228-90775C
Jun. 2013

AUTOSAMPLER FOR SHIMADZU ULTRA HIGH PERFORMANCE LIQUID CHROMATOGRAGH

Nexera X2 SIL-30AC

Instruction Manual

Read the instruction manual thoroughly before you use the product.
Keep this instruction manual for future reference.

Introduction

Read this manual thoroughly before using the instrument.

Thank you for purchasing this product.
This manual describes the installation, operation, usage cautions, and accessories and options for this product. Read this manual thoroughly before using the product and operate the product in accordance with the instructions in this manual.
The following instruction manuals are included with the product.

1. Nexera X2 SIL-30AC Instruction Manual (S228-90775)
2. Nexera X2 Safety Guideline (S228-91060)
3. Nexera X2 System Guide (S228-90870)

Before using the product, be sure to read the separate manual "Nexera X2 Safety Guideline", which describes product warranty, after-sales service, and precaution items for safety use. For installation procedure, hardware validation, etc. for the Nexera X2 series involving this product, refer to the "Nexera X2 System Guide".
Keep this manual for future reference.

IMPORTANT	• If the user or usage location changes, ensure that this Instruction Manual is always kept together with the product. • If this documentation or the warning labels on the instrument become lost or damaged, promptly obtain replacements from your Shimadzu representative. • To ensure safe operation, contact your Shimadzu representative if product installation, adjustment, or re-installation (after the product is moved) is required.

Notice

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- Nexera is a trademark of Shimadzu Corporation.
- Other company names and product names mentioned in this manual are trademarks or registered trademarks of their respective companies. The TM and ® symbols are omitted in this manual.

Original version is approved in English.

■ Indications used in the manual

In this manual, the following symbols are used.

⚠ **WARNING**	**Indicates a potentially hazardous situation which, if not avoided, could result in serious injury or possibly death.**
⚠ **CAUTION**	**Indicates a potentially hazardous situation which, if not avoided, may result in minor to moderate injury or equipment damage.**
NOTE	**Emphasizes additional information that is provided to ensure the proper use of this product.**

Contents

Chapter 1 Configuration

Chapter 2 Parts Identification and Function

Chapter 3 Preparation

Chapter 4 Basic Operation

Chapter 5 Application Operation

Chapter 6 Troubleshooting

Chapter 7 Maintenance

Chapter 8 Technical Information

Index

1 Configuration

1.1 Overview

The Shimadzu SIL-30AC autosampler is designed for use with the Shimadzu Ultra High Performance Liquid Chromatograph Nexera X2 series.
While ensuring the superior durability and repeatability of the SIL-20A/20AC series, the maximum allowable pressure is increased to 130 MPa and the internal capacity of the high-pressure valve is reduced to dampen peak diffusion. The injection capacity ranges from a volume of 0.1 μL up to a maximum of 50 μL (or 20 μL with the loop injection method).
There are two modes for sample injection; standard injection mode that facilitates condition setting, and repeat injection mode that the bracket sequence analysis can be performed in a certain cycle.
The instrument is equipped with a sample cooler that is able to control the sample temperature in the range between 4 and 40 °C. This feature helps users to perform serial analysis while cooling the samples that are apt to be decomposed in room temperature.

1.2 Sample Injection Modes

This autosampler has two sample injection modes, standard injection mode and repeat injection mode.

1.2.1 Standard Injection Mode

This is the simplest mode for making a standard injection. In this mode, specify the sample vial number, number of injections, injection volume and analysis time in the sample table setting screen (details are in Section "4.2 Creating an Analysis Sequence Table" P. 52). Also, set the needle stroke, the rinse volume, the rinsing speed, sampling speed, etc. in the Parameter setting screen.

1.2.2 Repeat Injection Mode

This mode can be used to inject a predetermined number of samples repeatedly, at periodic intervals. In this mode, a special repeat injection table is prepared (in addition to the default analysis sequence table). Preparing this repeat injection table allows bracket sequence analysis to be performed at a certain cycle while sampling table for standard injection is being executed.

1.3 Sample Injection Methods

The instrument can use two kinds of sample injection methods: total injection method and loop injection method, whichever best suits the desired application to be selected.

1.3.1 Total Injection Method (Standard)

Based on the set volume of injection, the sample is aspirated from the sample vial and the total volume is injected to the HPLC column.
Since the needle and injection port are incorporated in the flow line, high-pressure pumping of mobile phase during analysis has an effect of cleansing the flow line; thus carryover can be reduced to an absolute minimum even if the inside of the needle and the injection port are not rinsed.
A sample injection will be made according to the flow as shown in the next section.

1.3.2 Loop Injection Method (Option)

The loop injection method is effective for high-speed high-separation analysis because the delay volume can be reduced compared with the total injection method.
In addition, the loop injection method has two kinds of methods: partial loop method of injecting a set volume to the HPLC column, and full loop method of filling the entire loop with sample and injecting the sample to the HPLC column. The partial loop method is recommended if it is necessary to set the volume of injection. If precise analysis is required repeatedly, the full loop method is recommended.

1.4 Sample Injection

Sample injection operations involved with total injection method, that is the standard injection method of the instrument, are explained.
Sample injection will be made according to the flow as shown below.

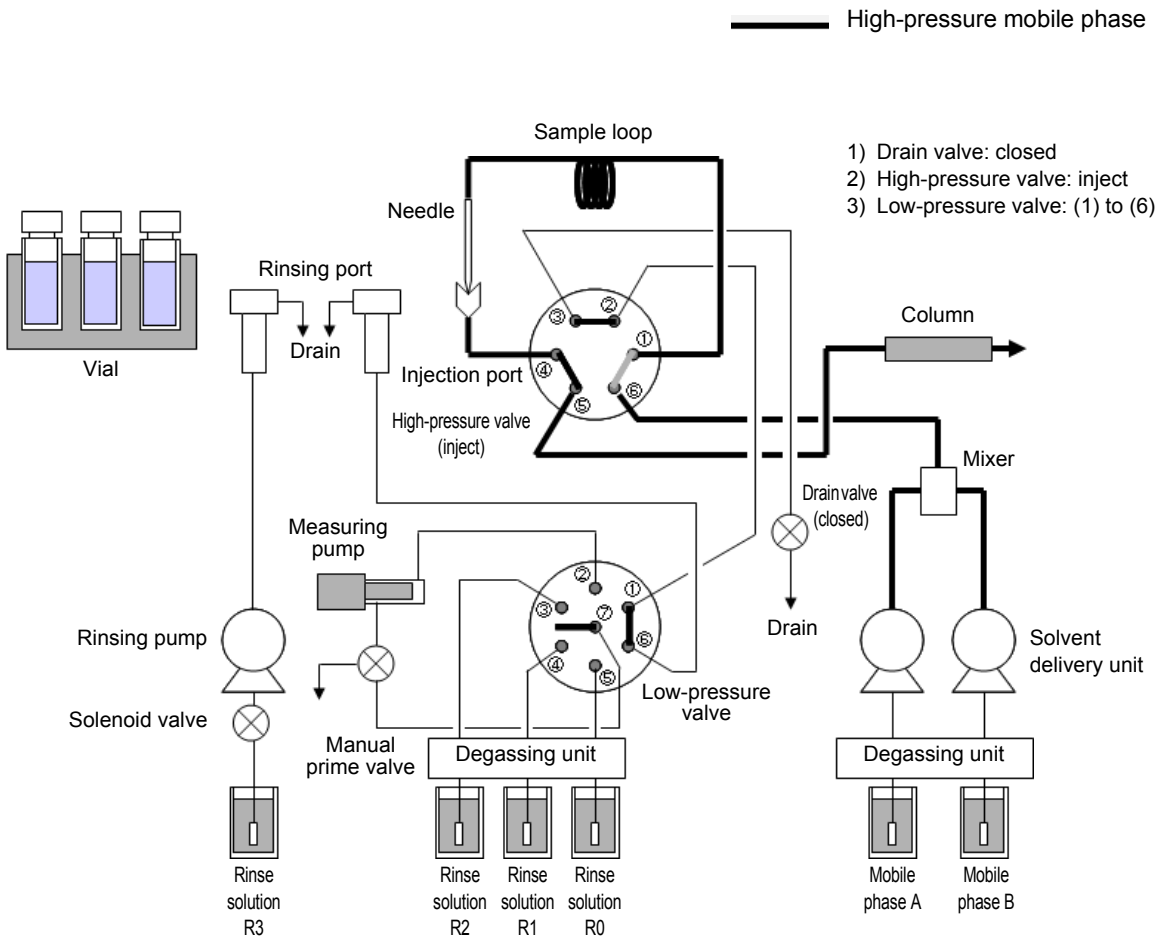


Fig. 1.1

1. Standby (READY)

Mobile phase from the reservoir is pumped, in order, through the high-pressure valve, sample loop, needle, injection port and back through high-pressure valve, before reaching the analysis column.

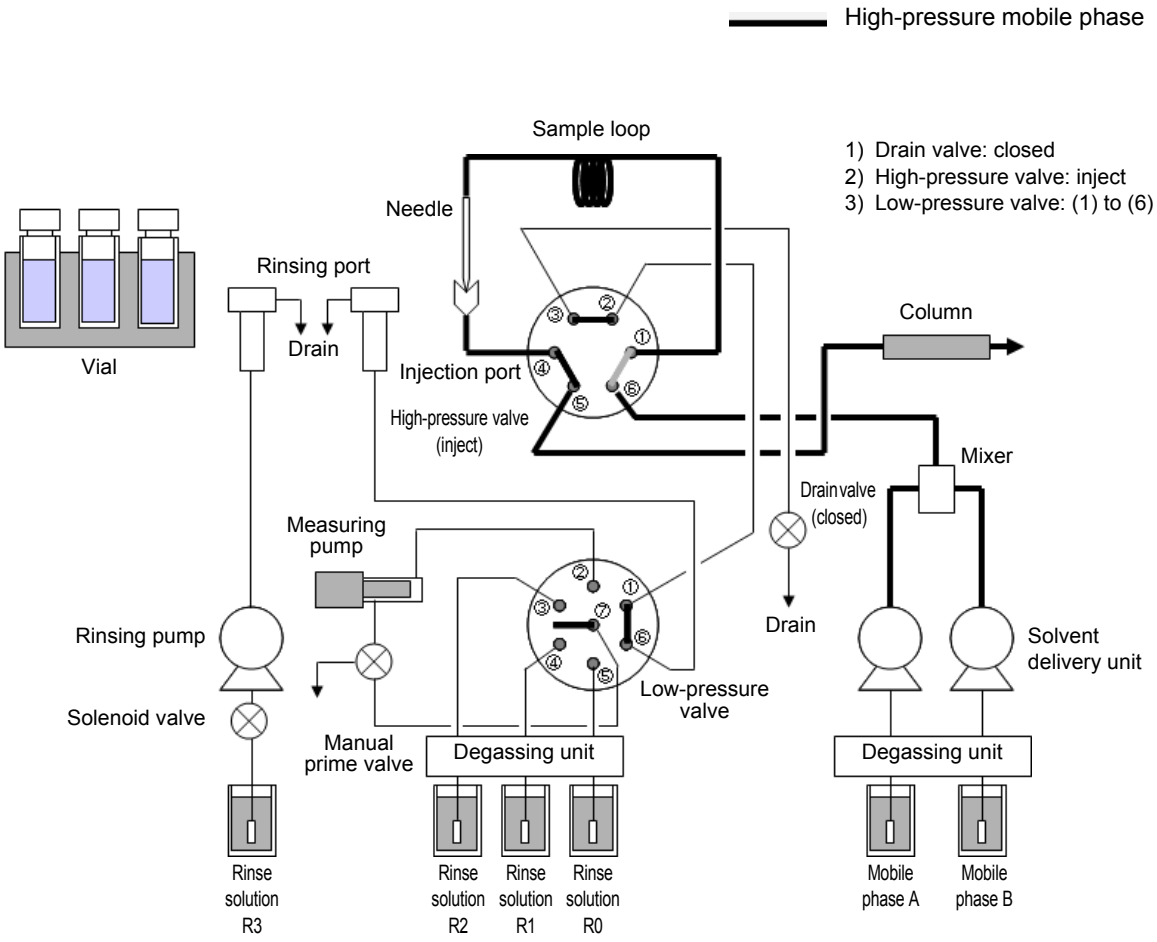


Fig. 1.2

2. Release of pressure in flow line

The high-pressure valve rotates to the load position (60° in the clockwise direction), and the high-pressure sample-loop mobile phase remaining in the sample loop flows through the needle → sample loop → high-pressure valve → low-pressure valve → rinsing port, and needle → injection port → high-pressure valve → drain valve, relieving the pressure in the sample loop.

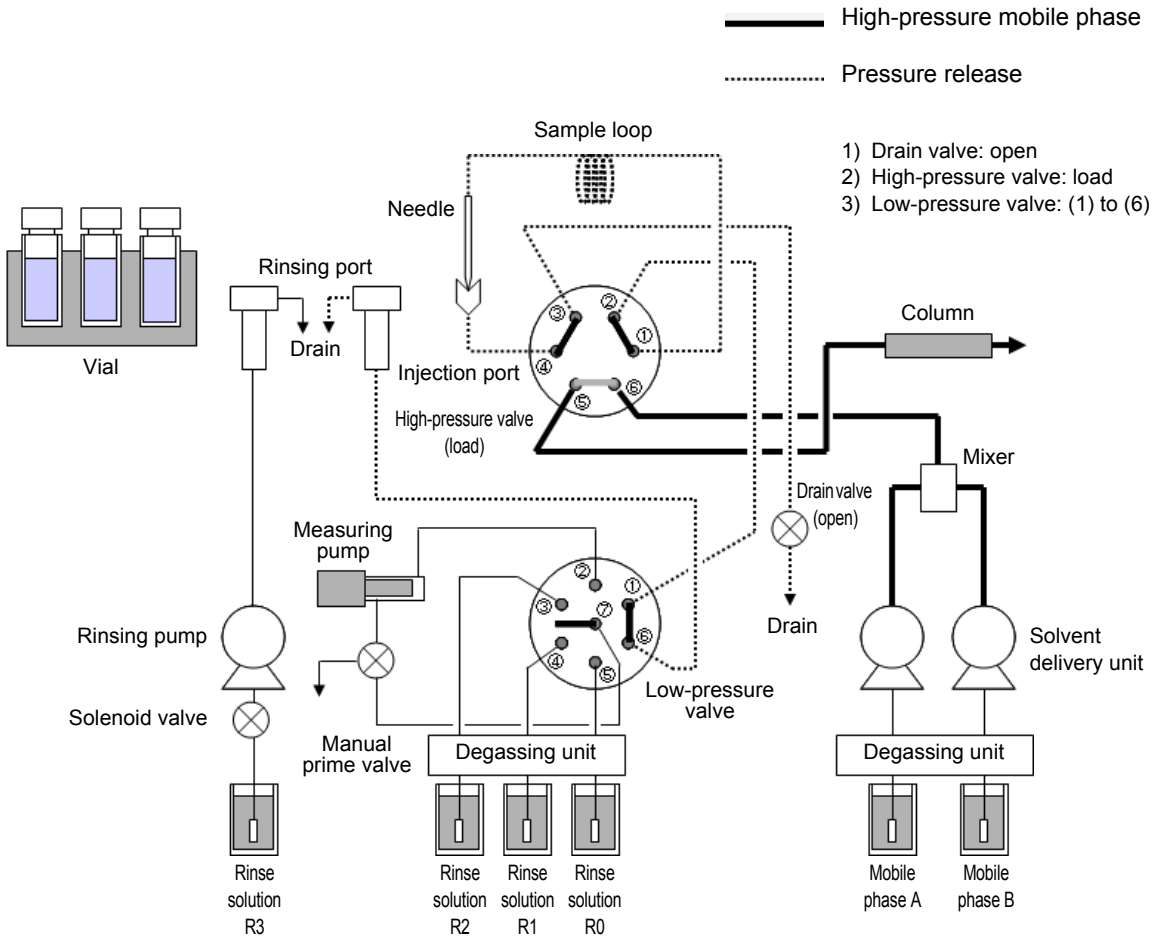


Fig. 1.3

3. Needle movement

The low-pressure valve rotates to the measuring position (210° in the counter-clockwise direction) and the needle moves up.

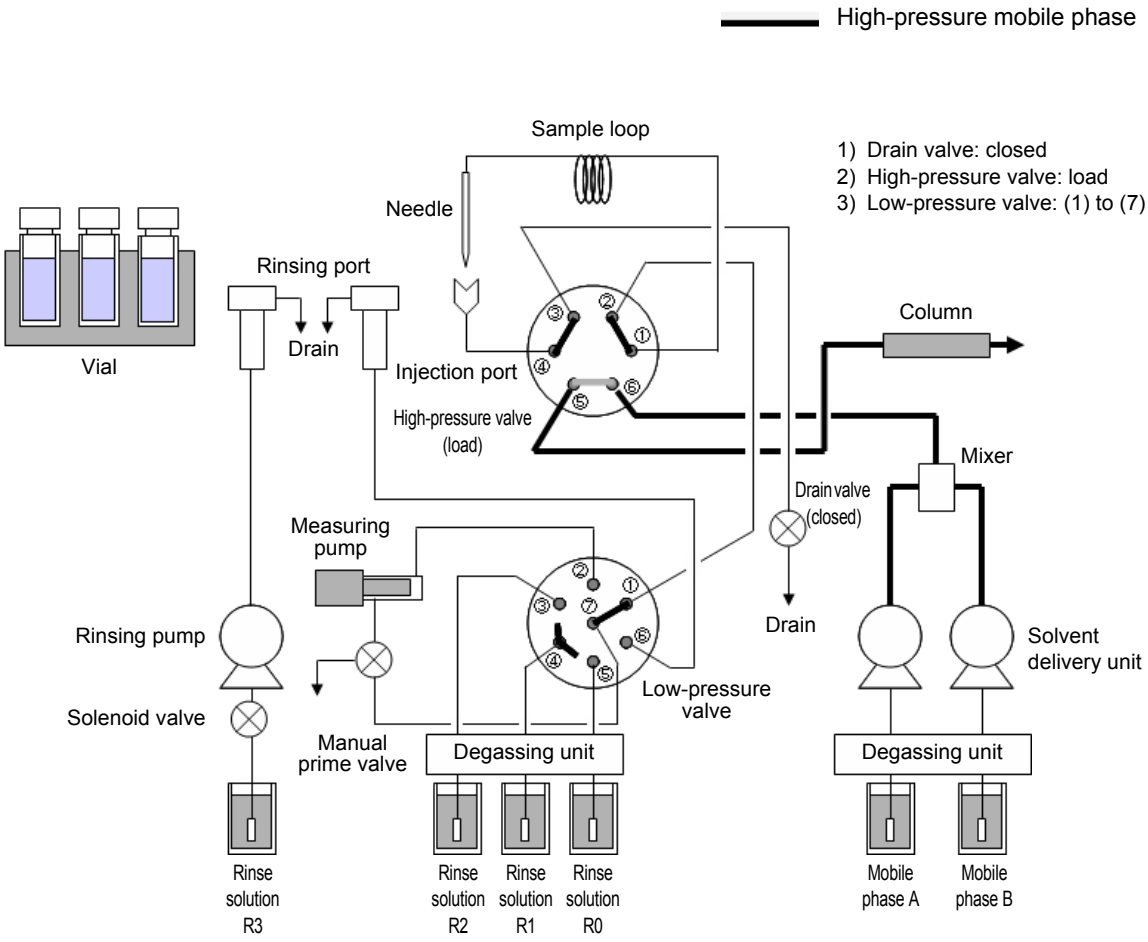


Fig. 1.4

4. External rinsing of needle before sample aspiration

The needle is inserted into the rinsing port and the external surface of the needle is rinsed with the rinse solution in the rinsing port.
It is possible to set to skip external rinsing of the needle before sample aspiration.
Rinsing can be performed with two kinds of rinse solution when a rinsing pump is used.

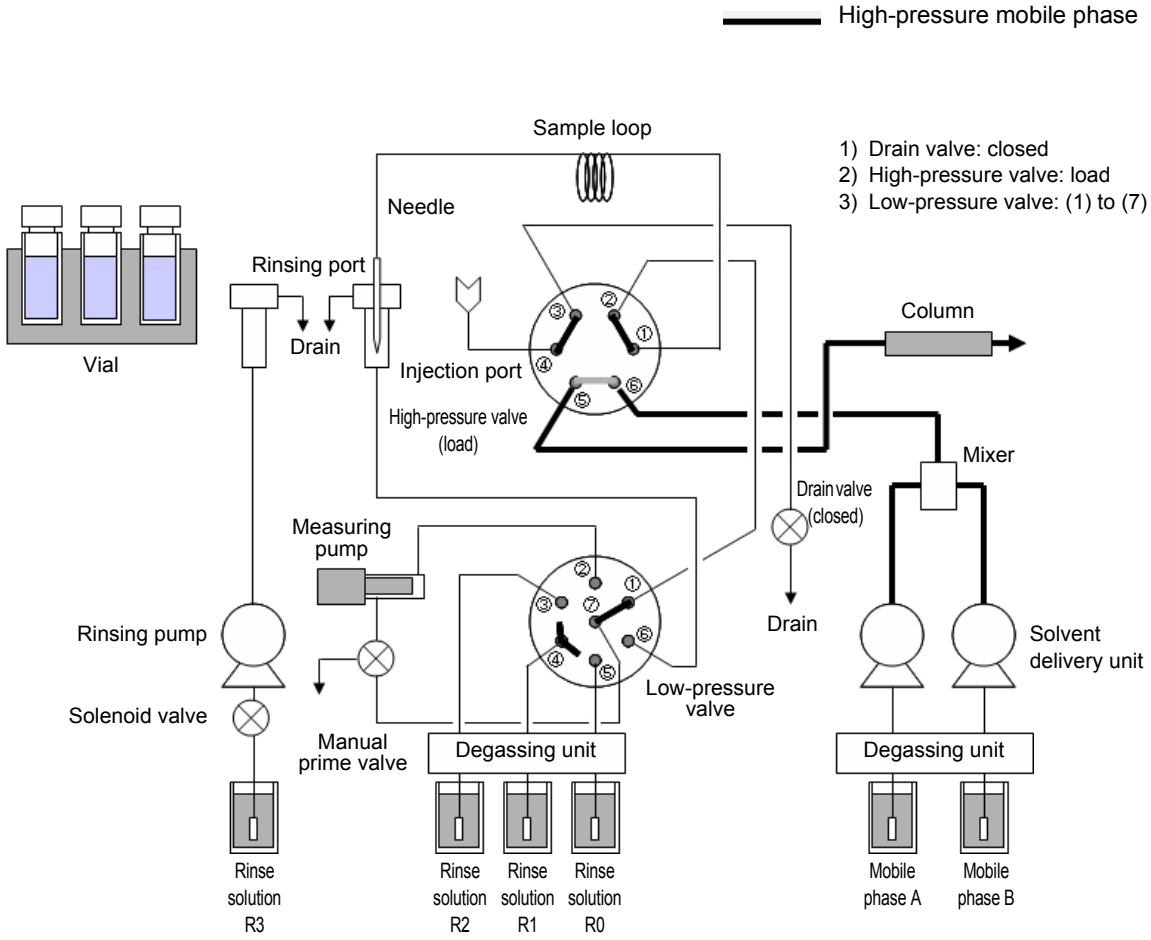


Fig. 1.5

5. Sample aspiration

The needle is inserted into the sample vial. Then the measuring pump draws the sample into the needle and sample loop.

