automatic' (selected) and a 20 µL value.
  - Compressibility:** Radio buttons for '95 \*10<sup>-6</sup> / bar' (selected) and 'No compensation'.
  - Maximum Flow Gradient:** A text box set to 100.000 mL/min<sup>2</sup>.
  - Primary Channel:** A dropdown menu set to 'Automatic'.
- Import Timetable...:** A light blue button at the bottom left.
- Timetable:** A scrollable area at the bottom right showing 'Timetable (1/100 events)'.

**Figure 25** Method settings 1260 Infinity II Quaternary Pump (G7111B)



Quat. Pump VWD Sampler DAD Column Comp.

**Quat. Pump (G7111B)** 

**Flow**  
0.800 mL/min

**Solvents**

A: 60.0 % Water

B:  40.0 % Acetonitrile

C:  0.0 %

D:  0.0 %

**Pressure Limits**  
Min: 0.00 bar Max: 600.00 bar

**Stoptime** **Posttime**

As Injector/No Limit  Off

10.00 min  1.00 min

Import Timetable...

**Advanced**

Timetable (1/100 events)  function centric view

Time [min]	A [%]	B [%]	C [%]	D [%]	Flow [mL/min]	Max. Pressure Limit [bar]
0.00	60.0	40.0	0.0	0.0	0.800	600.00
2.50	20.0	80.0	0.0	0.0	---	---

Add Remove Clear All Clear Empty

Cut Copy Paste Shift Times 0.00

**Figure 26** Timetable settings 1260 Infinity II Quaternary Pump (G7111B)

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#### 4 Change the method settings for the 1260 Infinity II Multisampler (G7167A)

The screenshot displays the software interface for the Multisampler (G7167A). The top navigation bar includes tabs for Quat. Pump, VWD, Sampler, DAD, Column Comp., Multisampler, Iso. Pump, Quat. Pump, Binary Pump, and Multisampler. The main window is titled "Multisampler (G7167A)".

**Injection**  
Injection volume: 1.00  $\mu\text{L}$

**Needle Wash**  
Standard Wash

**Stoptime** | **Posttime**

As Pump/No Limit |  Off  
 1.00 min |  1.00 min

**Advanced**

**Sampling Speed**  
Draw Speed: 100.0  $\mu\text{L}/\text{min}$   
Eject Speed: 400.0  $\mu\text{L}/\text{min}$   
Wait Time After Draw: 1.2 s

**Needle Height Position**  
Offset: 0.0 mm  
 Use Vial/Well Bottom Sensing

**High Throughput**  
Sample Flush-Out Factor: 5.0  
 Injection Valve to Bypass for Delay Volume Reduction  
 Enable Overlapped Injection  
 When Sample is Flushed Out  
 After Period of Time  
0.00 minutes after injection

**Injection Path Cleaning**

**Figure 27** Method setting 1260 Infinity II Multisampler (G7167A)

Quat. Pump | VWD | Sampler | DAD | Column Comp. | Multisampler | Iso. Pump | Quat. Pump | Binary Pump | Multisampler

**Multisampler (G7167A)**

**Injection**

Injection volume:   $\mu\text{L}$

**Needle Wash**

**Stoptime** | **Posttime**

As Pump/No Limit       Off  
  min        min

**Advanced**

**Injection Path Cleaning**

**Standard Wash**

Mode:

Time:  s

Location:

Repeat:

**Multi-wash**

Step	Solvent	Time [s]	Seat Back Flush	Needle Wash	Comment
1	Off		<input type="checkbox"/>	<input type="checkbox"/>	
2	Off		<input type="checkbox"/>	<input type="checkbox"/>	
3	Off		<input type="checkbox"/>	<input type="checkbox"/>	
Start Cond.	S1		<input checked="" type="checkbox"/>	<input type="checkbox"/>	

**Figure 28** Method setting 1260 Infinity II Multisampler (G7167A) - Injection Path Cleaning

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## 5 Change the method settings for the 1260 Infinity II Multicolumn Thermostat (G7116A)

**Temperature**

Left:  Not Controlled  40.0 °C  As Detector Cell  Unchanged

Right:  Not Controlled  25.0 °C  As Detector Cell  Unchanged  Combined

**Valve Position/Column**

Use Current Column / Position

Use Selected Column / Position

Position 1

Enforce column for run

**Stoptime** **Posttime**

As Pump/Injector  1.00 min

Off  1.00 min

**Advanced**

**Enable Analysis**

when front door open

Left:  With any temperature  When temperature is within ± 0.8 °C for 0.0 min

Right:  With any temperature  When temperature is within ± 0.8 °C for 0.0 min

**Valve Position/Column After Run**

Do not switch

Switch to position / column at beginning of run

Increase valve position / column

Use valve position / column

Position 1

Timetable (empty)

Ok Apply Cancel

**Figure 29** Method setting 1260 Infinity II Multicolumn Thermostat (G7116A)

## 6 Change the method settings for the 1260 Infinity II Diode Array Detector WR (G7115A)

Quat. Pump VWD Sampler DAD Column Comp.

**DAD (G7115A)**

**Signals**

	Acquire	Wave length	Band width	Reference Wavelength	Reference Bandwidth	
Signal A	<input checked="" type="checkbox"/>	254	4	<input checked="" type="checkbox"/> 360	100	nm
Signal B	<input type="checkbox"/>	254	4	<input type="checkbox"/> 360	100	nm
Signal C	<input type="checkbox"/>	210	4	<input type="checkbox"/> 360	100	nm
Signal D	<input type="checkbox"/>	230	4	<input type="checkbox"/> 360	100	nm
Signal E	<input type="checkbox"/>	280	4	<input type="checkbox"/> 360	100	nm
Signal F	<input type="checkbox"/>	260	4	<input type="checkbox"/> 360	100	nm
Signal G	<input type="checkbox"/>	270	4	<input type="checkbox"/> 360	100	nm
Signal H	<input type="checkbox"/>	290	4	<input type="checkbox"/> 360	100	nm

**Peakwidth**

> 0.0063 min (0.13 s response time) (40 Hz)

**Stoptime**      **Posttime**

As Pump/Injector       Off

1.00 min       1.00 min

**Advanced**

**Spectrum**

Store: None

Range from: 190 to 400 nm

Step: 2.0 nm

**Analog Output**

Zero Offset: 5 %

Attenuation: 1000 mAU

**Margin for negative Absorbance**      **Slit**

100 mAU      4 nm

**Autobalance**      **Lamps on required for acquisition**

Prerun       UV Lamp

Postrun       Vis Lamp

▶ Timetable (empty)

**Figure 30** Method setting 1260 Infinity II Diode Array Detector WR (G7115A)

- 7 Save the method as GRAD-1.M
- 8 Equilibrate the system for 10 min under checkout conditions
- 9 Run and evaluate the checkout method

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**Check Out the System**