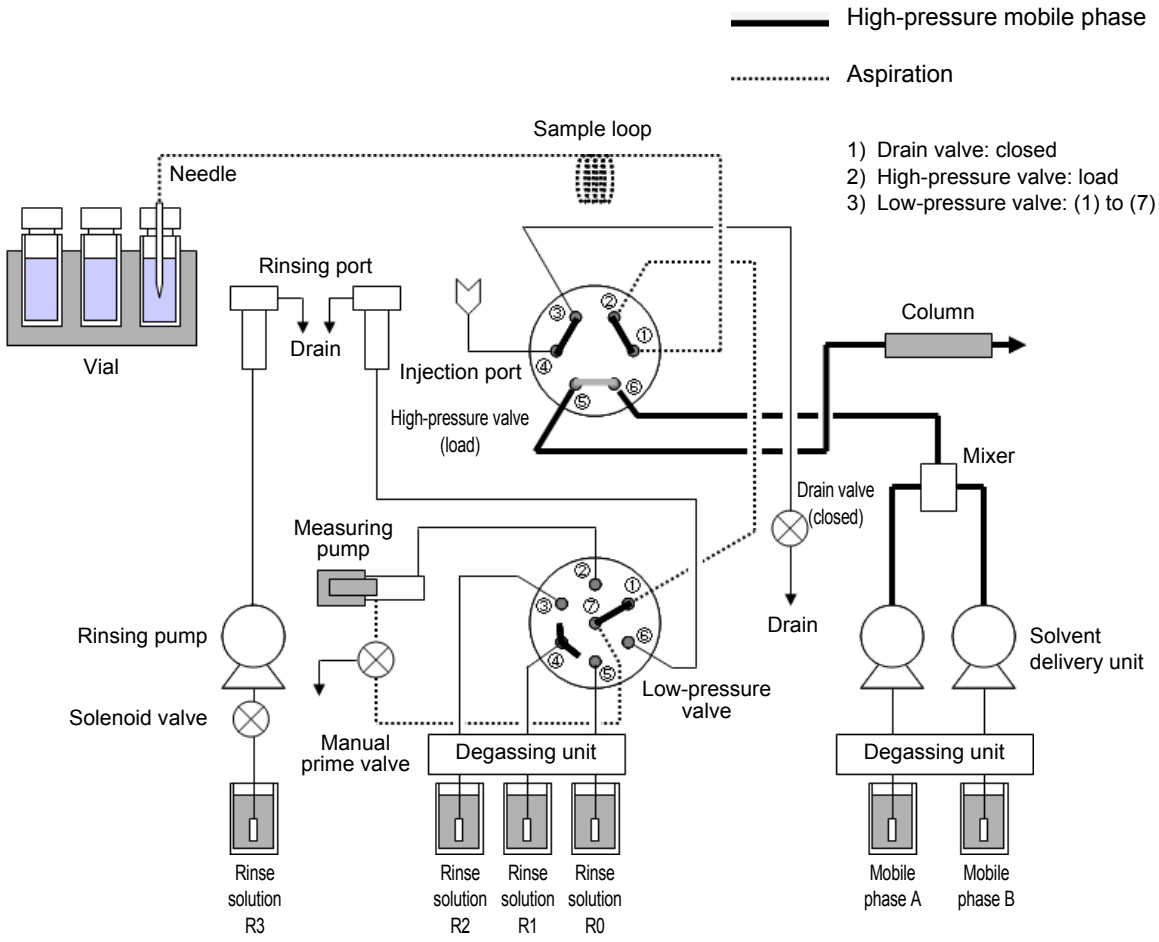


Fig. 1.6

6. External rinsing of needle after sample aspiration

The needle is inserted into the rinsing port, where its outer surfaces are rinsed with the rinse solution inside the port. It is also possible to set the autosampler not to perform rinsing. In addition, using a needle-rinsing pump allows rinsing to be performed with two types of rinse solutions.

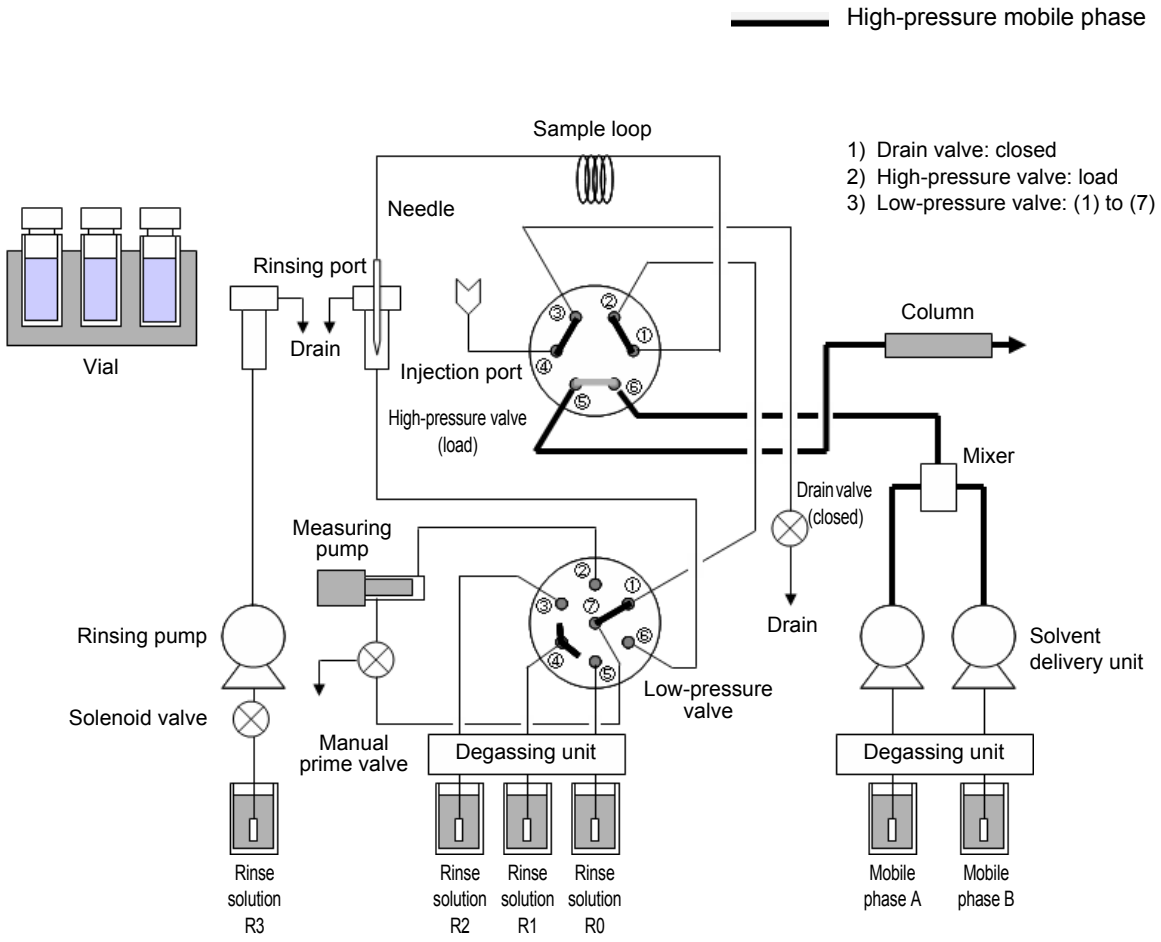


Fig. 1.7

7. Start of analysis (sample injection)

The needle is inserted into the injection port, and the high-pressure valve rotates 60° counterclockwise to the injection position. The sample is injected into the flow lines and, along with the mobile phase, passes through the high-pressure valve and into the column, where analysis begins.

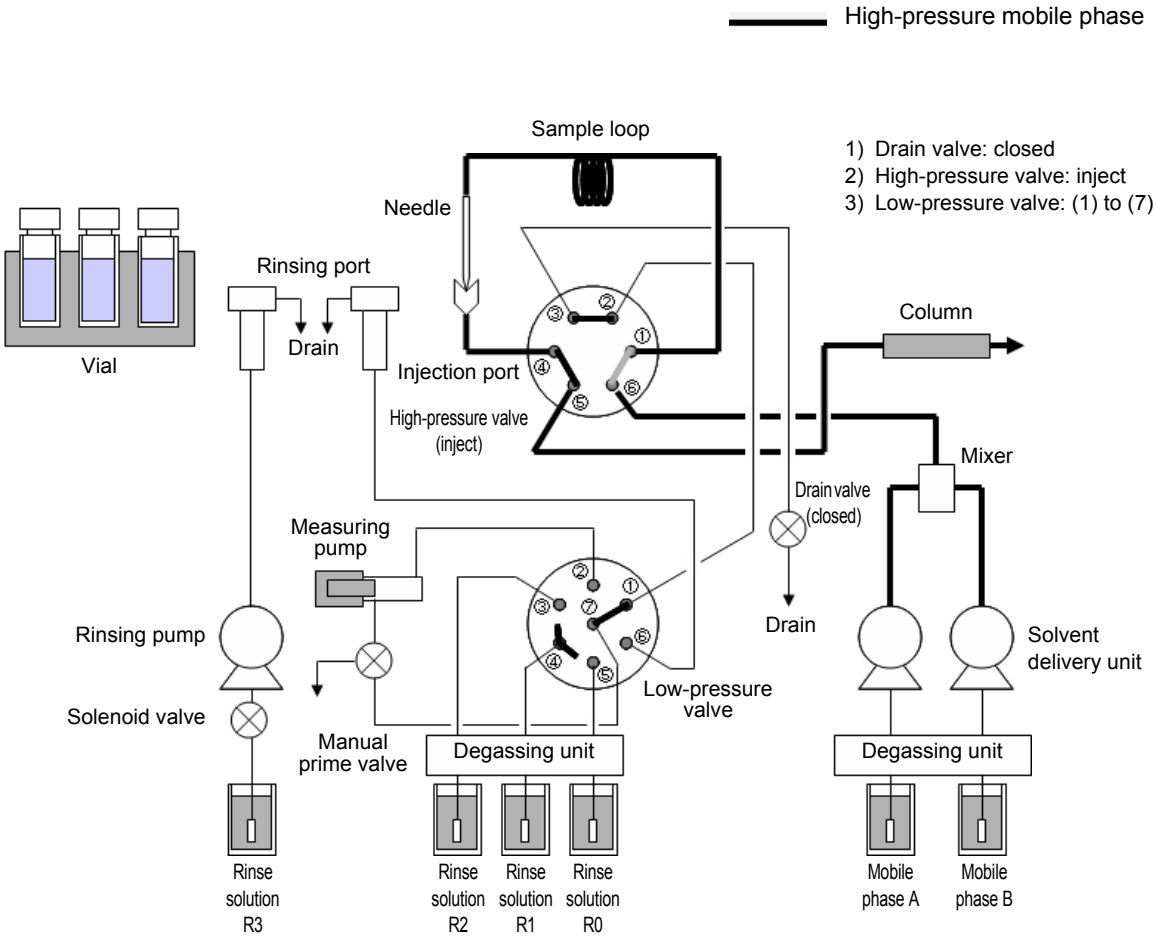


Fig. 1.8

8. Measuring pump home position setting

The measuring pump dispenses the sample and sets the home position.

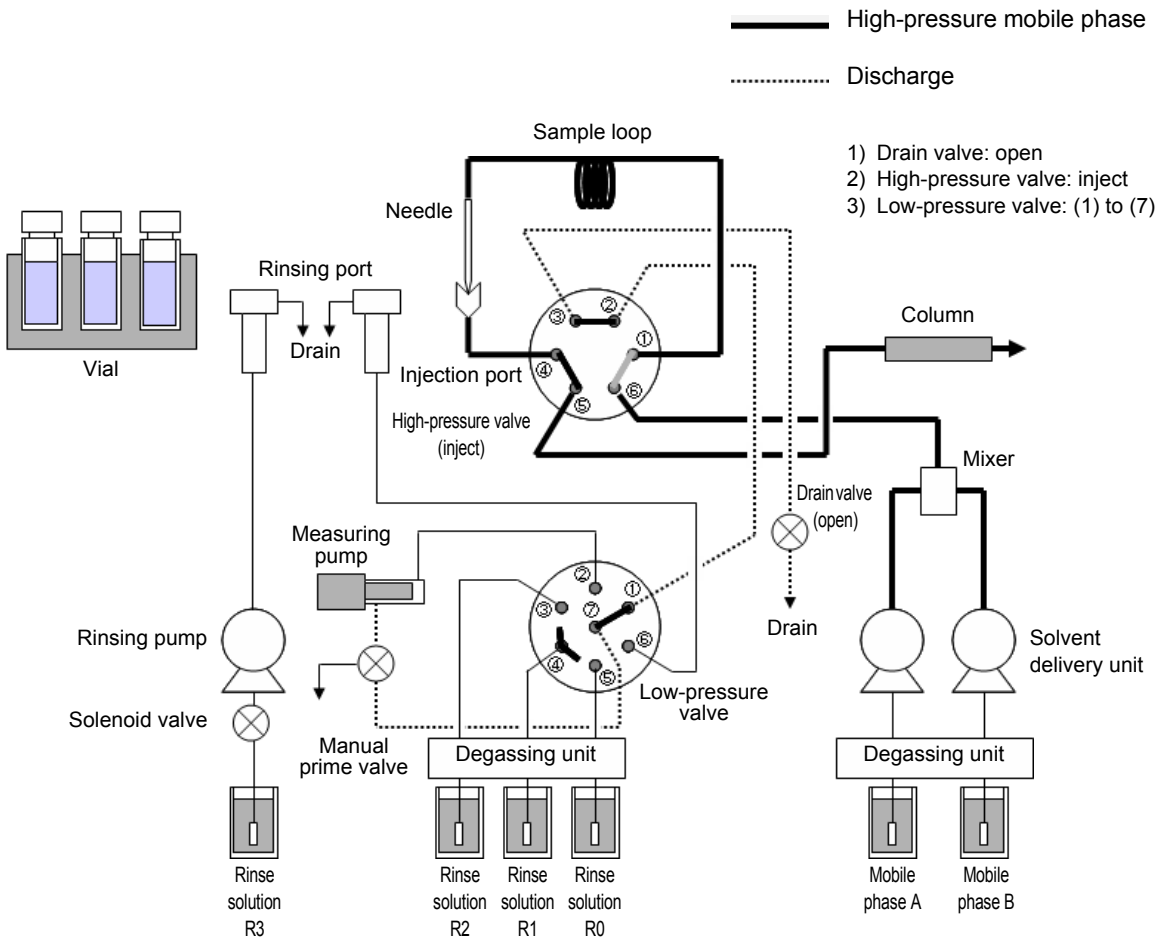


Fig. 1.9

9. Aspiration of rinse solution (R0)

The low-pressure valve rotates to the position (120° in the clockwise direction) where ports 5 and 7 are connected, and the plunger operates for aspiration of rinse solution (R0).

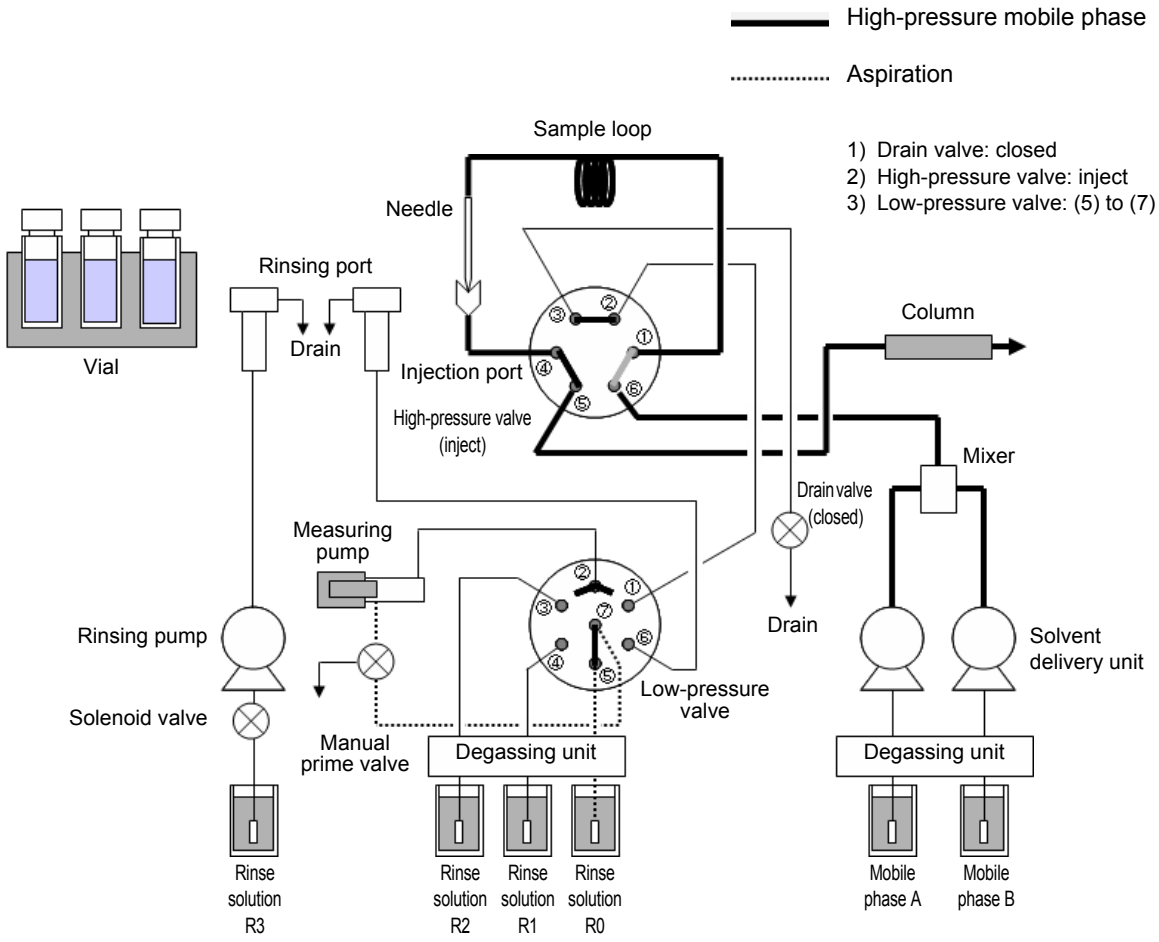


Fig. 1.10

10. Dispensing rinse solution (R0) to the measuring flow line

The low-pressure valve rotates 30° in the clockwise direction, and the plunger of the measuring pump operates for dispensing rinse solution (R0) to the drain valve to purge the measuring flow line.

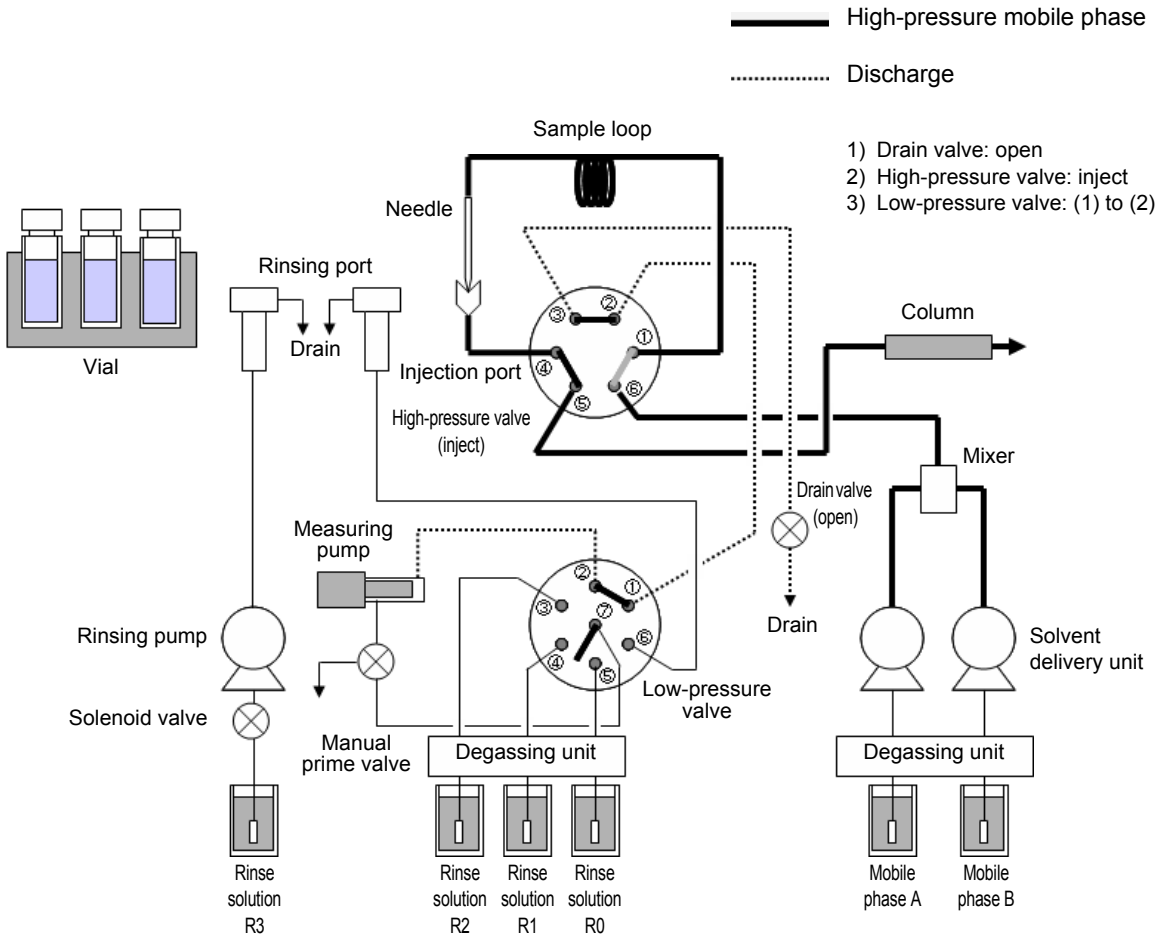


Fig. 1.11

11. Dispensing rinse solution (R0) to the rinsing port

The low-pressure valve rotates 30° in the counter-clockwise direction, and the plunger operates for aspiration of rinse solution (R0). Then the low-pressure valve rotates 60° in the counter-clockwise direction, and the plunger of the measuring pump operates for dispensing rinse solution (R0) to the rinsing port.

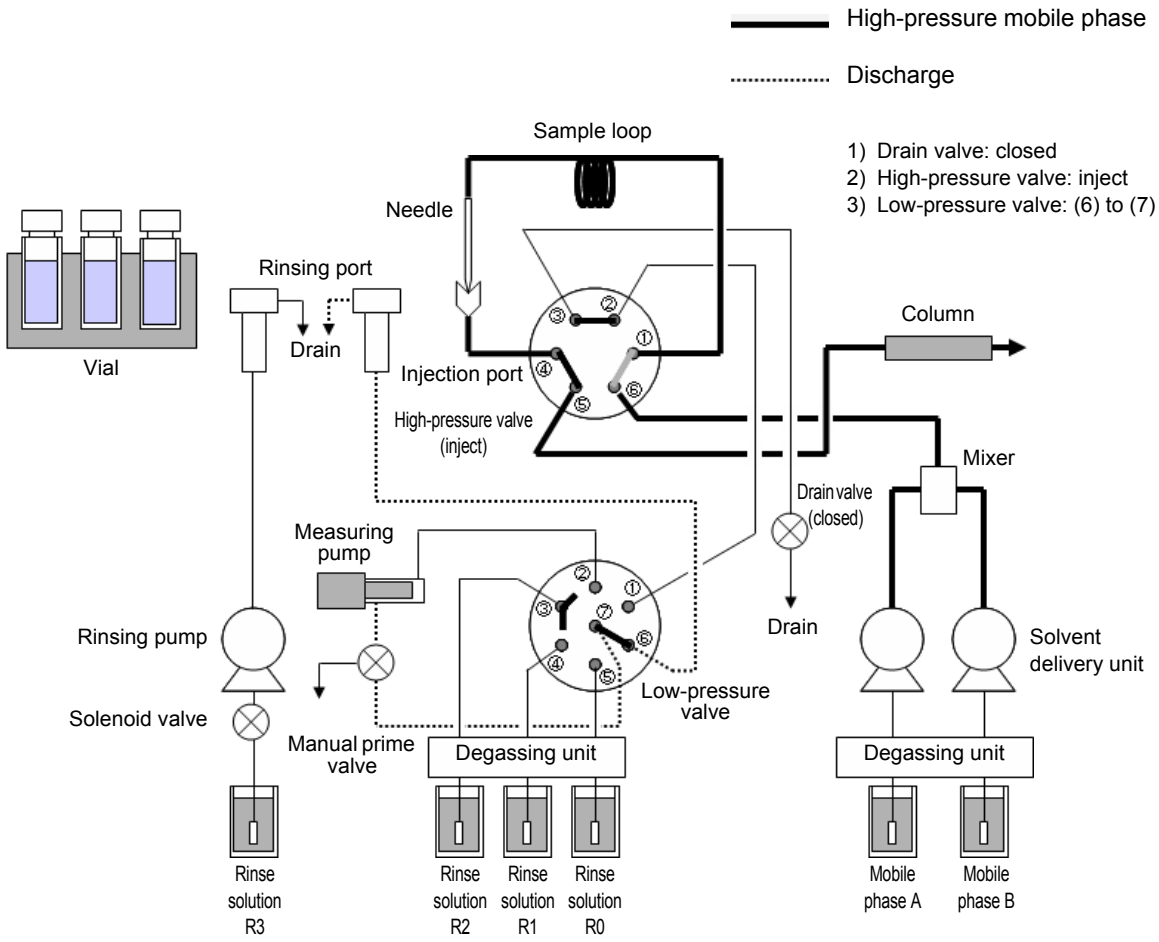


Fig. 1.12

12. (Reference) Internal rinsing of needle with rinse solution (R0, R1, R2)

When "2" is set for [RINSE TYPE] and internal rinsing of the needle is performed after sample injection, the specified rinse solution is aspirated by the measuring pump, and ports 1 and 2 of the low-pressure valve are connected, and rinse solution is dispensed to the high-pressure valve → sample loop → needle → injection port → high-pressure valve → drain valve.

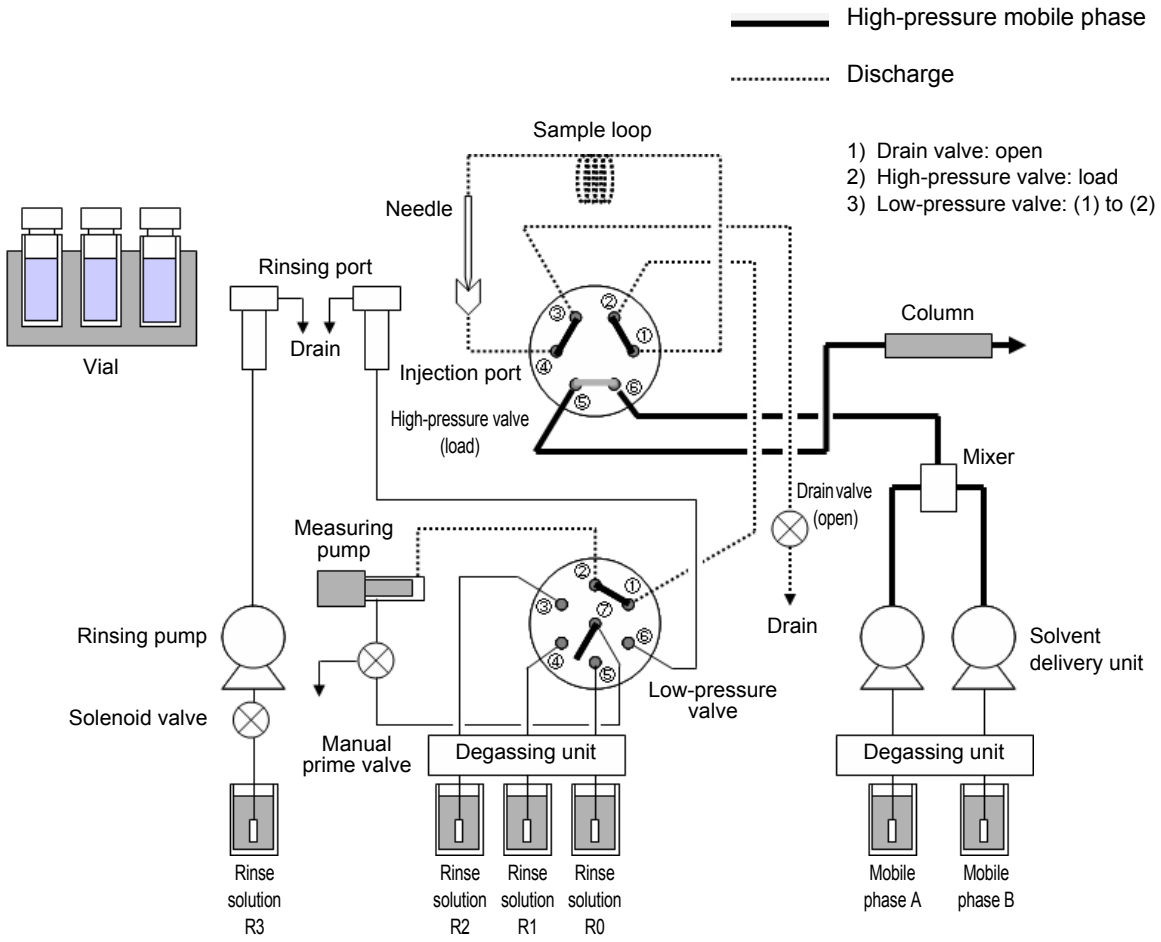


Fig. 1.13

■ External Rinsing of the Needle

This is the function that rinses the external surface of the needle by dipping the needle in the rinsing port or pumping the rinse solution using a rinsing pump before and after sample aspiration in order to eliminate contamination from the external surface of the needle.

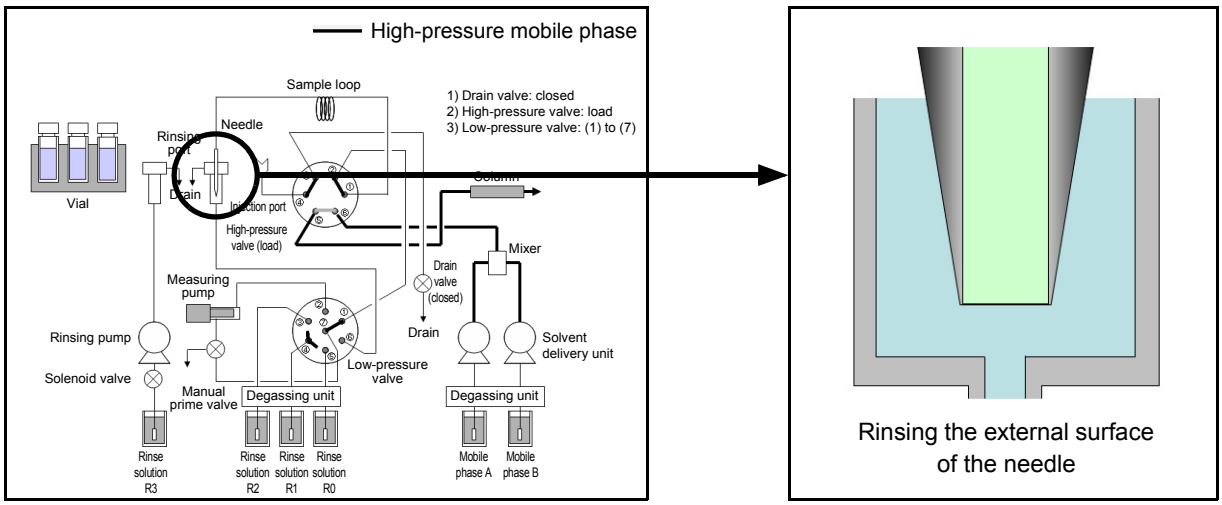


Fig. 1.14

■ Internal Rinsing of the Needle

This is the function that rinses the HPLC flow line using a maximum of three kinds of rinse solution (R0, R1, R2) during or after analysis in order to eliminate contamination from the flow line in the autosampler including the needle, the injection port, the sample loop, and the high-pressure valve. To perform internal rinsing of the needle, set "2" (internal/external rinsing of the needle) at [RINSE TYPE] in the parameter setting group.

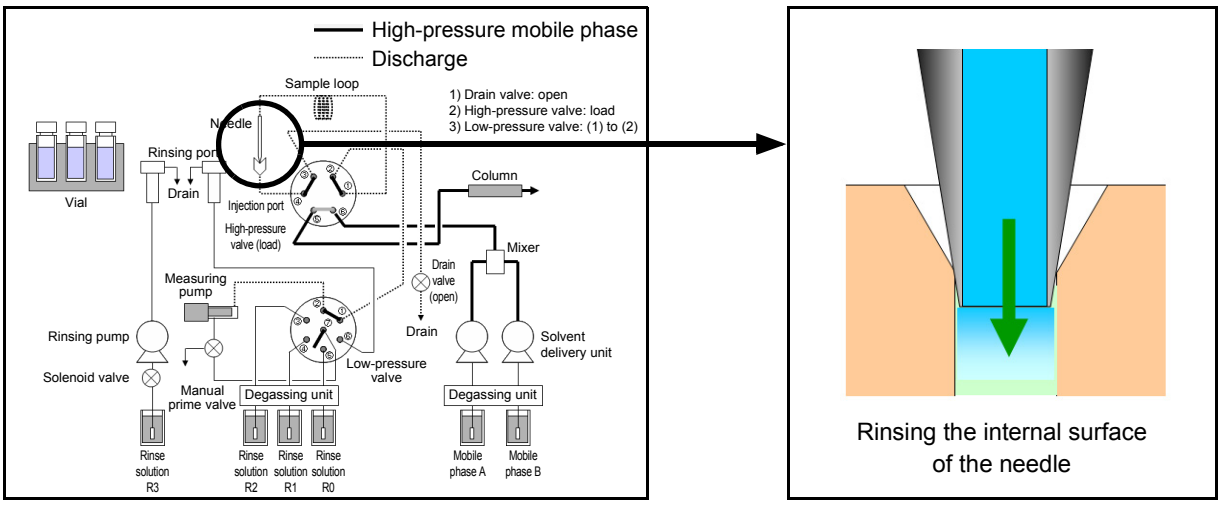


Fig. 1.15

■ Rinsing of the Injection Port

This is the function that rinses the injection port immediately after internal rinsing of the needle. This function is available only when internal rinsing of the needle is used. To perform rinsing of the injection port, set "2" (internal/external rinsing of the needle) at [RINSE TYPE] and select the solvent to be used at [INJ.P RINSE]. The major rinsing sequence is shown below.

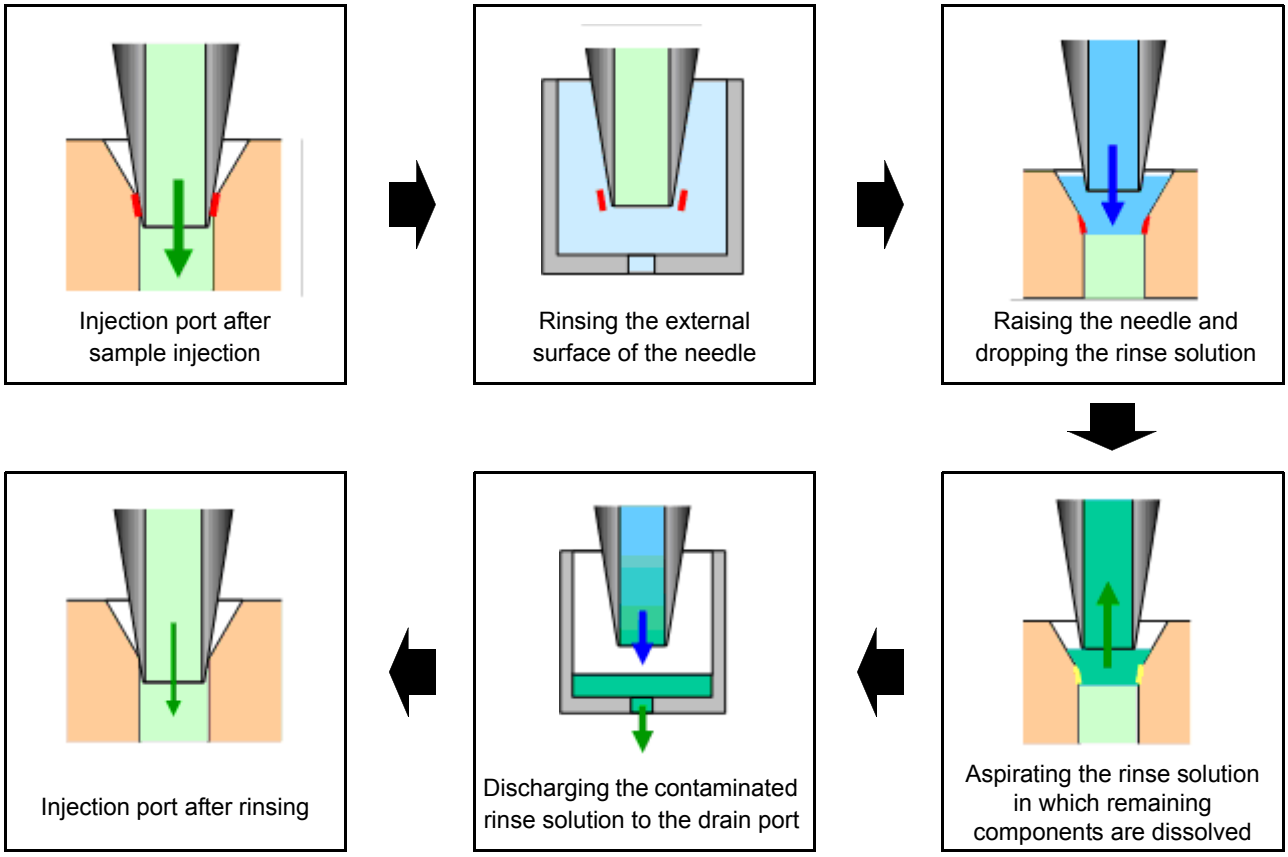


Fig. 1.16

■ Holding Capacity in the Flow Line (Reference)

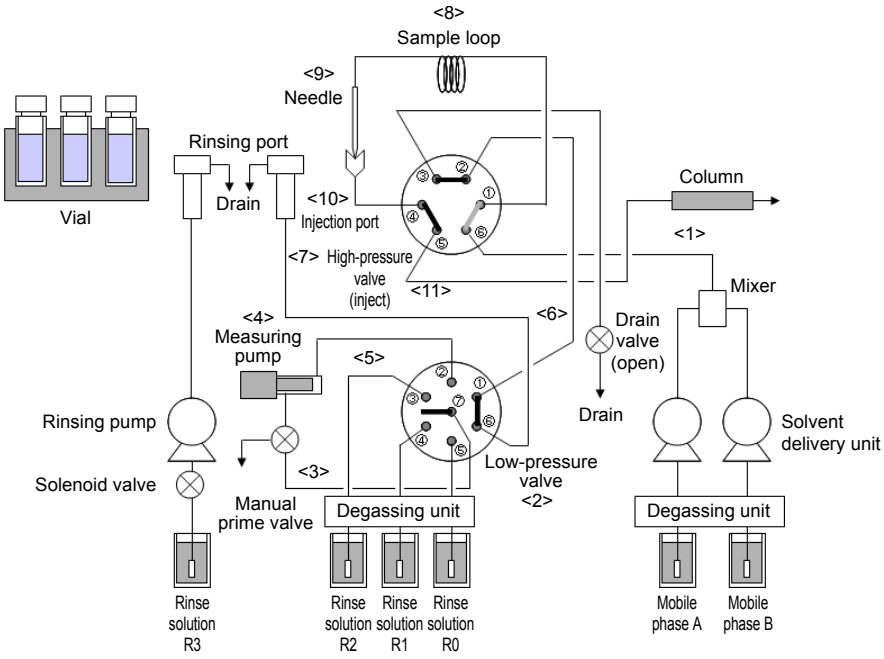


Fig. 1.17

NOTE

When "2" is set at [RINSE TYPE] (internal/external rinsing of the needle) and multiple rinse solutions are used for rinsing the flow line, rinse solution R0 must be used as mobile phase (initial concentration for gradient analysis).

No.	Flow Line	I.D. x Length (mm)	Capacity (μL)	Capacity from Mixer Outlet to Column Inlet (μL)	Capacity for Internal Rinsing of Needle (μL)
①	Mixer - HPV No. ⑥	ϕ0.3 × 300	21.2	21.2	
②	Between LPV ports	-	2.6		5.2
③	LPV No. ⑦ - Measuring pump inlet	ϕ0.5 × 135	26.5		26.5
④	Inside the measuring pump	-	34.5		34.5
⑤	Measuring pump outlet - LPV No. ②	ϕ0.5 × 170	33.4		33.4
⑥	LPV No. ① - HPV No. ②	ϕ0.5 × 360	70.7		70.7
⑦	Between HPV ports	-	0.7	1.4	1.4
⑧	Sample loop	ϕ0.3 × 1200	84.8	84.8	84.8
⑨	Needle	-	11.7	11.7	11.7
⑩	Injection port	-	1.0	1.0	11.0
⑪	HPV No. ⑤ - Column inlet	ϕ0.1 × 800	6.3	6.3	
			Total	126.4*1	269.2*2

*1 Equivalent to the delay volume for high-pressure gradient analysis (excluding the mixer capacity)

*2 Equivalent to the capacity in the flow line to be rinsed when performing internal rinsing of the needle

1.5 Component Parts

This instrument consists of the standard parts listed below. Check the parts against this list after unpacking. The standard parts provided depend on the power supply voltage. (See below.) After unpacking, verify that the correct types and quantity of parts have been provided.
The 2-digit numbers in the remark column in the table below indicate the power supply voltages for the part. -32, -42 indicates use with a 120 V power supply and -38, -48, -58 is a 220-240 V power supply.

No.	Part Name	Part No.	Q'ty	Remark
–	SIL-30AC		1	
–	Instruction Manual, SIL-30AC	S228-90775	1	
–	Nexera X2 System Guide	S228-90870	1	
–	Nexera X2 Safey Guideline	S228-91060	1	
–	Parts pack	S228-39988-93	1	-32, -38
		S228-39988-43		-42, -48, -58
1	Vial 1.5 mL (containing 10 pcs.)	S228-38446-91	1	
2	EVENT cable ASSY	S228-28253-91	1	
3	Rinsing port cover	S228-48328-01	1	
	Rinsing port cap (NO HOLE)	S228-47973-01	2	
	Rinsing port cap	S228-47973-02	2	
4	Suction filter ASSY	228-45708-93	3	Part No. of the replacement: S228-45708-91
	Tag (RINSE: R0)	S228-53057-41	1	
	Tag (RINSE: R1)	S228-53057-42	1	
	Tag (RINSE: R2)	S228-53057-43	1	
5	PTFE tube (ϕ7 mm $\times$ 800 mm)	S016-37507	1	
6	Drain tube ASSY (ETFE)	S228-44608-91	1	
7	Drain tube clamp	S228-43347	1	
	PTFE tube (ϕ8 mm $\times$ 50 mm)	S016-37519	1	
8	Cable clamp, UL-23G	S072-60314-03	1	
9	Cable, HFBR3600-1-021	S070-92025-51	1	
10	Drain tube SI	S228-25162-03	1	
11	Drain tube ASSY 30AC	S228-43271-93	1	
12	Cooling rack (1.5 mL $\times$ 105 pc)	S228-50761-92	1	
13	AC Power cord (for UL/CSA)	S071-60821-08	1	-32, -42
	AC Power cord (for VDE)	S071-60825-51	1	-38, -48, -58

No.	Part Name	Part No.	Q'ty	Remark
14	Control vial rack 1.5 mL	S228-44634-91	1	
15	SUS tubing HP OUT (0.1 × 600 mm)	S228-53184-91	1	I.D. identification mark*1
16	Jig rotor	S228-48899-91	1	
17	Tool for needle seal XR	S228-50570	1	
18	Clamp, DKN-10GSP	S072-60319-01	1	
19	Jig, rack teaching	S228-50895-91	1	

*1 This is the tubing of 0.1 mm ID, 600 mm long, to be used for connection of column oven CTO-20A/20AC. To identify the inner diameter, 0.1-mm ID tubing has a black tag. Red marking is provided to the tubing dedicated to the Nexera X2 series. Do not use any tubing not equipped with red marking to the Nexera X2 series.
Two UHPLC fittings (part No. S228-56867-41) are included in the package.

1.6 Optional Parts

Optional units available for this instrument are listed below.
Check the User Manual included with the optional equipment for installation instructions.
For information about other optional units not listed below, contact your Shimadzu representative.

■ PEEK Needle Seal

Used for pumping mobile phase exceeding pH 9.
When using the needle seal, the maximum allowable pressure is limited to 66 MPa.
If the pH exceeds 9, replace the needle seal only.

Part Name	Part No.
PEEK needle seal 30A	S228-53178-91

■ Loop Injection Base Kit

Used to switch the injection method from total injection method to loop injection method.

Part Name	Part No.	Remark
Loop injection base kit	S228-45421-91	(Major parts) SUS tubing (loop - LPV#1) SUS tubing HP IN (0.1 × 600 mm) Installation manual

NOTE

No sample loop is included in the loop injection base kit. Prepare a sample loop suitable for the injection volume from optional parts shown below.

■ Sample Loops for Loop Injection

Part Name	Part No.
Sample loop 5 μL	S228-52612-42
Sample loop 20 μL	S228-52612-43

⚠ CAUTION

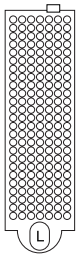
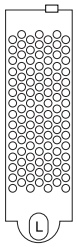
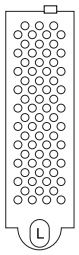
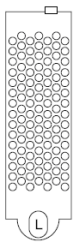
The following optional and maintenance parts for other SIL-20A series autosamplers are not available for this instrument.

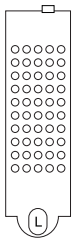
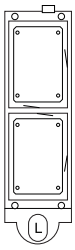
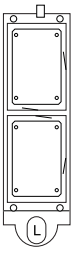
Part	Part No.	Remark
Sample loop ASSY (100 μL injection loop) Sample loop ASSY (500 μL injection loop) Sample loop ASSY (2 mL injection loop)	S228-45402-95 S228-45405-93 S228-45405-94	Because the sample measuring flow line is not the same, any sample greater than 50 μL cannot be measured or injected with the instrument.
Needle seal PEEK 20A Needle seal XR Needle seal XR kit (includes 3 pcs) Needle seal XR kit VESPEL needle seal	S228-42325-01 S228-50390 S228-50452-92 S228-50452-93 S228-48258-91	Pumping at a high pressure may cause leaks from the needle seal.
High-pressure valve rotor	S228-41310-92	The stator position does not match the flow line channel, resulting in leaks or false analysis.
High-pressure valve rotor XR	S228-48854	Pumping at a high pressure may cause leaks.
High-pressure valve stator XR	S228-48858-91	This part cannot be attached to the needle seal dedicated to this instrument.
Male nut fitting	S228-48485-91	Pumping at a high pressure may cause the tubing to be disconnected.
SUS housing kit XR modification kit	S228-46913-96 S228-50368-92	Pumping at a high pressure may cause leaks.

Red marking is provided to the tubing dedicated to the Nexera X2 series. Do not use any tubing not equipped with red marking to the Nexera X2 series.

■ Sample Vial Racks, Microtiter Plate Racks

Please select according to the purpose and content of analysis.
Contact your Shimadzu representative regarding recommended microtiter plates.

Name	Screen Display	Sample Vial Type, Volume	Capacity	Part No.
1 mL Sample Vial Rack 	1 mL-C	Glass 1 mL	175 vials	S228-37614-92
1.5 mL Sample Vial Rack 	1.5 mL	Glass 1.5 mL, Glass 1.1 mL, Glass w/spacer 0.3 mL, Plastic (PP) 1 mL, 0.2 mL	105 vials	S228-45409-92
1.5 mL Sample Vial Cooling Rack 	1.5 mL-C	Glass 1.5 mL, Glass 1.1 mL, Glass w/spacer 0.3 mL, Plastic (PP) 1 mL, 0.2 mL	70 vials	S228-44617-92
1.5 mL Sample Vial Cooling Rack (105 vials) 	1.5 mL	Glass 1.1 mL, 1.5 mL, Glass w/spacer 0.3 mL, Plastic (PP) 0.2 mL, 1 mL	105 vials	S228-50761-92

Name	Screen Display	Sample Vial Type, Volume	Capacity	Part No.
4 mL Sample Vial Rack 	4 mL-C	Glass 4 mL, Plastic (PP) 4 mL, 0.3 mL to accommodate 4 mL sample vials	50 vials	S228-37616-92
Microtiter Plate Rack 	MTP-96, MTP-384	Microtiter plates (96-well, 384-well)	2 plates	S228-37545-92
Deep-well MTP Rack 	DWP-96, DWP-384	Deep-well (96-well, 384-well)	2 plates	S228-37546-92
Control Vial Rack	CntR	Glass 1.5 mL, 1.1 mL, Glass w/spacer 0.3 mL, Plastic (PP) 1 mL, 0.2 mL	10 vials	S228-44634-91
Changer Rack	Changer	For optional rack changer Microtiter plate (96-well) Deep-well (96-well)	1 plate	S228-45499-92

Temperature Control Performance

The following table indicates the temperature control performance when using various types of optional sample racks.

Sample Rack Name	Temperature Control Performance*
Sample vial rack for 1 mL vials	Vial bottom temperature = temperature setting ±3 °C
Sample vial cooling rack for 1.5 mL vials	Vial bottom temperature = temperature setting ±3 °C
Sample vial rack for 4 mL vials	Vial bottom temperature = temperature setting ±3 °C
Microtiter plate rack	Well bottom temperature = temperature setting ±6 °C
Deep-well MTP rack	Well bottom temperature = temperature setting ±6 °C

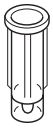
∗Cooling Performance of the Sample Cooler

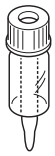
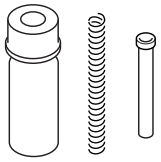
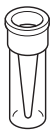
The cooling performance varies depending on various conditions, such as the surrounding environment and sample volume in a vial . The value indicated in the upper table is measured on condition of the following.

Sample Rack Name	Environment	Liquid Volume	Vial used	Measurement site
Sample vial rack for 1 mL vials	30 °C 70%	Water 700 µL	Flat bottom glass vial	Vial bottom center
Sample vial cooling rack for 1.5 mL vials	30 °C 70%	Water 1 mL	Glass vial	Vial bottom center
Sample vial rack for 4 mL vials	30 °C 70%	Water 3 mL	Glass vial	Vial bottom center
Microtiter plate rack	30 °C 70%	Water 200 µL	Nalge Nunc round bottom MTP	Well bottom center
Deep-well MTP rack	30 °C 70%	Water 1 mL	Nalge Nunc round bottom MTP	Well bottom center

■ Sample Vials

The following types of sample vials can be used in the sample vial racks.

	Part Name	Capacity	Material	Part No.	Application	Remark
	4 mL Sample Vial 	4 mL	Borosilicate glass	S228-21287-91	General	100 ea. w/cap and silicone rubber septum
×	4 mL Sample Vial	4 mL	Polypropylene	S228-31537-91	General	
	1.5 mL Sample Vial 	1.5 mL	Borosilicate glass	S228-15652-92	General	
×	1.1 mL Sample Vial 	1.1 mL	Borosilicate glass	S228-21283-91	General and small-capacity Needle stroke is less than 50 mm	100 ea. w/cap and silicone rubber septum
×	1 mL Sample Vial 	1 mL	Polypropylene Cap: polyethylene	S228-31600-91	General and small-capacity Needle stroke is less than 50 mm	200 ea. w/cap (See TIP " Sample vials" P. 28.)

	Part Name	Capacity	Material	Part No.	Application	Remark
	1 mL Sample Vial	1 mL	Borosilicate glass Cap: polyethylene	S228-39699-91	General	250 ea. w/cap
×	0.3 mL Sample Vial 	300 µL	Borosilicate glass	S228-16847-92	Small-capacity Needle stroke is less than 50 mm	100 ea. w/cap and silicone rubber septum
	0.3 mL Sample Vial (Spares)	300 µL	Borosilicate glass	S228-16850-91		100 ea.
×	0.3 mL Sample Vial 	300 µL	Borosilicate glass	S228-21284-91	Small-capacity	100 ea. w/spring, used inserted into aforementioned 4 mL sample vial
	0.3 mL Sample Vial (Spares)	300 µL	Borosilicate glass	S228-21285-91		100 ea.
×	0.2 mL Sample Vial 	200 µL	Polypropylene Cap: polyethylene	S228-35217-91	Disposable vials for small-capacity analysis Needle stroke is less than 50 mm	100 ea. w/cap (See TIP " Sample vials" P. 28.)
	Silicone Rubber Septum	-	Silicone rubber w/ PTFE cover	S221-26718-93	1.5 mL Sample Vial 1.1 mL Sample Vial 0.3 mL Sample Vial (S228-16847-92)	100 ea.
	PTFE Septum	-	PTFE	S228-15655-91	Same above	100 ea.
	Silicone Rubber Septum	-	Silicone rubber w/ PTFE cover	S221-21290-91	4 mL Sample Vial	100 ea.
	PTFE Septum	-	PTFE	S228-23469-91	4 mL Sample Vial	100 ea.

∗ PTFE (Poly Tetra-Fluoro Ethylene)

NOTE

Using Sample Cooler

Sample vials marked with a × may have different thermal conductivity properties due to the shape or material of the sample vial. Consequently, there may be circumstances in which the sample cooler set temperature and the internal temperature of the sample vials may differ.

■ Microtiter Plates

When using a sample cooler, do not use microtiter plates with a raised bottom, shallow wells, and a gap above 2 mm between the bottom of the wells and the bottom of the plate. Using this type of plate will create a gap between the cooling plate on the rack and the microtiter plate or deep-well plate. Water condensation may occur in this gap, making it impossible to obtain accurate analysis results.

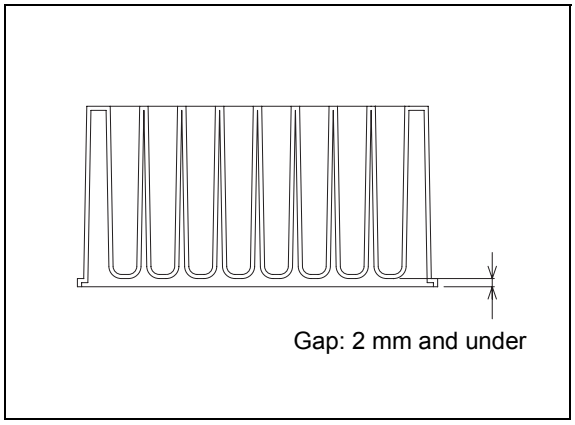


Fig. 1.18

1) Well-fitted sealing mat

Plate Type	Product	Contact	Remark
Microtiter plate	267245 series	Nalge Nunc International	Material: PP, capacity: 0.5 mL
Deep-well plate	278752		Material: PP, capacity: 2 mL
	AXYGEN P-DW-20-C	Greiner	Material: PP, capacity: 2 mL
Mat	AXYGEN AM-2ML-RD		Material: Silicone * The mat may be raised as time passes if large volumes are filled in the wells.
	276011	Nalge Nunc International	
	276002 series		Material: Thermoplastic elastomer * The mat may be raised and cannot be used in the rack changer.

2) Heat sealing mat

Plate Type	Product	Contact	Remark
Microtiter plate	4titude 4Ti-0110	AB gene	Material: PP, capacity: 0.3 mL
Deep-well plate	4titude 4Ti-0130		Material: PP, capacity: 1.85 mL
Mat	ABgene Easy Peel AB-0745		Material: Foil
	4titude Peel-Seal 4Ti-0521		Material: Foil
	4titude Pierce-Seal 4Ti-0531		Material: Foil
	Permanent sheet PP 298-37851	Wako Pure Chemical Industries	Material: Foil

3) Adhesive sealing mat

Plate Type	Product	Contact	Remark
Microtiter plate	267245 series	Nalge Nunc International	Material: PP, capacity: 0.5 mL
Deep-well plate	278752	Greiner	Material: PP, capacity: 2 mL
	AXYGEN P-DW-20-C		Material: PP, capacity: 2 mL
Mat	USA SCIENTIFIC NAL-96 Sealing Film 2923-5000	USA Scientific	Material: Upper: PE, lower: PP * No adhesives at wells * Exclusive for 96-well plates * The mat may be raised as time passes if large volumes are filled in the wells.

NOTE

The sealing mats having the features (1) and (2) described below may cause the needle or needle seal flow line to be clogged. If you use such a sealing mat, pay careful attention to clogging.

1. Adhesive-backed mats (adhesive on the entire face that will contact the plate)
   Regardless of the sample solvent type, as the instrument is used, the adhesive may attach to the external needle surface and the internal surface of the flow line, causing sample aspiration or peak area calculation to fail, or clogging of the flow line.
2. Mats made of PET (polyethylene terephthalate)
   If the sample solvent is acetonitrile based or DMSO based, the mat is likely to swell and may be creased after sealing, impairing airtightness.
   When the sample solvent is water based or methanol based, there will be no problem in practical use.

NOTE

Sample vials

Both glass and plastic (polypropylene) sample vials are available for use, however, to prevent problems during analysis the following precautions should be observed when selecting the type of sample vial.

Glass Sample Vials

There is a possibility that ionic substances, such as acids or bases, may be adsorbed onto the glass surface. If ionic substances are analyzed under these circumstances, precision will be poor and the reliability of the analysis will be lost. When using glass sample vials, employ a sample solvent to restrict adsorption of the substance. The following kinds of sample solvents are used.

- 10 mM to 100 mM aqueous perchloric acid solution or a mixture of that and an organic solvent.
  (Use acetonitrile, methanol, or ethanol as the organic solvent.)
- An organic solvent solution of 10 mM trifluoroacetic acid (TFA).
  (Use acetonitrile, methanol, or ethanol as the organic solvent. However, trifluoroacetic acid will be detected when detecting absorbance in the 200 nm to 220 nm range.)

In the event that one of these sample solvents cannot be used, use a plastic sample vial. Note that alkalis and hydrogen fluorides will chemically attack glass.

Plastic Sample Vials

The hydrophobic properties of substances are the cause of surface adsorption in plastic sample vials. The precision of analysis will be poor in this situation as well. The higher the polarity of the sample solvent, the greater the effect. The adsorption of hydrophobic substances can be suppressed through the use of low-polarity sample solvents, but if the polarity is too low, the possibility arises of additives in the plastic dissolving off the surface of the sample vial. Consequently, use the following sample solvents.

- Mixtures of water or a buffering solution with an organic solvent.
  The organic solvent content should be 20% to 50% (V/V). (Use acetonitrile, methanol, or ethanol as the organic solvent.)

In the event that one of these sample solvents cannot be used, use a glass sample vial. As discussed above, note that organic solvents promote the deformation of plastic.

■ Rack Changers II

Rack changers make it possible to perform analysis with up to 12 microtiter plates or deep-well plates.

Part Name	Part No.	Remark
Rack Changer II	S228-45164-31 S228-45164-32 S228-45164-38 S228-45164-41 S228-45164-42 S228-45164-48 S228-45164-58	With rack cooling function

■ Rack Changer II 1.5 mL Sample Vial Plate Kit

Serial analysis is possible with up to twelve 1.5 mL sample vial plates using Rack Changer II and this kit.
A maximum of 54 sample vials (1.5 mL) can be set on one 1.5 mL sample vial plate.

Part Name	Part No.	Remark
1.5 mL sample vial plate kit (3 plates)	S228-50830-91	Three 1.5 mL sample vial plates (for one stack) are included. If you use 12 plates, 4 kits are required.

When using 1.5 mL sample vial plates on the Rack Changer/C not equipped with a window on the front panel, be sure to use the Extended Plate Kit for Rack Changer (part No.: S228-50829-91).

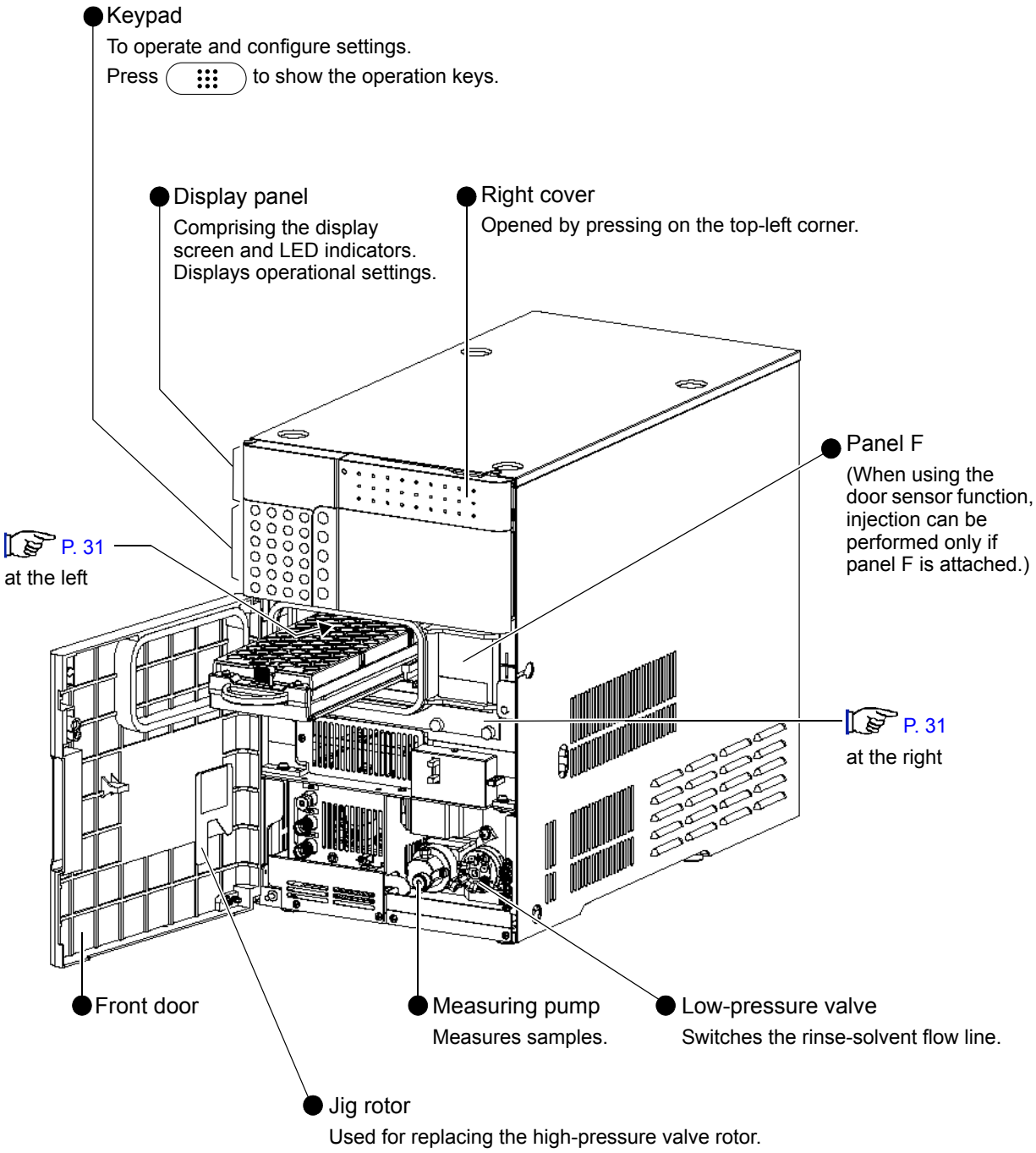
■ Maintenance Kit

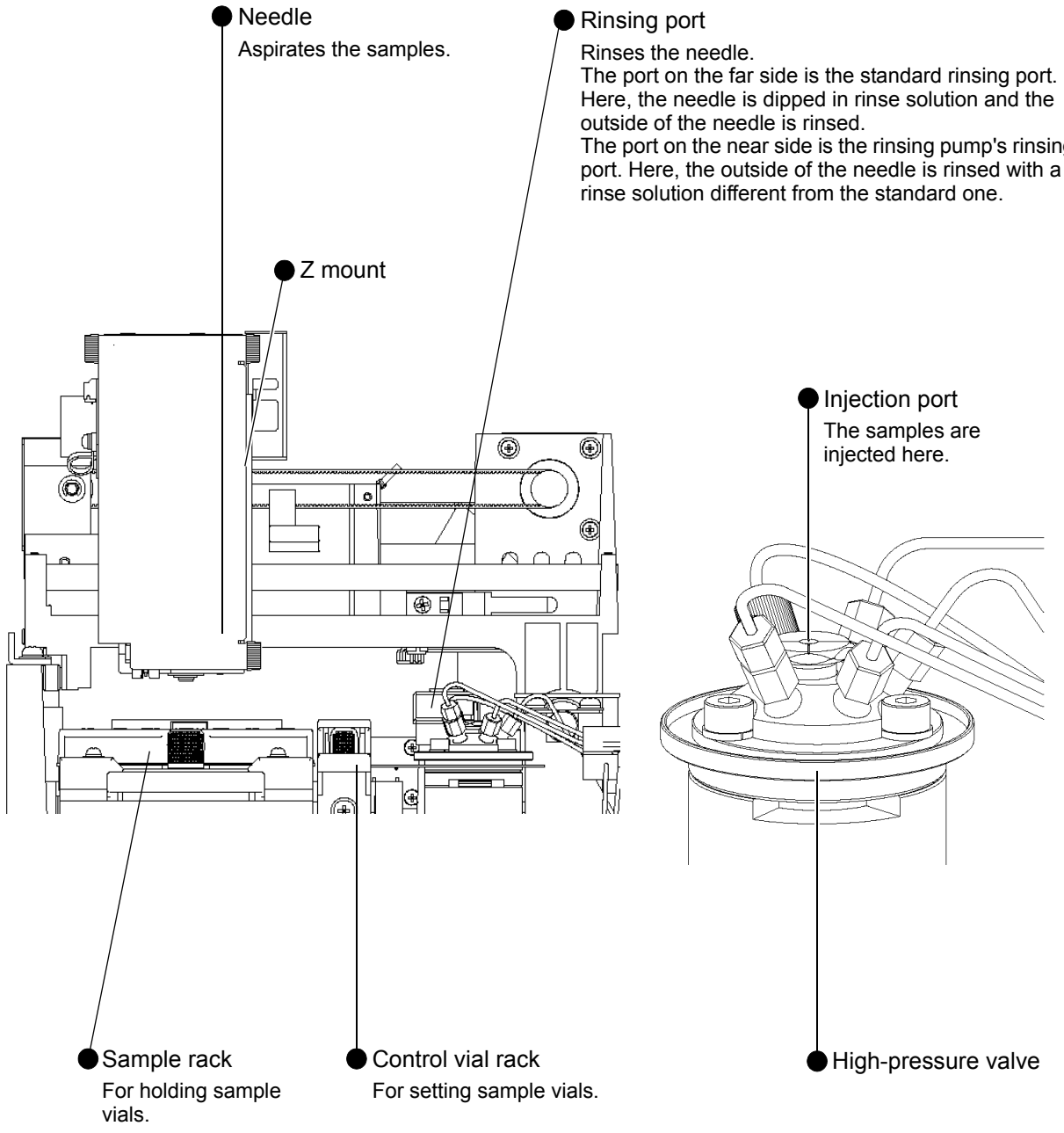
A set of consumable parts and plumbing parts. "8.2.3 Maintenance Kit" P. 283

Part Name	Part No.	Remarks
Maintenance Kit	S228-45413-98	Rotor/stator for high-pressure valve, rotor for lower-pressure valve, etc. are included.

2 Parts Identification and Function

2.1 Front Section Interior




Needle
Aspirates the samples.
Rinsing port
Rinses the needle.
The port on the far side is the standard rinsing port. Here, the needle is dipped in rinse solution and the outside of the needle is rinsed.
The port on the near side is the rinsing pump's rinsing port. Here, the outside of the needle is rinsed with a rinse solution different from the standard one.
Z mount
Injection port
The samples are injected here.
Sample rack
For holding sample vials.
Control vial rack
For setting sample vials.
High-pressure valve

● Rinsing pump inlet

Introduces a rinse solution for external rinsing of the needle from the reservoir bottle to the rinsing pump. No degassing unit is used.

● Rinsing pump outlet

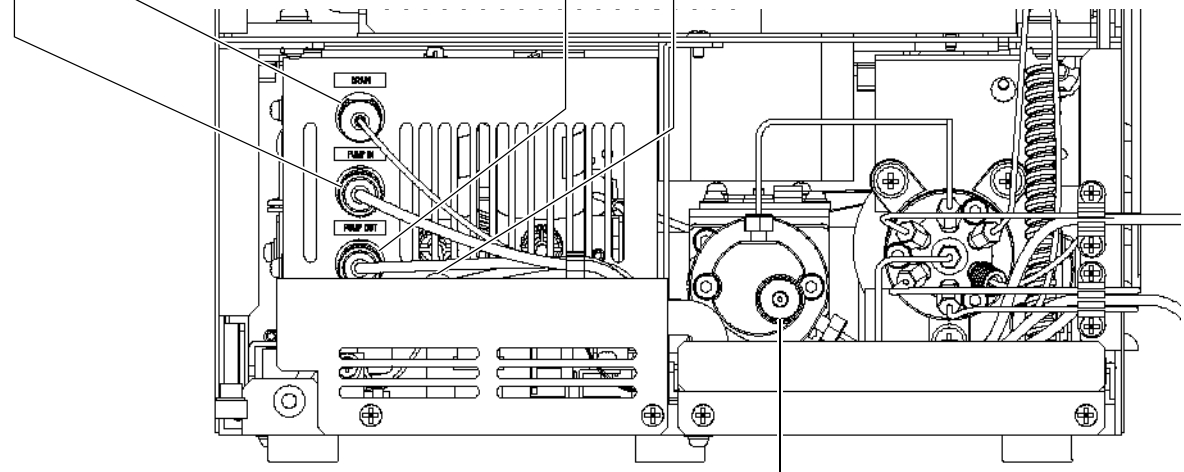
Delivers a rinse solution for external rinsing of the needle from the rinsing pump to the rinsing port.

● Drain valve inlet

Introduces waste liquid into the solenoid valve after internal rinsing of the needle.

● Drain valve outlet

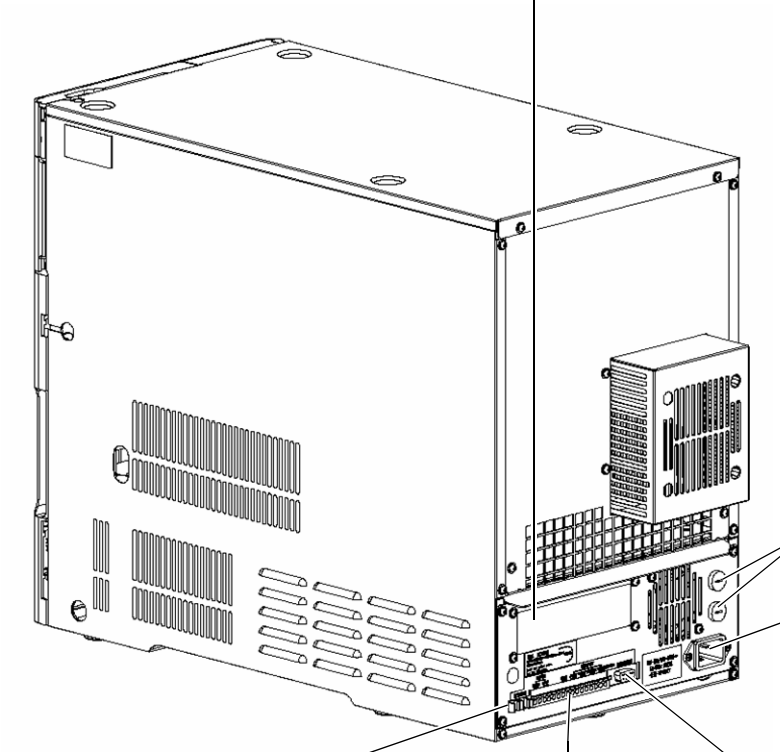
Discharges waste liquid out of the instrument through the solenoid valve after internal rinsing of the needle.



● Manual prime valve

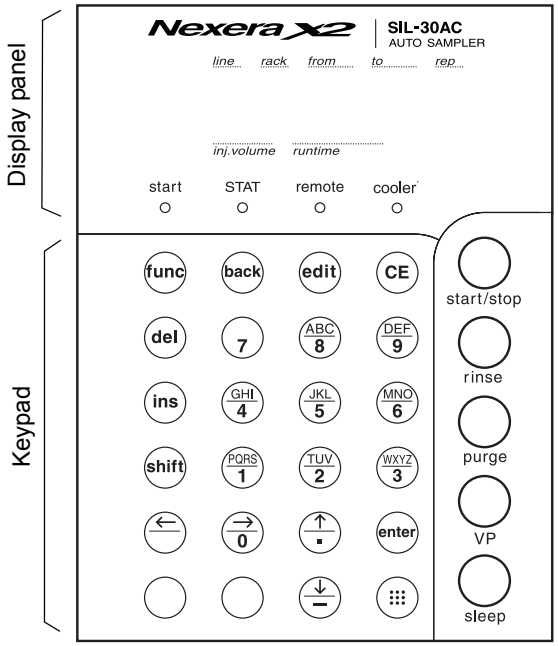
Used to draw a rinse solution using the manual syringe provided with the instrument.

2.2 Back


Houses the CBM-20Alite.
Fuse holders
AC Power cord connector
Changer connector
[REMOTE] connector
Connects to the CBM-20A.
External input/output terminal

2.3 Names and Functions of Displays and Keypad

This instrument is controlled through the keypad.
The display allows verification of the instruments status.

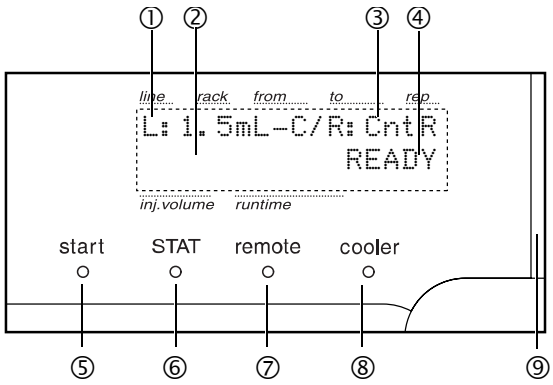


NOTE

The display screen may become hot when in use.

2.3.1 Display Panel

The display panel consists of a display screen and LED indicators.
Names and functions of the screen areas and the indicators are given below.

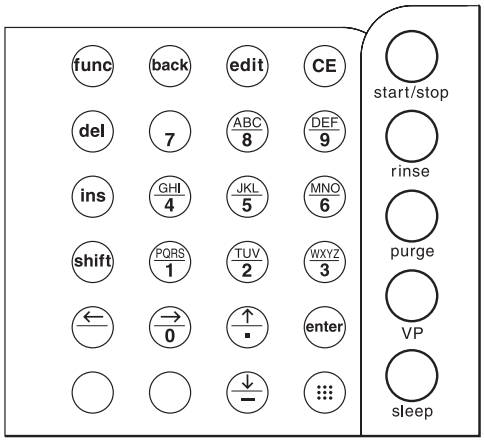


No.	Display or Indicator	Function
①	Rack display	Displays the model of the sample rack set in the autosampler.
②	Status display 1	When connected to a rack changer and using a rack-changer rack, [CNG-LINK] is displayed. When [20A] is set for [OP MODE], [20A MODE] is displayed. "[OP MODE]" P. 148
③	Control vial rack display	Indicates whether or not there is a control vial rack.
④	Status display 2	Displays the status of operation.
⑤	start LED	Illuminates when sample injection starts.

No.	Display or Indicator	Function
⑥	STAT LED	Priority analysis indicator ON when priority analysis is executed.
⑦	remote LED	Remote control mode indicator ON when the instrument is controlled by system controller.
⑧	cooler LED	Illuminates when using a sample cooler. Flashes if the monitor temperature is not within 1 °C of the set temperature.
⑨	Status indicator	Green : when power is ON. Flash : while the needle is moving up and down on the sample rack. Red : when an error is generated. Orange : during sleep mode.

2.3.2 Keyboard

The 27 keys on the keypad are used to operate the instrument and set parameters. The functions of the keys are given below.



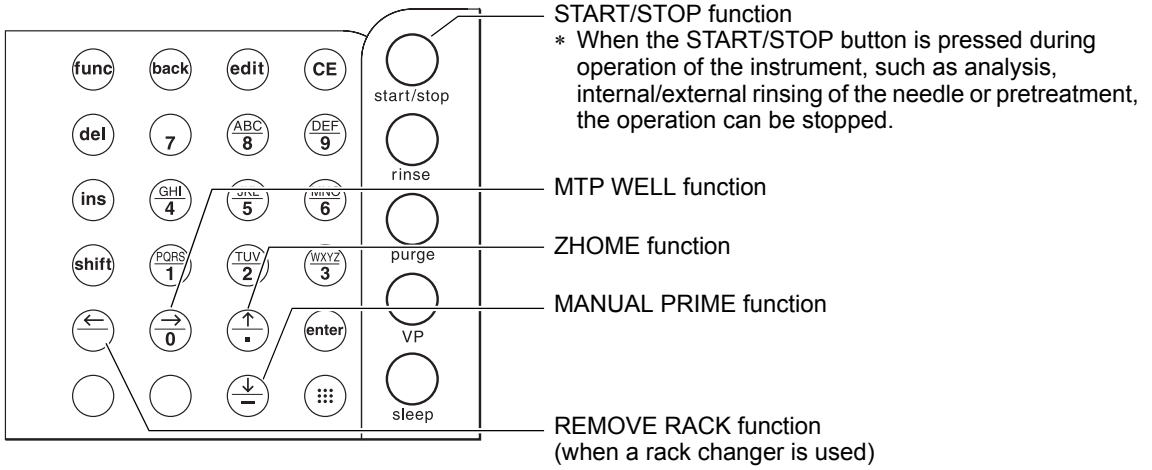
Key	Name	Function
⁞⁞	Display key	To show the operation keys.
func	Function key	Used to select auxiliary functions. Press this key after selecting the VP function groups to go to the VP function menus.
back	Back key	Press this key while setting auxiliary functions to go back to the previous setting screen.
edit	Edit key	Activates edit mode for repeat injection table or analysis sequence table (from initial screen).
CE	Clear key	Initializes the screen. ∗ Cancels values input since [Enter] was last pressed. ∗ Clears error messages and cancels alarms.

Key		Name	Function
del		Delete key	Deletes the set line in the repeat injector table or the sample table.
ins		Insert key	For adding lines to the repeat injection or analysis sequence table.
shift		Shift key	For performing the operations for arrow keys indicated in the top part of the numeric keypad. When this key is pressed, [Shift pressed] appears on the display screen. Press any key or shift again to cancel [Shift pressed].
ABC - WXYZ		Letter keys	For entering the well number when using a microtiter plate or a deep-well plate.
Shift +	←	Left cursor key	For moving the cursor to the left on table parameter setting screens in the repeat injection or analysis sequence table. For moving from the initial screen directly to the special rack removal setting (REMOVE RACK) screen when a rack changer is connected.
	→	Right cursor key	For moving the cursor to the right on table parameter setting screens in the repeat injection or analysis sequence table. For moving from the initial screen directly to the microtiter plate setting (MTP WELL) screen.
	↑	Up cursor key	For moving the cursor up on table parameter setting screens in the repeat injection or analysis sequence table. For moving from the initial screen directly to the needle position moving (ZHOME) screen.
	↓	Down cursor key	For moving the cursor down on table parameter setting screens in the repeat injection or analysis sequence table. For moving from the initial screen directly to the rinse solution flow line purging (MANUAL PRIME) screen.
. - 9		Numeric keypad	For entering numerical values. Pressing the ↑ . key moves from the initial screen directly to the needle position moving (Z HOME) screen.
enter		Enter key	Validates input values.
−		Minus key	For displaying a minus sign on the cooler temperature setting screen. For moving from the initial screen directly to the rinse solution flow line purging (MANUAL PURGE) screen.
start		Start key	Starts sample injection. Stops sample injection.
rinse		Rinse key	For rinsing needle in rinse solution.
purge		Purge key	Pumps rinse solution through flow lines for a specified period of time.
VP		VP key	For switching from initial screen to VP mode.

Key	Name	Function
sleep	Sleep key	Turns off display screen. Has no effect on operation.

NOTE

When the operation key on the keypad is pressed while the display panel shows the initial screen, the corresponding function will be executed or will be selected directly.



3 Preparation

3.1 Turning Power ON/OFF

1 Press the power switch to turn the power ON.
Press it again to turn the power OFF.

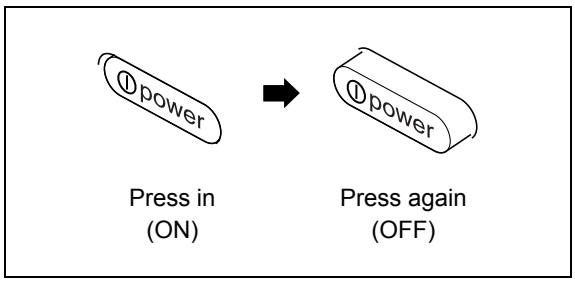
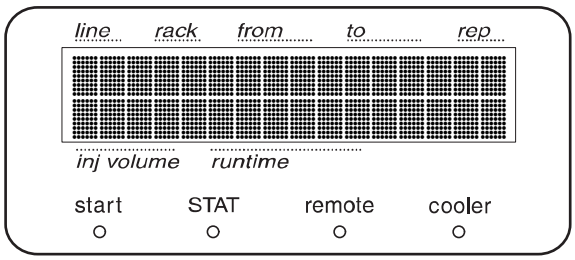
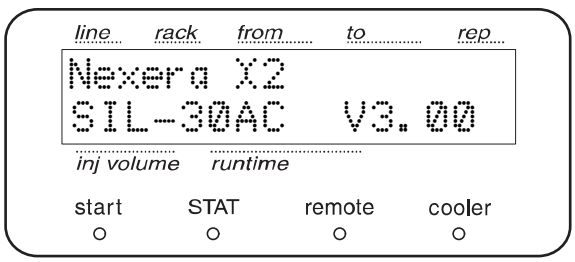


Fig. 3.1

2 When the power is first turned on, all the dots in the display matrix and all the indicators illuminate, as shown on the right.

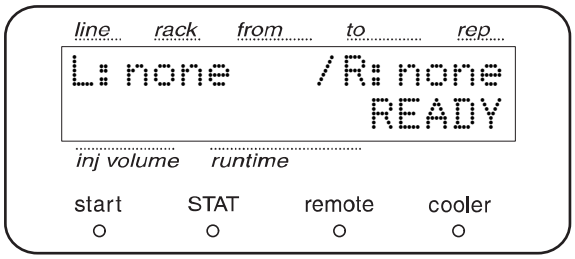


3 The memory is automatically tested, and after the memory check passes, the version number of the control program is displayed momentarily.
The status indicators turns green.
The screen displays the S/W version number [V*.**].



4 When the needle is in the standby state (in the injection port position), the initial screen on the right appears on the display.

Initial Screen



NOTE

- If there is a large amount of data to be backed up, such as when there are many lines set in the sample table, it may take a while before initialization starts.
- If an error is detected, an alarm sounds and an error message appears.
"6.3 Error Messages" P. 222

⚠ CAUTION

There are two door sensors provided: a micro switch sensor and a photo sensor. Ensure that the photo sensor has not failed during the daily inspection. Turn ON the power switch on the instrument, and when initialization is complete, open the door and check that "DOOR IS OPEN" is displayed on the display panel of the instrument. Then close the door and check that it changes to "READY".

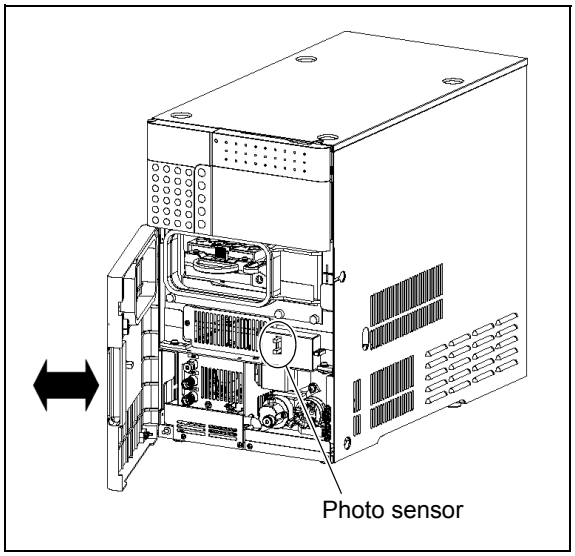


Fig. 3.2

3.2 Preparing Rinse Solution Bottles and Waste Containers

Prepare rinse solution bottles and waste containers before connecting the plumbing.

⚠ WARNING

Do not use cracked or damaged waste container(s). They could be broken.

⚠ CAUTION

The waste container must be positioned lower than the instrument (for example, on the floor). If it is positioned higher than the instrument, liquid will not be drained, and will leak from the connections. Check that drain tubings are attached in the same way as shown in "2.3.1 Connecting Leakage Drain Tubing" in the Nexera X2 SYSTEM GUIDE. Attach a drain tube clamp (accessory) to the mouth of the waste container, and make sure that the tip of the drain tubing connected to the rinse solution outlet is not immersed in the waste (The upper outlet is for the rinse solution, the center outlet is for condensation and the lower outlet is for liquid leaked inside the equipment.). If the tip of the drain tubing is immersed in the waste, the waste solution may flow inside of the instrument and may break the instrument.

3.3 Rinse Solution Selection

Select the rinse solution as follows, depending on the mobile phase. To ensure the accuracy in sample injection, be sure to install a degassing unit in the flow line of rinse solution and mobile phase.

■ For reversed phase, ion exchange, and aqueous normal phase

- The ratio of methanol to water should be 50%/50%(V/V). If precipitation occurs upon sample contact, select a rinse solution without salts, as for the mobile phase.
- When the target compound is acid, base, or ionic substance, and sample is likely to remain on the outside surface of the needle, add acid such as formic acid and acetic acid to the organic solvent of methanol, acetonitrile, etc., or use a 0.1% TFA solution or organic solvent solution, or their mixture solution.

■ For non-aqueous normal phase, GPC

- Use the same solvent (s) used for the mobile phase.
- When the target compound is an acid, base, or ionic substance, and rinse mode is required, use a 0.1% TFA aqueous solution, an organic solvent solution, or a mixture of both.

⚠CAUTION

When using a highly volatile acid for rinse solution:
If rinse solution contains highly volatile acids (1% or higher formic acid or acetic acid solution, 0.1% or higher TFA solution, etc.), follow the instructions given below. If analysis is continued for a long time with such a rinse solution, volatile components may cause metals in the instrument to corrode.

- When the analysis is complete, purge the rinse solution with water or methanol that does not include any acids, and open the front door to ventilate the instrument.
- Avoid using a solution that contains an acid concentration higher than those listed above. Dilute the solution sufficiently before use.

⚠CAUTION

When using a buffer solution for mobile phase:

If a buffer solution is used for mobile phase, follow the instructions given below. Depending on the type of buffer solution, tubing may become clogged.

- During injection from an autosampler, rinse solution and mobile phase are mixed in the tubing that extends between the high-pressure valve and the low-pressure valve. Before use, be sure to confirm that no salt deposition will occur even if rinse solution and mobile phase are mixed.
- When sample is injected, the flow lines shown with solid lines in Fig. 3.3 are filled with rinse solution. Those shown with dashed lines are filled with mobile phase. The high-pressure valve rotates before measuring sample as shown in Fig. 3.4 and squeezes mobile phase, which is compressed under high pressure, from high-pressure valve port Nos. 4 and 5 as well as Nos. 1 and 6. Depending on the pumping pressure, rinse solution and mobile phase may be mixed in the ◯ in the figure, which may result in salt deposition.

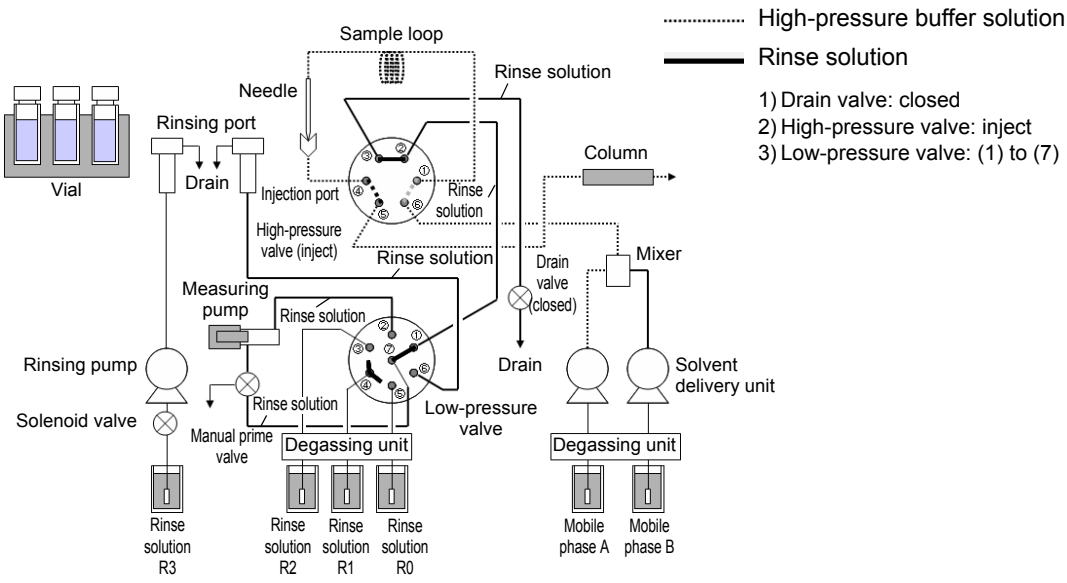


Fig. 3.3

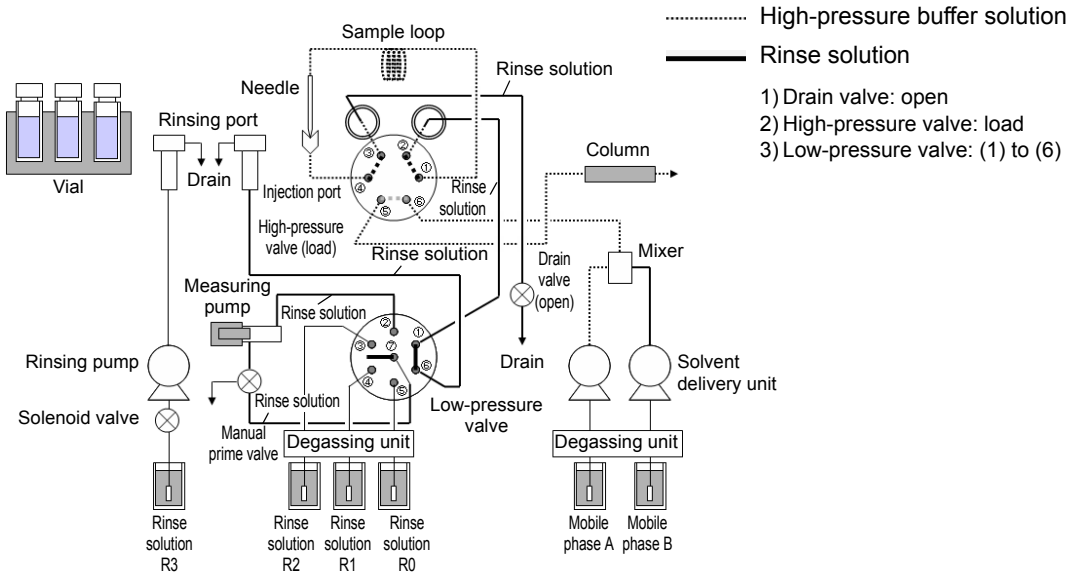


Fig. 3.4

- To prevent salt deposition, lower organic solvent concentration of rinse solution to 50 % or less when using a buffer solution exceeding a concentration of 50 mmol.

3.4 Purging Air Bubbles

Air bubbles are likely to appear in the tubing when the instrument has been inactive for a prolonged period or when the room temperature changes.
Air bubbles inside the flow lines will adversely affect sample injection precision. Be sure to use a degassing unit and for connection with the degassing unit, use stainless tubing attached to low-pressure valve port Nos. 3, 4 and 5.

☞ Nexera X2 SYSTEM GUIDE, "2.3.3 Plumbing for Low Pressure Flow Line"

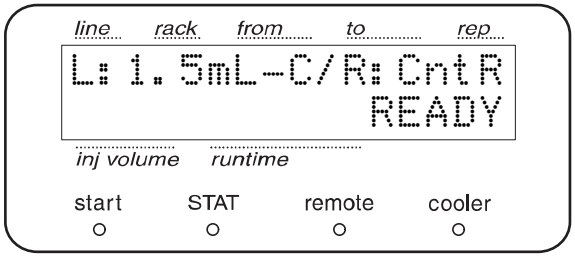
Before starting analysis, air bubbles must be purged with the PURGE function.
Also, perform the purge operation in the following cases:

- When the autosampler has not been used for a long period
- When the rinse solution has been changed
- When the room temperature has changed

NOTE

When replacing the solvent with an incompatible solvent, first replace with a compatible solvent as an intermediate rinse solution before replacing with the desired solvent.

1 Press **CE** to return to the initial screen.



NOTE

When 35 μL/s (default) is set for [RINSE SPEED], the average flow rate of the rinse solution dispensed by the measuring pump is approximately 1.0 mL/min.
When purging the flow line completely with a new rinse solution, perform manual priming in the rinse flow line and perform purge operation for 10 minutes for each line. To purge three kinds of rinse solution, set "2" (internal/external rinsing of the needle) at [RINSE TYPE].

☞ "[RINSE TYPE]" P. 80, "[MANUAL PRIME]" P. 103 in 5.

To change the purge time, see "[PURGE TM RP, PURGE TM ML, PURGE TM R0, PURGE TM R1, PURGE TM R2]" P. 83.

2 Use a manual syringe and draw three kinds of rinse solution into the flow line.

☞ "[MANUAL PRIME]" P. 103 in "5.2.5 Control Settings Group"

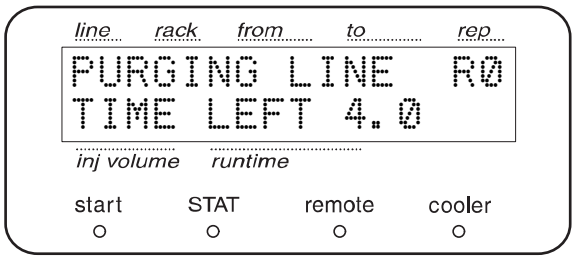
3 Set "2" at [RINSE TYPE] and set the purge time for three rinse flow lines.

"[PURGE TM RP, PURGE TM ML, PURGE TM R0, PURGE TM R1, PURGE TM R2]" P. 83 in "5.2.2 Parameter Settings Group"

NOTE

Without purging three rinse flow lines, sample injection accuracy could decrease.

4 Press purge .
Rinse solution will be applied to purge the flow lines.



5 Set the needle rinsing condition at [RINSE TYPE] for the subsequent analysis.

"[RINSE TYPE]" P. 80 in "5.2.2 Parameter Settings Group"

NOTE

If analysis starts with "2" at [RINSE TYPE], external/ internal rinsing of the needle will be set.

NOTE

To stop purging in mid-operation, press purge .
Purging will stop when the pump has discharged the rinse solution entirely, and if there is another rinse solution to purge, the next purge operation will start. To finish all purge operations, press purge repeatedly until no rinse solution to purge remains.

If the rinse solution flow line is connected to a degassing unit which has a large internal capacity. As such, the entire flow line may not be filled with rinse solution after a single purge operation. In this case, repeat the purge operation 2 or 3 times until rinse solution is discharged from the drain outlet.

3.5 Preparing Samples

⚠CAUTION

If insoluble substances, such as dust or solids, are mixed in the mobile phase or sample, they may cause clogging of the needle, needle seal, high-pressure valve stator or rotor, or instrument's flow line including the outlet tubing.
Besides the sliding surface on the high-pressure valve stator or rotor may be scratched, resulting in leaks within a very short time.
To prevent the entry of dust or solids, filter the mobile phase and sample through the membrane filter (0.22 μm) beforehand.
High-viscosity samples may not be injected correctly according to the volume setting. Dilute high-viscosity samples before use.

1 Completely dissolve the sample in a solvent with an identical or similar composition to the mobile phase.

2 Filter the sample through a membrane filter.

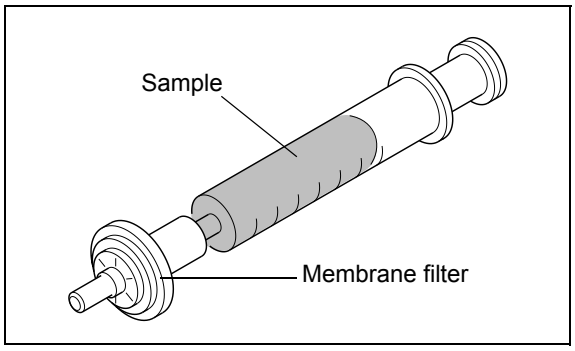


Fig. 3.5

3 Carefully load the sample(s) into a vial or microtiter plate well.

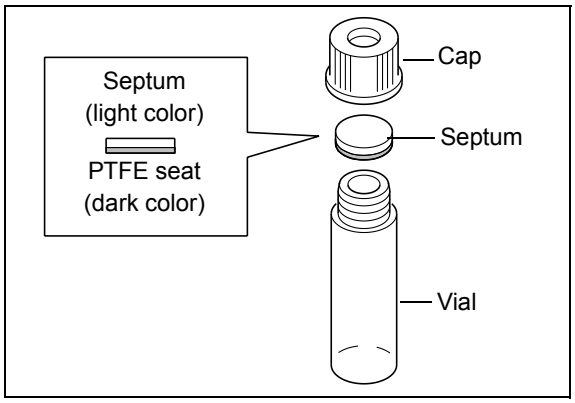


Fig. 3.6

⚠CAUTION

- When loading into a vial, place the silicon septum on the vial so that the PTFE surface is facing down (liquid side) before attaching the cap.
  If the septum is positioned incorrectly, the solvent may dissolve the silicon rubber.
- Only use vial septum provided by Shimadzu.
  Septum provided by other vendors may produce fragments that could clog the flow line or may prevent successful penetration by the needle.

4 Basic Operation

4.1 Preparing the Samples

⚠CAUTION

When a sample cooler is used, condensation may occur if the door is left open during temperature regulation. Also, when using the door sensor, the autosampler will not operate if the front door is open.

4.1.1 Setting Samples in the Sample Rack

There are several types of sample vial racks available -- 1 mL, 1.5 mL, 1.5 mL cooling (70 vials, 105 vials), 4 mL, microtiter plate, and deep-well MTP.

☞ "1.6 Optional Parts" P. 21

The upper surface of the rack is marked with numbers at the vial positions. Specify these numbers when setting sample number parameters.

⚠CAUTION

When inserting or removing the sample vial rack in or from the instrument, be sure to hold the sample vial rack with two hands.

1 Open the door.

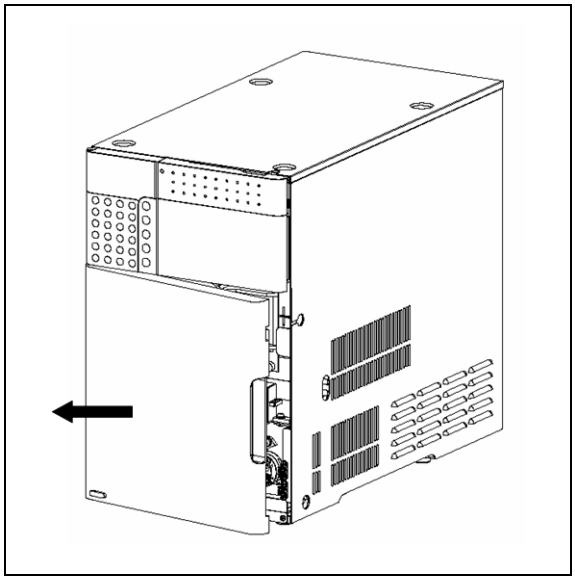


Fig. 4.1

2 Place the vial in the sample rack with the cap facing up.

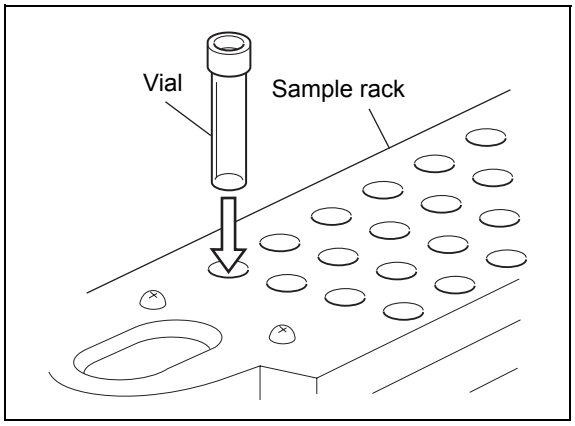


Fig. 4.2

3 Slide the sample rack along the guides all the way to the back of the instrument. The sample rack clicks into place when it is inserted correctly.

⚠CAUTION

If the sample rack is not inserted correctly, the needle may miss the vial opening and become bent.
When inserting or removing the sample vial rack in or from the instrument, be sure to hold the sample vial rack with two hands.

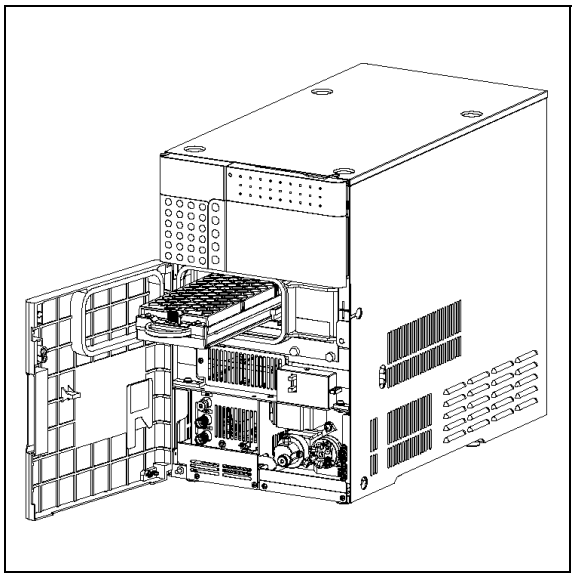


Fig. 4.3

4 Close the door.

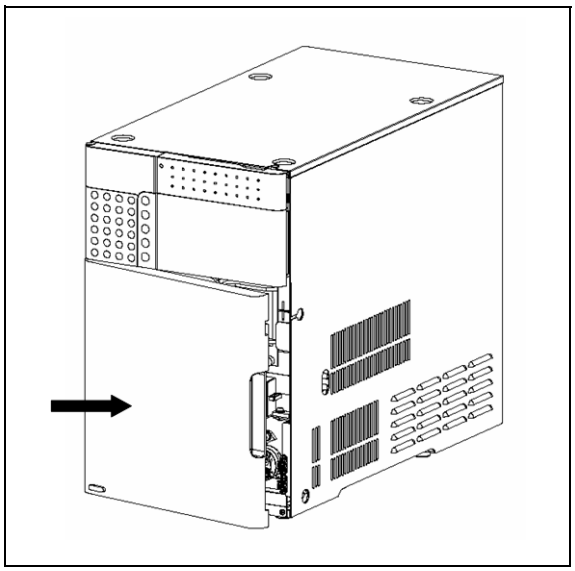


Fig. 4.4

4.1.2 Setting Samples in Microtiter Plates

When a microtiter plate is used for the first time or replaced with a different type of microtiter plate (96-well, 384-well, deep well), the sampling position intervals must be calibrated. This is referred to as the [teaching] procedure.

"[ADJUST MTP]" P. 137

1 Open the door.

2 Set the microtiter plate(s) in the MTP rack.
Set so that A1 well comes to left front.

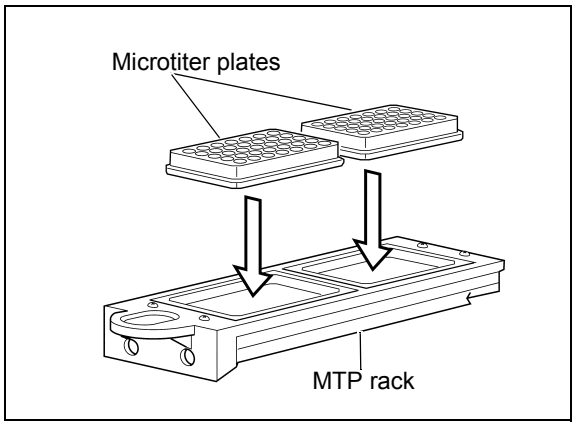


Fig. 4.5

NOTE

Up to two microtiter plates can be used.

3 Slide the MTP rack all the way to the back of the instrument. The MTP rack clicks into place when it is inserted correctly.

"Fig. 4.3" P. 47

⚠ CAUTION

When inserting or removing the sample vial rack in or from the instrument, be sure to hold the sample vial rack with two hands.

4 Close the door.

■ About the MTP Rack

The MTP rack position is as in the right figure.

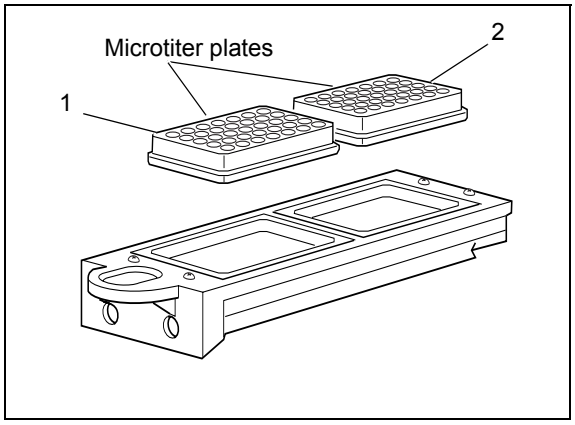


Fig. 4.6

4.1.3 Setting Samples Using a Sample Cooler

If the sample vials are cooled, the cooler LED illuminates.

■ Set the Sample Vial

1 Inject sample into the sample vial.

NOTE

The amount of sample introduced into the vial must not exceed the corresponding height shown on the right. If more than this amount is introduced, sufficient cooling of the liquid may not be possible.

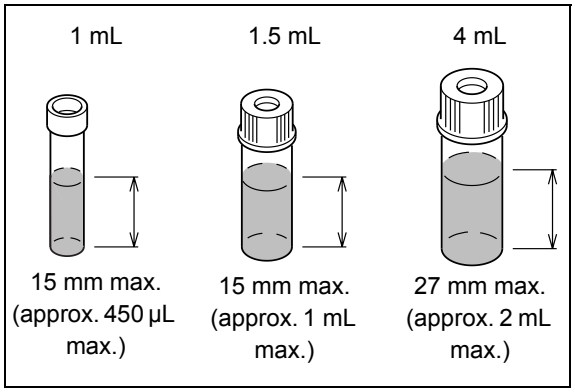


Fig. 4.7

2 Press the latch of front side of the cover to open the cover.

NOTE

Refer to the instruction manual that came with the rack for proper rack operation.
When using the 1 mL or 0.2 mL polypropylene sample vials, the sample vials may stick to the cover and be raised when the cover is opened. Remove the cover when using these sample vials. For details, refer to the sample vial rack supplementary information included in the package.

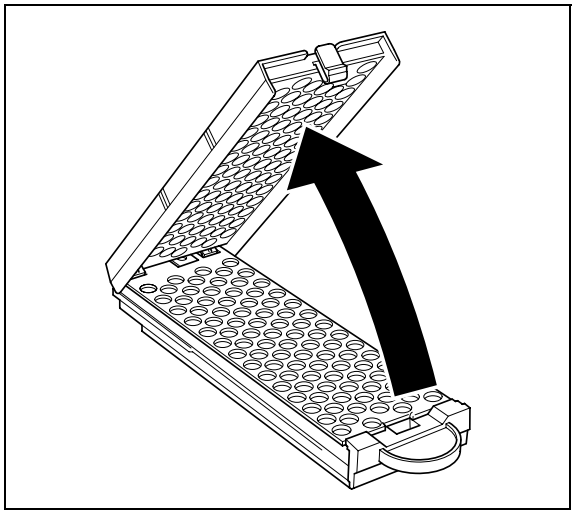


Fig. 4.8

⚠CAUTION

When inserting or removing the sample vial rack in or from the instrument, be sure to hold the sample vial rack with two hands.

3 Place the vial in the sample rack with the cap facing up.

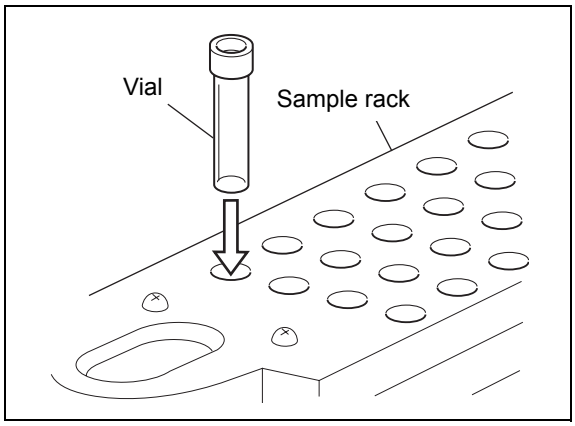


Fig. 4.9

4 Insert the hooks on rear side of the cover into square holes at the rear side on the sample rack.

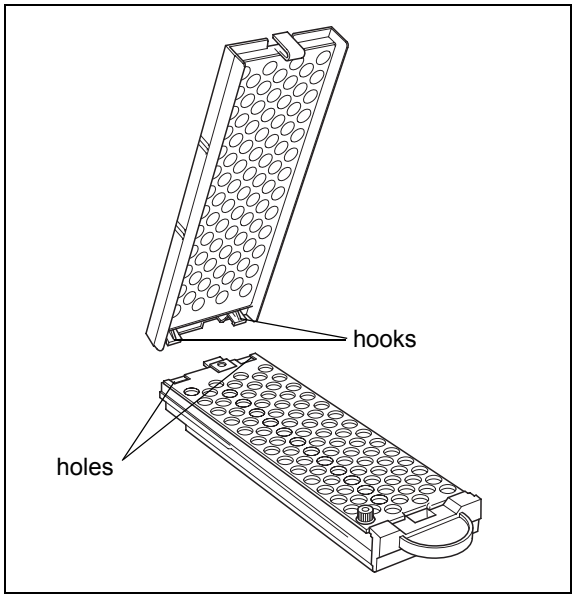


Fig. 4.10

5 The cover clicks if closed correctly.

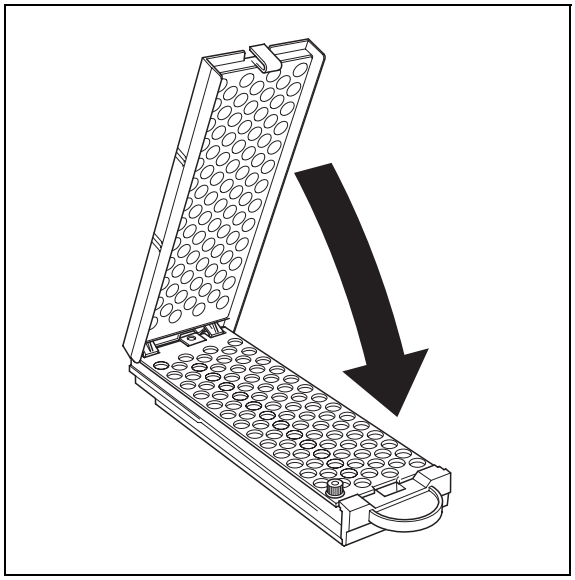


Fig. 4.11

4.2 Creating an Analysis Sequence Table

The procedure for creating a sample table is described below using an actual example.

The procedure for creating the following sample table is described using the settings for line 0. The line number is set automatically.

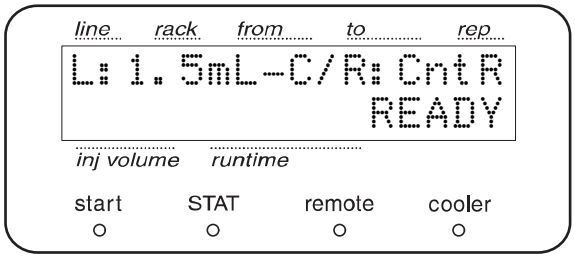
NOTE

The display cannot be used when CBM-20A/20Alite is connected.

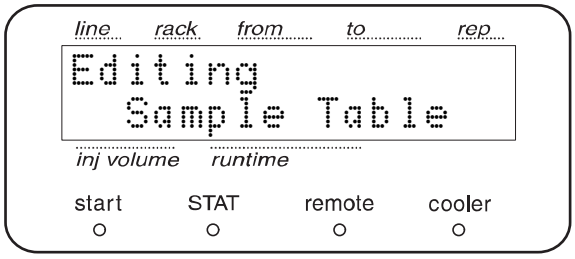
Sample table example

line	rack	from	to	rep	inj volume	runtime
0	1	1	9	10	20	5.00
1	1	20	30	10	10	10.00
2	1	30	39	1	10	30.00

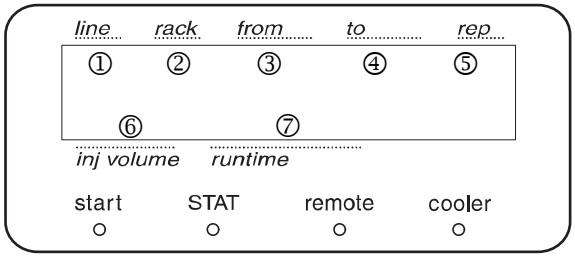
1 Press **CE** to return to the initial screen.



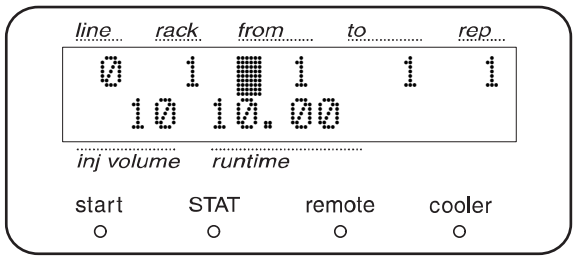
2 Press **edit**.
The analysis sequence table title screen will be briefly displayed, and then replaced by the parameter setting screen.



Explanation of parameter setting screen
① Line number
② Rack number
③ Initial sample vial number
④ Last sample vial number
⑤ Number of repeat injections
⑥ Injection volume (µL)
⑦ Analysis time (min)



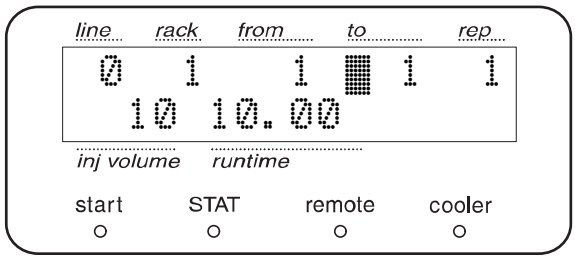
3 Use the numeric keypad to enter the rack number [1], and press **enter** .
The cursor will move to the [from] field.



NOTE

When using a rack changer, set rack numbers from 1 to 12.
Enter [0] to inject from the control vial rack.
"[MTP ORDER]" P. 89

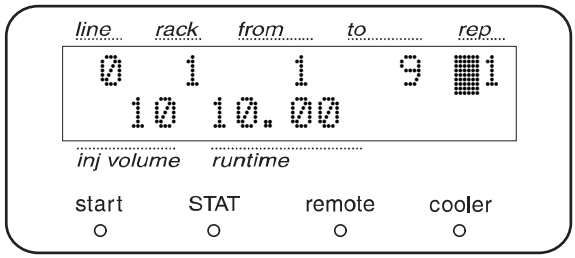
4 Use numeric keypad to enter the number [1] of the first vial to be injected, and press **enter** .
The cursor will move to the [to] field.
When using a microtiter or deep-well plate, enter the combination of an alphabetic character and number, such as "A01".



NOTE

Only the line number and rack number are displayed on a new parameter setting screen.

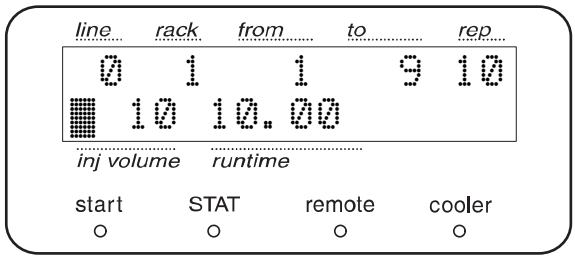
5 Enter [9], the number of the last vial to be injected, by numeric keypad and press **enter** .
The cursor will move to the [rep] field.



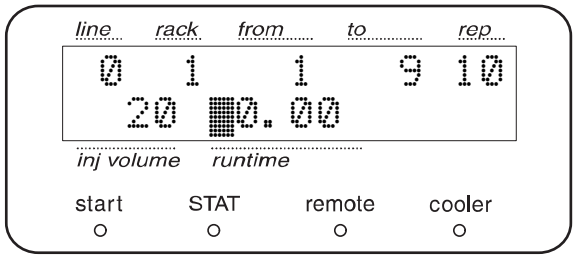
NOTE

If only one vial is to be injected, use the same number as in step 4.

6 Enter [10], the number of injections to be made repeatedly from one sample vial, by numeric keypad and press **enter** .
The cursor will move to the [inj. volume] field on the second line.



7 Enter [20], the injection volume (unit: µL), by numeric keypad and press **enter** .
The cursor will move to the [runtime] field.

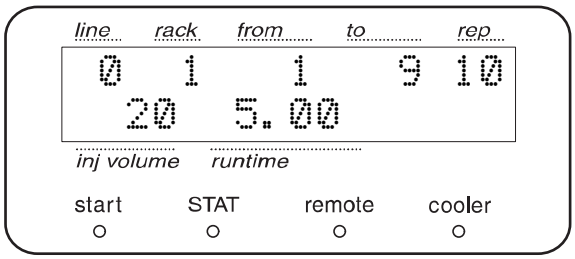


NOTE

Injection volumes from 0.1 to 9.9 µL can be entered in 0.1 µL increments, and from 10 to 50 µL can be entered in 1 µL increments.
When using the loop injection method, a loop injection base kit and a sample loop for loop injection are required optionally. Note that the maximum volume of injection is 20 µL.

"[MAX INJ. VOLUME]" P. 108, "1.6 Optional Parts" P. 21

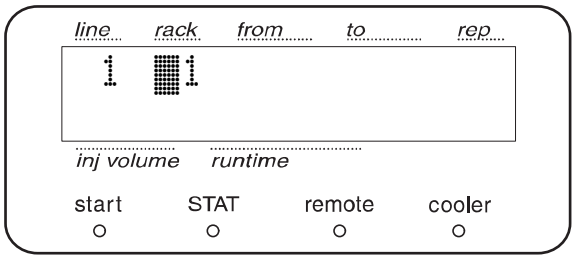
8 Enter [5.00], the analysis time (in minutes), and press **enter**.
The input values will be validated.



NOTE

Any value between 0.01 and 9999 minutes can be set. Values higher than 100 minutes must be in 1 minute increments.

9 Press **enter**.
The sample table on the next line appears.



10 Perform the required settings by repeating steps 1 to 9.

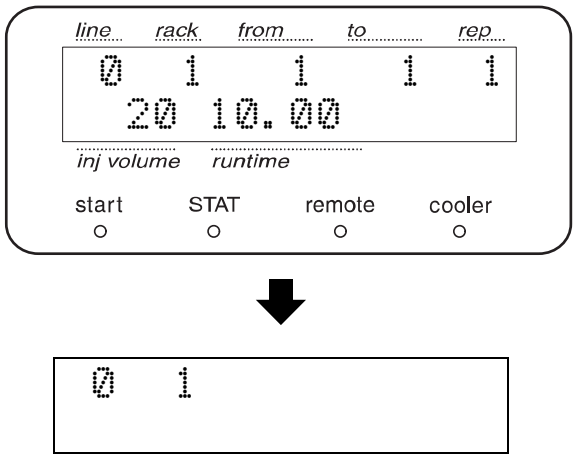
NOTE

- Up to 99 lines can be set in the sample table. If a repeat injection table is created using the procedure described in "5.4.1 Creating a Repeat Injection Table" P. 122, up to 99 lines including the number of insertions can be set.
- If no further lines are required, press **CE**.

11 Press **CE** to go back to the initial screen.

■ Deleting a Line

1 Display the line to be deleted, using the same procedure as when setting parameters. To display lines greater than 1, press (shift), (↓) and (↑) until the desired line is displayed.



2 Press (del).
The line displayed will be deleted, and the line that follows will be displayed. If no lines follow, the display will be as shown on the right.

■ Inserting a Line

1 Display the line number where the new line is to be inserted by pressing (shift), (↓) and (↑).

2 Press (ins).
The new line will be added to the table.

4.3 Selecting the Needle Rinsing Condition

The needle rinsing condition of the instrument is determined by the setting at [RINSE TYPE] in the parameter settings group.
The default set value is "1"; the following needle rinsing conditions are set.

- Only one kind of rinse solution (rinse solution R0) is used for external rinsing of the needle.
- Internal rinsing of the needle and rinsing of the injection port are not available.

"[RINSE TYPE]" P. 80

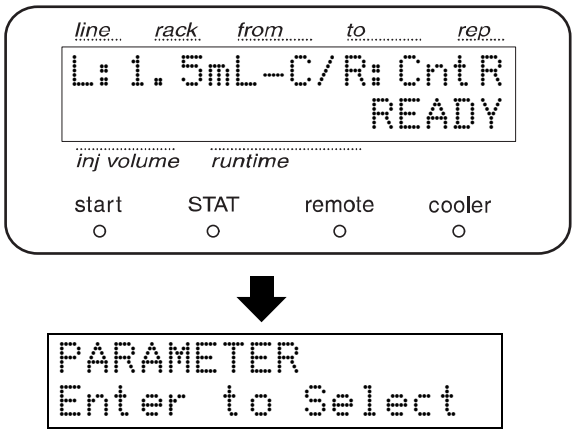
NOTE

- When the system controller CBM-20A/20Alite is connected, make the settings on the autosampler injection setting screen of the web controller screen. When the workstation LabSolutions is connected, make the settings on the system parameter screen.
- To control it in the 20A compatibility mode, make the settings using the numeric keypad on the instrument.

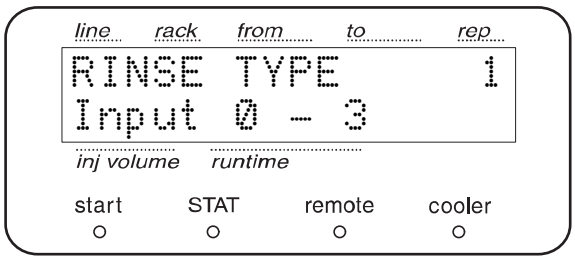
1 Press **CE**.
The initial screen will be displayed.

2 Press **func**.

* Pressing **back** moves back to the immediately previous item.



3 Select the needle rinsing method. Select the rinsing method using the numeric keypad and press **enter**.



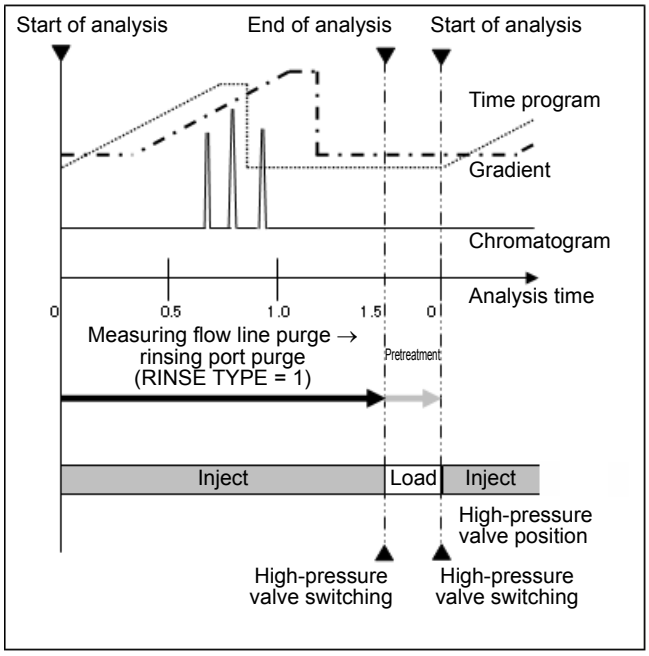
Set Value	Function
0	No rinsing of the needle
1	Perform external rinsing of the needle. (default)
2	Perform internal/external rinsing of the needle.
3	No rinsing of the needle (fast)

"[RINSE TYPE]" P. 80

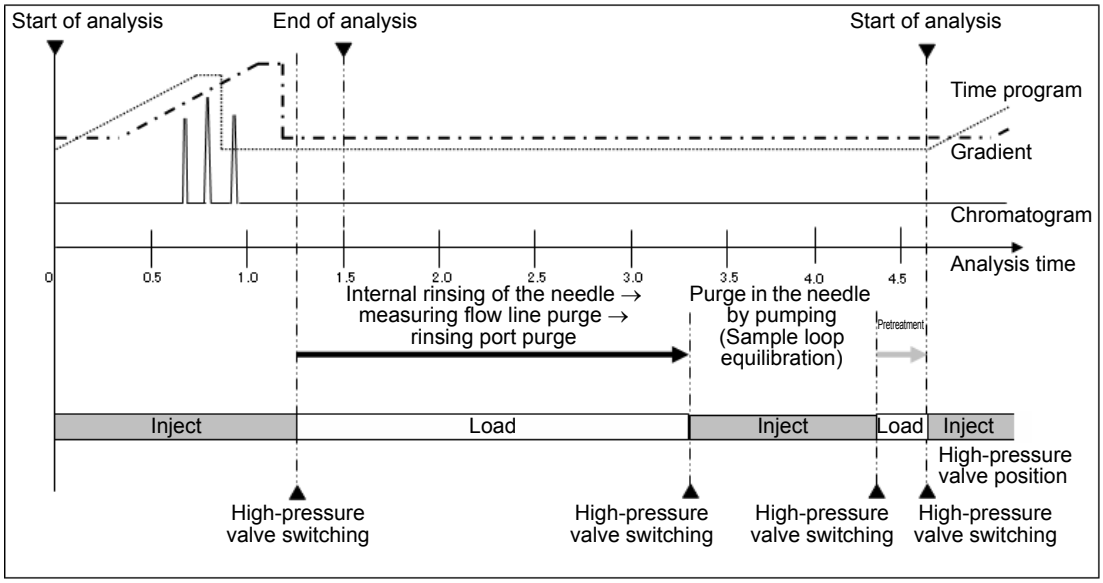
NOTE

The outlines of analysis sequences with respective needle rinsing methods are explained below.

1. When RINSE TYPE = 0, 1, 3:
   When "no rinsing", "external rinsing of the needle" or "no rinsing (fast)" is selected for the needle rinsing method, the measuring flow line and the rinsing port will be purged immediately after the start of analysis, and then the pretreatment process for the next analysis will start. External rinsing of the needle can be performed before and after sample aspiration during the pretreatment process.



2. When RINSE TYPE = 2:
   When internal/external rinsing of the needle is selected for the needling rinsing method, the high-pressure valve will be switched to the load position during or after analysis; internal rinsing of the needle will be performed; the measuring flow line and the rinsing port will be purged; the high-pressure valve will be switched to the injection position, the solvent in the sample loop and the needle will be purged with mobile phase, and then the pretreatment process for the next analysis will start. External rinsing of the needle can be performed before and after sample aspiration during the pretreatment process.



4.4 Starting Injection

1 Check that the front door is closed.

2 Press **start**.
The start indicator on the display panel illuminates, and the autosampler injects the first specified sample.

NOTE

When a data processor is used, it takes some time to print analysis results. To allow for this, set the autosampler's analysis time (RUN TIME) to a value slightly longer than that of the data processor's analysis time (STOP TIME). The value should allow time for the report to be printed. Note that starting the data processing unit by the event cable (provided as standard accessory) may fail, if analysis cycle of autosampler is shorter than that of the data processing unit.
For how to set the data processor's analysis time, refer to the data processor manual.

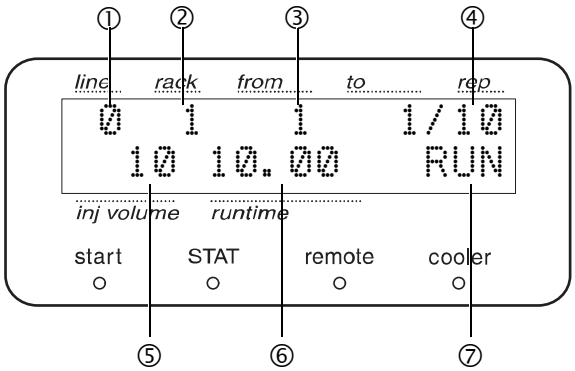
NOTE

If the open/close door detection is enabled and the door is opened during sample injection, operation ceases. Operation resumes as soon as the door is closed.

3 During analysis, the display screen indicates the status in the way shown on the right.

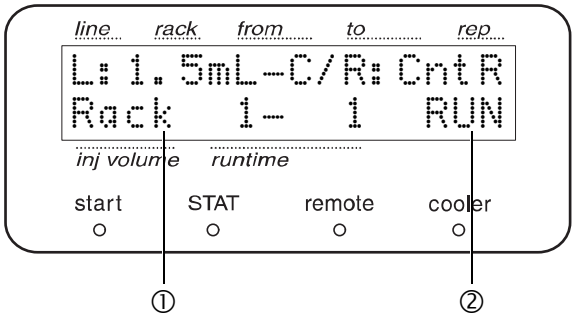
1) Controlled locally:

① Number of the line being executed
② Number of the rack being executed
③ Number of the sample vial being executed
④ Repeat status
⑤ Injection volume
⑥ Elapsed analysis time
⑦ Operating status



2) Controlled from a system controller:

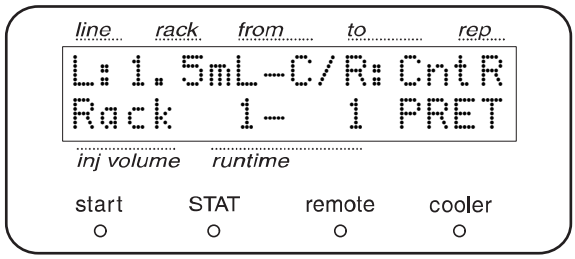
① When [20A] is set for [OP MODE]: [20A MODE]
☞ "[OP MODE]" P. 148
② Operating status and number of sample vial being analyzed



NOTE

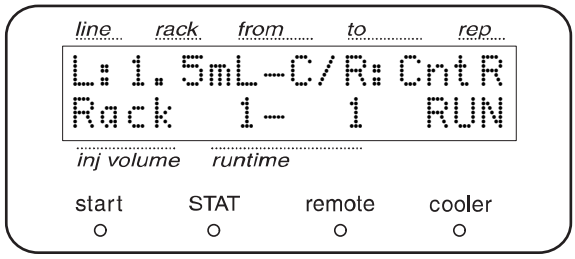
The following operating statuses are shown.

<1> [PRET]: Indicated during sample aspiration



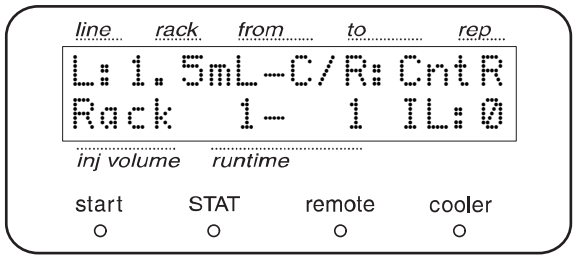
<2> [RUN] lit: indicated during analysis

[RUN] (flash): Indicates in the waiting for execution, such as the internal rinsing of the needle or equilibration of the sample loop, during analysis



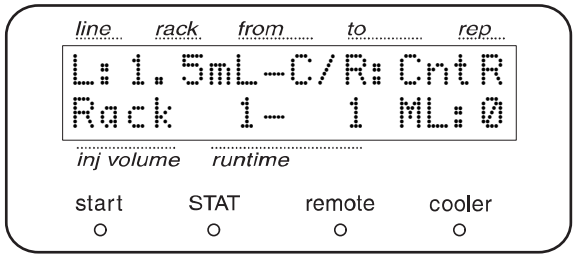
<3> [IL: 0]: Indicates during internal rinsing of the needle with rinse solution R0

- When it is executed during analysis, [RUN] and [IL: 0] are displayed alternately.
- When it is executed after analysis, [POST] and [IL: 0] are displayed alternately.
- In addition to [IL: 0], [IL: 1] (rinse solution 1) and [IL: 2](rinse solution 2) are displayed.



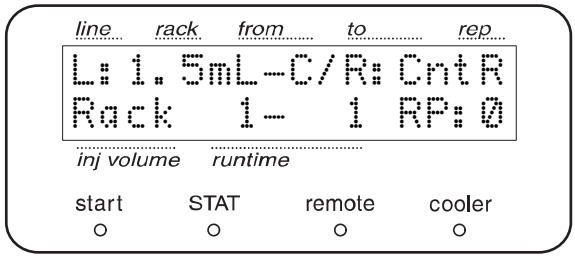
<4> [ML: 0]: Indicates during purge operation of the measuring flow line with rinse solution R0

- When it is executed during analysis, [RUN] and [ML: 0] are displayed alternately.
- When it is executed after analysis, [POST] and [ML: 0] are displayed alternately.
- In addition to [ML: 0], [ML: 1] (rinse solution 1) and [ML: 2](rinse solution 2) are displayed.

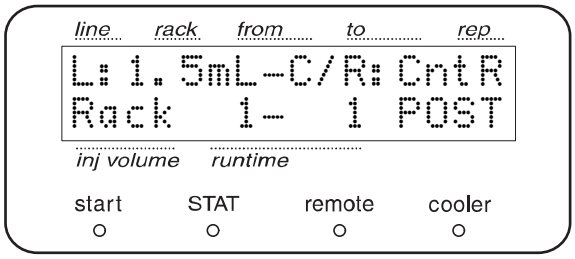


<5> [RP: 0]: Indicates during purge operation of the rinsing port with rinse solution R0

- When it is executed during analysis, [RUN] and [RP: 0] are displayed alternately.
- When it is executed after analysis, [POST] and [RP: 0] are displayed alternately.
- In addition to [RP: 0], [RP: 1] (rinse solution 1) and [RP: 2](rinse solution 2) are displayed.

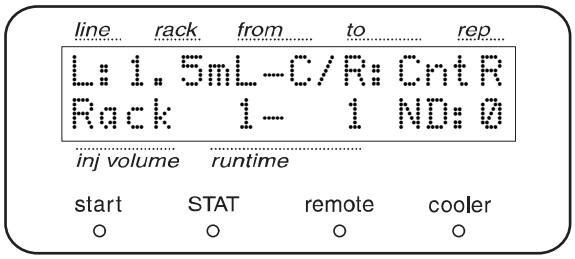


<6> [POST]: Flashes in the waiting for execution, such as equilibration of the sample loop, after analysis



<7> [ND: 0]: Indicates while dispensing rinse solution R0 from the needle tip in the pre-treatment process

- In addition to [ND: 0], [ND: 1] (rinse solution 1) and [ND: 2](rinse solution 2) are displayed.



4.5 Stopping Injection

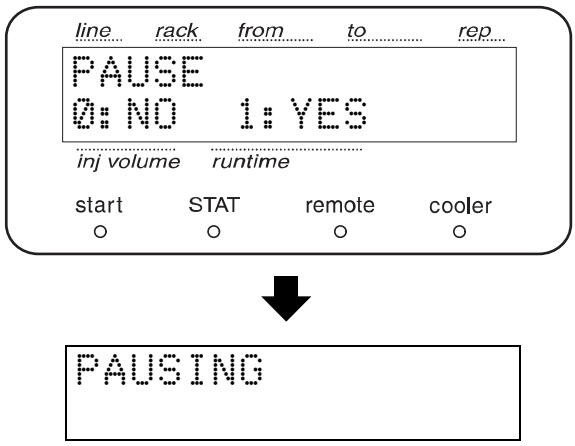
There are two ways to stop the injection process. It can either be stopped temporarily (i.e., paused) or stopped immediately.

■ To Pause the Injection Process Temporarily

1 After selecting the [CONTROL] screen, press **func** repeatedly until [PAUSE] is displayed.

2 Select [1:ON] to return to the initial screen.
(The [start] LED will flash.)
Operation will pause when analysis of the current sample ends.

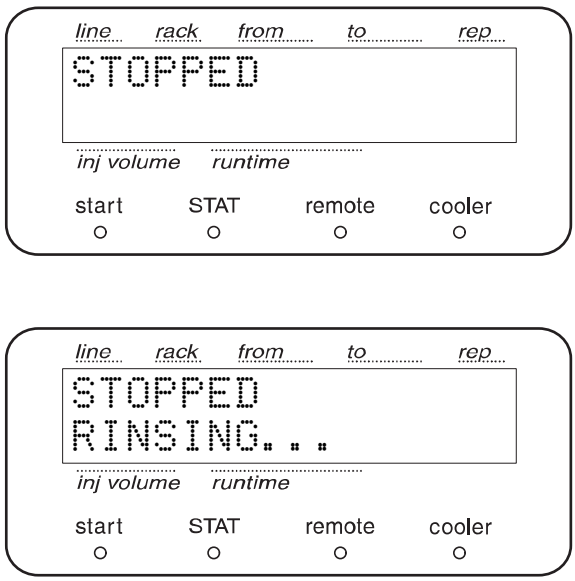
3 When ready to resume operation (starting from the next analysis) press **start** .



■ To Immediately Stop the Injection Process

1 Press **start** during analysis.
The message shown on the right appears on the display, informing the operator that operation has stopped.

2 When the operation has stopped, start rinsing of the needle and cancel the stopped state.

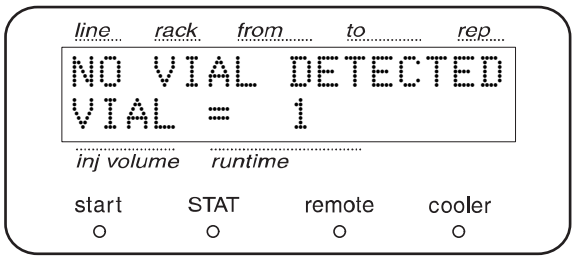


NOTE

If the aspirated sample remains in the needle, such as immediately stopping the injection process, the needle moves to the drain port and discharges the sample from the needle, and then rinsing of the needle starts.

■ Skipping Injection for Specific Sample Vials

If a sample vial registered in the sample table is not set in the sample rack, the injection operation for this vial is skipped and continued from the next vial. When a sample vial is skipped, its number is displayed in the way shown on the right. Press CE to clear this display. (When injection from a sample vial in a control vial rack is skipped, [c] is displayed in front of the vial number (e.g., [c1]).)



4.6 Finishing Injection

1 Rinse the flow lines.
"4.7 Rinsing" P. 63

2 Press the power switch to turn the power off.
"3.1 Turning Power ON/OFF" P. 38

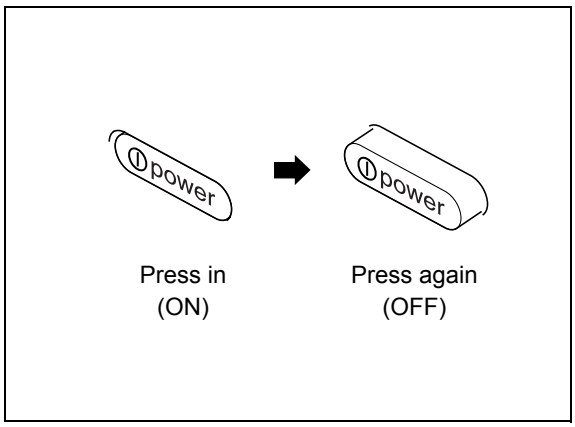


Fig. 4.12

4.7 Rinsing

To ensure long-term, safe, and trouble-free operation, follow these rinsing procedures when analysis have been completed.

⚠CAUTION

When a buffer solution has been used as a mobile phase or rinse solution, the flow lines must be cleaned with distilled or de-ionized water.
Otherwise, any remaining buffer solution evaporates and crystallizes over time. This residue could damage the instrument or clog the flow lines.

The mobile phase and sample flow lines must be cleaned separately. Follow the procedures below.

4.7.1 Rinsing the Mobile Phase Flow Lines

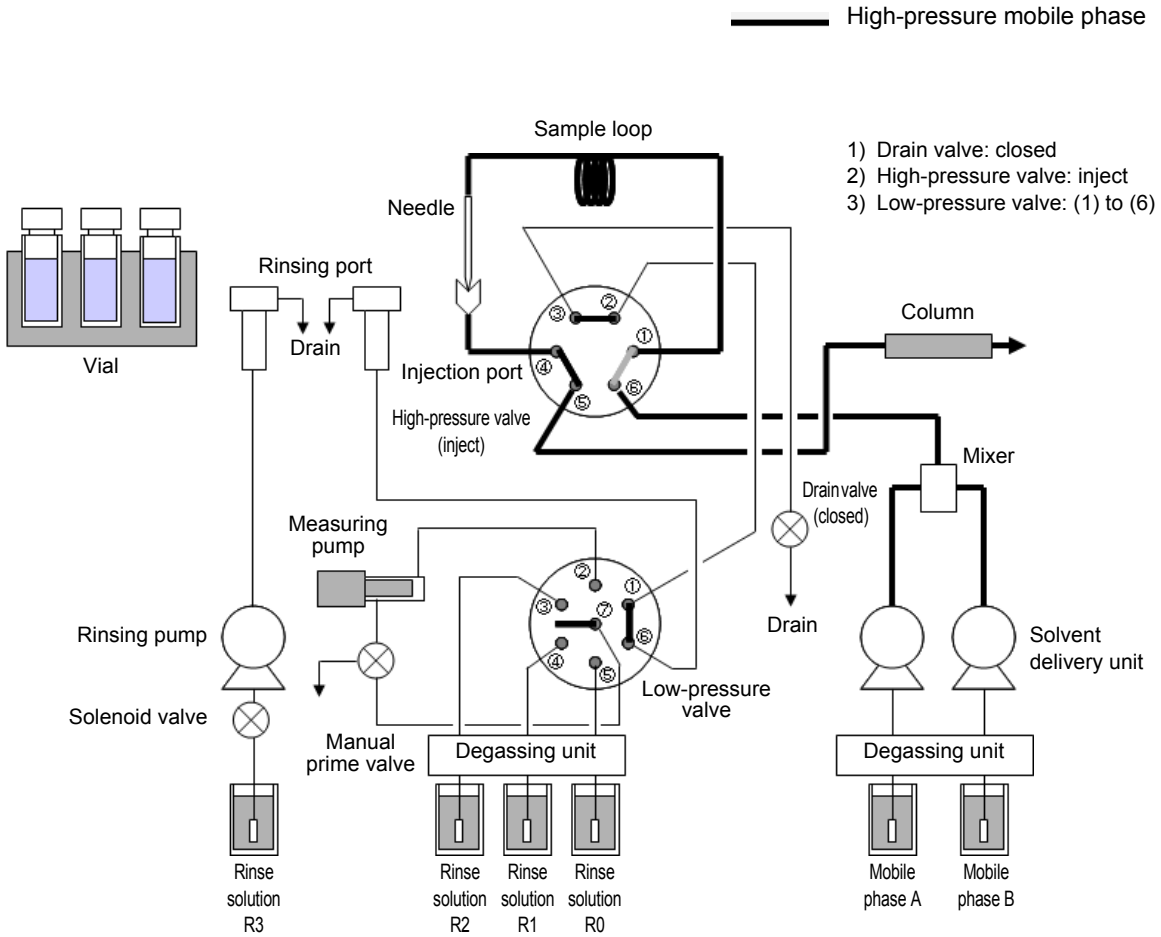


Fig. 4.13

1 Replace the mobile phase in the reservoir with distilled or de-ionized water.

2 Remove the column from the flow line, run the pump until the mobile phase in the flow lines shown in the figure above has been completely purged with distilled water or de-ionized water.

3 Stop the pump.

4 Replace the mobile phase (distilled water or de-ionized water) with methanol in the reservoir bottle.

5 Run the pump again, until the water in the mobile phase flow lines has been completely replaced with methanol.

6 Stop the pump.

4.7.2 Rinsing the Sample Flow Lines

1 Replace the water in the rinse solution container with distilled or de-ionized water.

2 Press (purge).
Rinse solution is delivered from the measuring pump.

NOTE

- To purge three kinds of rinse solution, set "2" (internal/external rinsing of the needle) at [RINSE TYPE].
"[RINSE TYPE]" P. 80.
- The default setting for the purge time is 10 minutes. Reset the purge time when the set value has been changed.
"[PURGE TM RP, PURGE TM ML, PURGE TM R0, PURGE TM R1, PURGE TM R2]" P. 83

3 Replace the mobile phase (distilled water or de-ionized water) with methanol in the reservoir bottle.

4 Perform manual priming in the flow line replaced with methanol, and purge the flow line for 10 minutes.

4.7.3 Cleaning the High-pressure Valve

0.1-mm ID tubing is likely to be clogged with wear debris, which are produced from the high-pressure valve. If the tube is clogged, follow the procedure below to solve this.

1 Disconnect the tubing (outlet tubing) from high-pressure valve port No. 5 and ensure that there is no clogging in the upstream flow line. For details on the procedure for removing or tightening UHPLC fittings at the outlet tubing, refer to the Nexera X2 SYSTEM GUIDE.

☞ Nexera X2 SYSTEM GUIDE, "2.3.4 Prior to Plumbing the High Pressure Flow Line"

⚠CAUTION

Before carrying out cleaning operation, ensure that the pump pressure of the mobile phase displayed on the screen of the solvent delivery pump is reduced to zero.

2 Clean the outlet tubing by reversing the flow and remove clogs. If the clogs cannot be removed, replace the tubing.

☞ "7.13.2 Reverse Rinsing of the Flow Lines" P. 269, "7.14 Replacing the Outlet Tubing" P. 271

3 Demount the high-pressure valve following the procedure of "7.6 Replacing and Inspecting High-Pressure Valve Rotor and Stator".
Clean the rotor and stator with an ultrasonic cleaning device.
(Perform this operation about every 10,000 injections.)
Replace the rotor with a new one after approximately 10,000 injections. Replace the stator with a new one after approximately 20,000 injections.

☞ "7.6 Replacing and Inspecting High-Pressure Valve Rotor and Stator" P. 247

4.7.4 Cleaning Rinsing Port and Rinsing Port Cover

Follow the procedure below if a leak occurs at the rinsing port slope or the rinsing port slope becomes dirty.

1 Check that the tip of the drain tubing is not immersed in the waste.
Use the drain tube clamp to prevent the immersion of the tip of the drain tubing.
Nexera X2 SYSTEM GUIDE, "2.3.1 Connecting Leakage Drain Tubing"

2 Remove the rinsing port cover.
"7.12.1 Before Replacing the Rinsing Port Cap" P. 266

3 Clean the rinsing port cover, rinsing port slope or other dirty portions using a soft cloth or paper soaked with water.

4 Replace the rinsing cover according to procedure 4. of "7.12.2 Replacing the Rinsing Port Cap" P. 266.

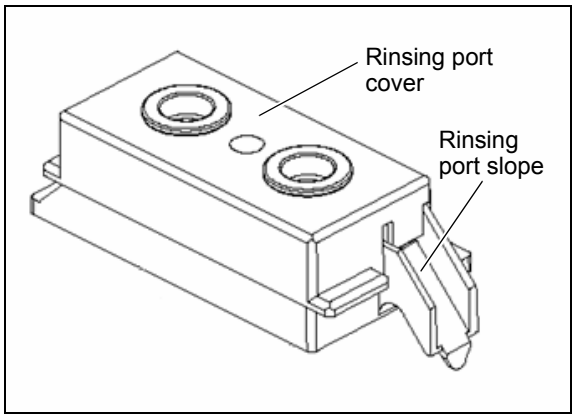


Fig. 4.14

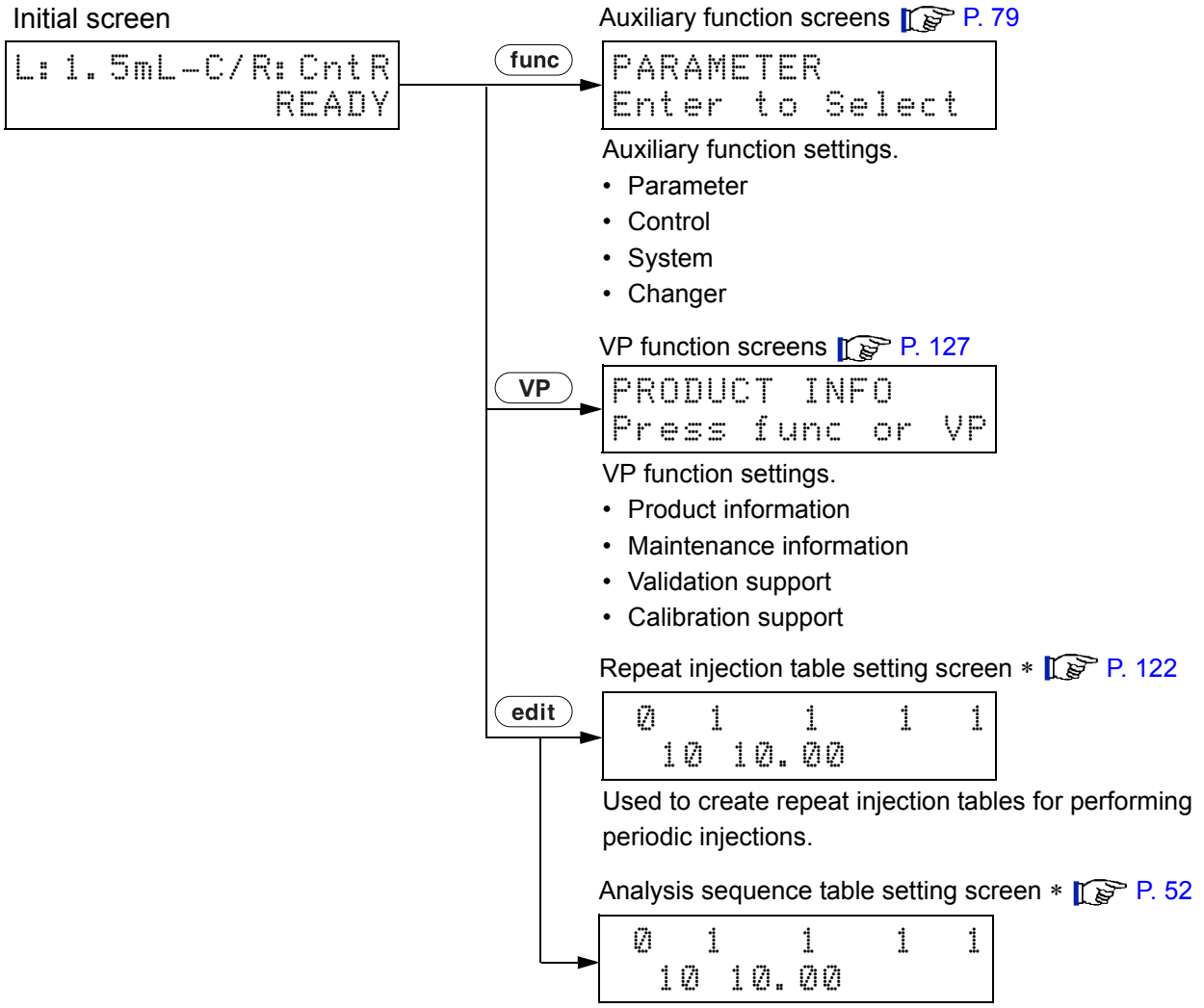
5 Application Operation

5.1 Display Panel

5.1.1 Types of Screens

Turn the power ON, the initial screen appears.
By pushing the keys (func), (VP) and (edit), the screen can be switched from the initial screen to one of the four screens described below.

- Auxiliary function screens
- VP function screen
- Repeat injection table setting screen or analysis sequence table setting screen



* The display cannot be used when CBM-20A/20Alite is connected.

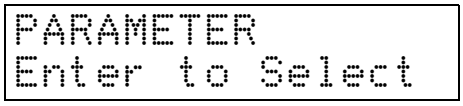
5.1.2 Auxiliary Settings Screens

In this section, auxiliary settings screens are shown in the following flow diagrams.
In each screen, press **func** to show the next screen, and press **back** to return.
In auxiliary settings group, press **enter** to enter each group.
Press **CE** to return the initial screen.

NOTE

To control it in the 20A compatibility mode, note the following points:
Blank: Some setup on the instrument is required because this is not downloaded from the CBM-20A/20Alite or workstation.
◎: The set values at the workstation (CBM if controlled by CBM) are downloaded to the instrument.
△: When "0" (REMOTE) is set, the set values at CBM are downloaded to the instrument.
When "1" (LOCAL) is set, the set values on the instrument are used.

■ Parameter Settings Group



●: displayed, blank: not displayed

20A Compatibility Mode	Name	Description	RINSE TYPE 0: No rinsing 1: External rinsing of the needle 2: External/internal rinsing of the needle 3: No rinsing (high-speed)				Remark	See
			0	1	2	3		
	RINSE TYPE	Select the needle rinsing method.	●	●	●	●	Default: 1 Set value: 0: No rinsing 1: External rinsing 2: External/internal rinsing 3: No rinsing (high-speed)	P. 80
◎	COOLER TEMP	Set the sample cooler temperature. The rack changer (if used) is set to the same temperature.	●	●	●	●	Default: 15 °C Set value: 4-40 °C	P. 82
◎	PURGE TM RP	Set the purge time of the rinsing port with rinse solution R0.	●	●	-	●	Default: 10.0 min Set value: 0: OFF, 0.1-25 min	P. 83
	PURGE TM ML	Set the purge time of the measuring flow line with rinse solution R0.	●	●	-	●	Default: 10.0 min Set value: 0: OFF, 0.1-25 min	P. 83

20A Compatibility Mode	Name	Description	RINSE TYPE 0: No rinsing 1: External rinsing of the needle 2: External/internal rinsing of the needle 3: No rinsing (high-speed)				Remark	See
			0	1	2	3		
◎	PURGE TM R0	Set the purge time of the measuring flow line and the rinsing port with rinse solution R0. (When [DIP-R SOL] = 0) Set the purge time of the rinsing port with rinse solution R0. (When [DIP-R SOL] = 1 or 2)	-	-	●	-	Default: 10.0 min Set value: 0: OFF, 0.1-25 min	P. 83
	PURGE TM R1	Set the purge time of the rinsing port with rinse solution R1.	-	-	●	-	Default: 10.0 min Set value: 0: OFF, 0.1-25 min	P. 83
	PURGE TM R2	Set the purge time of the rinsing port with rinse solution R2.	-	-	●	-	Default: 10.0 min Set value: 0: OFF, 0.1-25 min	P. 83
	ML PURGE VOL	Set the volume in the measuring flow line to be replaced.	●	●	●	-	Default: 100 µL (RINSE TYPE: 0/1) 600 µL (RINSE TYPE: 2) 100 µL fixed (RINSE TYPE: 3) Set value: 0-2000 µL	P. 83
◎	RINSE SPEED	Set the speed for replacing the rinse solution.	●	●	●	-	Default: 35 µL/s Set value: 1-35 µL/s	P. 84
◎	SAMPLE SPEED	Set the sample measuring speed.	●	●	●	-	Default: 5 µL/s Set value: 0.1-15.0 µL/s	P. 84
	DISP SPEED	Set the speed for dispensing the sample to the sample loop. * Displayed when loop injection method (INJECTION TYPE: 1) is selected	●	●	●	-	Default: 1.0 µL/s Set value: 0.1-15.0 µL/s	P. 85
◎	NEEDLE STROKE	Set the distance of moving the needle down.	●	●	●	●	Default: 51 mm	P. 85
△	MTP WELL	Set the well type when using a microtiter plate or a deep-well plate.	●	●	●	●	Default: 0 Set value: 0: REMOTE, 1: LOCAL	P. 89
	MTP ORDER	Set the sample injection order when using a microtiter plate or a deep-well plate.	●	●	●	●	Default: 0 Set value: 0: A1A2, 1: 1A1B	P. 89
	EVENT	Control the external output terminals.	●	●	●	●	Default: 0 Set value: 0-123	P. 91
	Repeat Inj Table	Set when performing bracket sequence analysis.	●	●	●	●	Default: 0 Set value: 0: OFF, 1: ON	P. 91

20A Compatibility Mode	Name	Description	RINSE TYPE 0: No rinsing 1: External rinsing of the needle 2: External/internal rinsing of the needle 3: No rinsing (high-speed)				Remark	See
			0	1	2	3		
	AIR GAP	Set the air volume to be trapped before and after the sample.	●	●	●	●	Default: 0.1 (INJECTION TYPE: 1) 0.0 (INJECTION TYPE: 0) Set value: 0: OFF, 0.1-5 μL	P. 92
	LOOP INJ TYPE	Select either partial loop or full loop method. * Displayed when loop injection method (INJECTION TYPE: 1) is selected	●	●	●	●	Default: 0 Set value: 0: PARTIAL, 1: FULL	P. 92
	EXCESS VOLUME	For setting the excess volume on aspiration. * Displayed when loop injection method (INJECTION TYPE: 1) is selected	●	●	●	●	Default: 10 Set value: 4-20 μL	P. 92
	LOOP FILL FACTOR	Set how many times of the loop volume the sample should be injected in the case of full loop method. * Displayed when loop injection method (INJECTION TYPE: 1) and full loop method (LOOP INJ TYPE: 1) are selected	●	●	●	●	Default: 3.0 Set value: 1.0-5.0	P. 93

NOTE

When the instrument is controlled by the system controller, set up the values other than [EVENT], [Repeat Inj Table] in the parameter settings group on the system controller (or workstation).

■ External Rinsing Parameter Settings Group

●: displayed, blank: not displayed

20A Compatibility Mode	Name	Description	RINSE TYPE 0: No rinsing 1: External rinsing of the needle 2: External/internal rinsing of the needle 3: No rinsing (high-speed)				Remark	See
			0	1	2	3		
◎	RINSE MODE	Select the needle rinsing method.	-	●	●	-	Default: fixed to 0 (RINSE TYPE: 0/3) 3 (RINSE TYPE: 1/2) Set value: 0-3	P. 93
	DIP-R SOL	Select the rinse solution type to be used in the rinsing port for external rinsing of the needle.	-	-	●	-	Default: 1 Set value: 0: R0, 1: R1, 2: R2	P. 94
◎	DIP-R TIME	Set the dip time of the needle in the rinsing port.	-	●	●	-	Default: 0 Set value: 0-60 sec	P. 94
◎	DIP-R VOL	Set the volume of rinse solution in the rinsing port to be replaced.	-	●	●	-	Default: fixed to 100 (RINSE TYPE: 0/3) 500 (RINSE TYPE: 1/2) Set value: 0-2000 μL	P. 94
◎	FLOW-R METHOD	Set the needle rinsing method when using a rinsing pump.	-	●	●	-	Default: 0 Set value: 0-6	P. 95
◎	FLOW-R TIME	Set the rinsing time with the rinsing pump.	-	●	●	-	Default: 2 sec Set value: 1-9 sec	P. 96

NOTE

These items are not effective when the instrument is controlled by the system controller. Set up the values on the system controller (or workstation).

■ Internal Rinsing Parameter Settings Group

```
INT.RINSE PARAM
Enter to Select
```

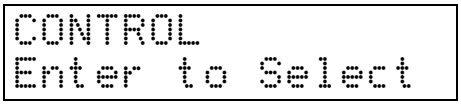
●: displayed, blank: not displayed

20A Compatibility Mode	Name	Description	RINSE TYPE 0: No rinsing 1: External rinsing of the needle 2: External/internal rinsing of the needle 3: No rinsing (high-speed)				Remark	See
			0	1	2	3		
	START TM	Set the time to start the internal rinsing of the needle. * Displayed when total injection method (INJECTION TYPE: 0) is selected	-	-	●	-	Default: -1.0 Set value: -1: AUTO, 0.00-999 min	P. 96
	SOL. SEQ	Set the rinse solution type and order for internal rinsing of the needle.	-	-	●	-	Default: 1 Set value: 0: R0, 1: R1, 2: R2	P. 97
	SOL. VOLUME	Set the volume of rinse solution to be used for internal rinsing of the needle.	-	-	●	-	Default: 300 Set value: 0-2000 μL	P. 98
	LOOP S.TM	Set the time to start equilibration of the sample loop. * Displayed when total injection method (INJECTION TYPE: 0) is selected	-	-	●	-	Default: -1.0 Set value: -1: LOAD, 0.00-999 min	P. 98
	LOOP HOLD TIME	Set the time to keep equilibration of the sample loop. * Displayed when total injection method (INJECTION TYPE: 0) is selected * Displayed when a time period is specified at [LOOP S.TM].	-	-	●	-	Default: 1.00 min Set value: 0.00-999 min	P. 99
	INJ.P RINSE	Set the rinsing of the injection port.	-	-	●	-	Default: R0: * R1: 0 R2: * Set value: 0, 1	P. 100

NOTE

These items are not effective when the instrument is controlled by the system controller. Set up the values on the system controller (or workstation).

■ Control Settings Group



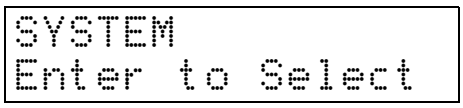
●: displayed, blank: not displayed

20A Compatibility Mode	Name	Description	RINSE TYPE 0: No rinsing 1: External rinsing of the needle 2: External/internal rinsing of the needle 3: No rinsing (high-speed)				Remark	See
			0	1	2	3		
	Clear SMPTBL	Delete the analysis sequence table.	●	●	●	●	Default: 0 Set value: 0: No, 1: Yes	P. 101
	Clear RepInjtbl	Delete the analysis sequence table used for bracket sequence analysis.	●	●	●	●	Default: 0 Set value: 0: No, 1: Yes	P. 101
	STAT	Set priority analysis.	●	●	●	●		P. 102
	PAUSE	Pause the sequence.	●	●	●	●	Default: 0 Set value: 0: No, 1: Yes	P. 103
	MANUAL PRIME	Draw the rinse solution using a manual syringe.	●	●	●	●	Default: 3 Set value: R0-R2, 3: OFF	P. 103
	Z HOME	Raise the needle for transportation.	●	●	●	●		P. 105
	TEST INJ PORT	Check that the needle is moved down correctly into the injection port.	●	●	●	●		P. 105
	PURGE (Ext Pump)	Purge the rinsing port using a rinsing pump and perform the external rinsing of the needle.	●	●	●	●		P. 105
	HPV TEST	Test the high-pressure valve switching operation.	●	●	●	●		P. 106
	I-RINSE EXE	Start internal and external rinsing of the needle.	-	-	●	-		P. 106

NOTE

[STAT] and [PAUSE] in the control settings group are not effective when the instrument is controlled by the system controller. Set up the values on the system controller (or workstation).

■ System Settings Group



●: displayed, blank: not displayed

20A Compatibility Mode	Name	Description	RINSE TYPE 0: No rinsing 1: External rinsing of the needle 2: External/internal rinsing of the needle 3: No rinsing (high-speed)				Remark	See
			0	1	2	3		
	LOCAL	Separate the autosampler from the external controller.	●	●	●	●	Default: 0 Set value: 0: Remote, 1: Local	P. 106
	KEY CLOSE	Lock the keypad.	●	●	●	●		P. 107
	BRIGHTNESS	Adjust the screen brightness.	●	●	●	●	Default: 4 Set value: 1-4	P. 107
◎	CNT RACK STRK	Set the distance of moving the needle down at the control vial rack.	●	●	●	●	Default: 52 mm Default: 17-54 mm	P. 107
	NEEDLE LOOP VOL	Set the maximum volume that can be aspirated at one time when the loop injection method is selected. * Displayed when loop injection method (INJECTION TYPE: 1) is selected	●	●	●	●	Default: 100 Set value: 10-2000 µL	P. 107
	MAX INJ. VOLUME	Set the maximum volume of injection. * Displayed when total injection method (INJECTION TYPE: 0) is selected	●	●	●	●	Default: 50 Set value: 10-2000 µL	P. 108
	VALVE LOOP VOL	Set the capacity of sample loop (option). * Displayed when loop injection method (INJECTION TYPE: 1) is selected	●	●	●	●	Default: 5.0 µL Set value: 1.0-20 µL	P. 108
	SELECT EVENT1	Switch event output 1 between event output and start output.	●	●	●	●	Default: 0 Set value: 0: Ev, 1: St	P. 109
	SELECT EVENT2	Switch event output 2 among event output, start output, and output at the end of analysis sequence table.	●	●	●	●	Default: 0 Set value: 0: Ev, 1: St, 2: Sd	P. 109
	SELECT EVENT3	Switch event output 3 between error output and event output.	●	●	●	●	Default: 0 Set value: 0: Error, 1: Event	P. 110
	USE SMALL VIAL	Set when using small-capacity vials.	●	●	●	-	Default: 0 Set value: 0: OFF, 1: ON	P. 110
	CBM LINK	Switch the remote connector between internal and external.	●	●	●	●	Default: 1 Set value: 0: Int, 1: Ext	P. 111
	BEEP MODE	Set the buzzer tone.	●	●	●	●	Default: 0 Set value: 0: On, 1: Alm, 2: Off	P. 111

■ Changer Settings Group

20A Compatibility Mode	Name	Description	Remark	See
	DISP RACK STATUS	Display the status of rack-changer racks 1 to 12.		P. 112
	STACK A CODE	Enter the stack code of rack-changer stack A.	Set value: 0: 96MTP, 1: 96DWP, 2: 1.5 mL, 3: 384MTP, 4: 384DWP	P. 112
	STACK B CODE	Enter the stack code of rack-changer stack B.		
	STACK C CODE	Enter the stack code of rack-changer stack C.		
	STACK D CODE	Enter the stack code of rack-changer stack D.		
◎	STACK A STRK	Set the needle stroke for stack A.	Default: 45 mm Set value (stack code): 10-52 mm (0),10-52 mm (1), 10-46 mm (2), 10-52 mm (3), 10-52 mm (4)	P. 114
◎	STACK B STRK	Set the needle stroke for stack B.		
◎	STACK C STRK	Set the needle stroke for stack C.		
◎	STACK D STRK	Set the needle stroke for stack D.		
	REMOVE RACK	Set when removing the rack-changer rack from the instrument.		P. 116
	SET DUMMY RACK	Set the condensation-prevention rack on the rack-changer rack.		P. 116
	AUTO EXCHANGE	Change the rack to the next one during sample analysis.	Default: 1 Set value: 0, 1	P. 117
	REMOVE DUMMY	Set whether or not to remove the rack-changer rack from the autosampler at the end of analysis sequence table.	Default: 0 Set value: 0, 1	P. 117
	RC STACK SCAN	Set whether or not to check the presence or absence of racks when inserting a stack.	Default: 0 Set value: 0, 1	P. 118
	Clear RACK INFO	Delete the rack presence/absence information as well as dummy rack position.		P. 119
	RC INITIALIZE	Check the presence or absence of racks at all the stacks.		P. 119
	LED LIGHT	Turn on the LED inside the Rack Changer II for about 10 seconds.	Enabled on the models with LED illumination	P. 119

NOTE

These items are displayed only when the Rack Changer II or Rack Changer/C is provided as an option.

5.1.3 VP Function Screens

In this section VP function screens are shown in the following flow diagrams.
Press VP on initial screen to show each group screen.
Press CE to return to the initial screen.
Press func or back to switch the settings screen within the groups selected by VP.
Press CE to return to the initial screen in the group.

■ Product Information Group

Name	Description	See
SERIAL NUMBER	Display the serial number.	P. 128
S/W ID	Display the program version number.	P. 128
RC SERIAL NUMBER	Display the rack changer's serial number.	P. 128
RC S/W ID	Display the rack changer's program version number.	P. 128

■ Maintenance Information Group

Name	Description	See
TOTAL OP TIME	Display the total operating time.	P. 129
NDL SEAL USED	Display the use frequency of the needle seal.	P. 129
HPV SEAL USED	Display the use frequency of the high-pressure valve rotor.	P. 129
HPV STATOR USED	Display the use frequency of the high-pressure valve stator.	P. 130
LPV SEAL USED	Display the use frequency of the low-pressure valve rotor.	P. 130
LPV STATOR USED	Display the use frequency of the low-pressure valve stator.	P. 130
EXT PUMP USED	Display the use frequency of the rinsing pump.	P. 130
NDLE FLUSH	Perform internal rinsing of the needle. * Displayed when total injection method (INJECTION TYPE: 0) is selected	P. 131
P-SET	Use when replacing the measuring pump plunger and the plunger seal.	P. 131
HPV ROTATION	Use after high-pressure valve rotor replacement.	P. 131
LPV ROTATION	Use after low-pressure valve rotor replacement.	P. 132
PART REPLACEMENT	Input logs of part replacement.	P. 132

Name	Description	See
MAINTENANCE LOG	Display maintenance logs.	P. 132
OPERATION LOG	Display operation logs.	P. 133
ERROR LOG	Display error logs.	P. 133

■ Validation Support Information Group

Name	Description	See
DATE	Display the date.	P. 134
TIME	Display the clock time.	P. 134
MEMORY CHECK	Run a memory check.	P. 135
POSITION SENS	Run a check with position sensors.	P. 135
LEAK SENSOR TEST	Run a check with leak sensors.	P. 136

■ Calibration Support Information Group

Name	Description	See
Input PASSWORD	Input the password. (Default: 00000)	P. 137
ADJUST MTP	Adjust the position of microtiter plates or deep-well plates.	P. 137
ERASE MTP ADJ	Delete microtiter plate position data.	P. 142
ASP FACTOR	Correct the injection volume accuracy.	P. 143
LEAK THR	Set the operation level of the leak sensor.	P. 145
NDL SEAL	Change the needle seal replacement alert value.	P. 145
HPV SEAL	Change the high-pressure valve rotor replacement alert value.	P. 145
HPV STATOR	Change the high-pressure valve stator replacement alert value.	P. 145
LPV SEAL	Change the low-pressure valve rotor replacement alert value.	P. 145
LPV STATOR	Change the low-pressure valve stator replacement alert value.	P. 146
EXT PUMP	Change the rinsing pump replacement alert value.	P. 146
CANCEL DOORSW	Set automatic door open/close detection.	P. 146

Name	Description	See
CANCEL RACKDET	Set automatic rack detection.	P. 147
CANCEL VIALDET	Set automatic vial detection.	P. 148
OP MODE	Set the mode for communications with the external controller.	P. 148
INITIALIZE PARAM	Initialize parameters.	P. 149
CHANGE PASSWORD	Change the password.	P. 149
ADJUST RACK	Adjust the rack position.	P. 150
ADJUST INJ PORT	Adjust the injection port position.	P. 151
ERASE RACK.P ADJ	Delete rack position data.	P. 154
ERASE INJ.P ADJ	Delete injection port position data.	P. 154
TEMP DELTA	Correct the sample cooler temperature.	P. 154
INJECTION TYPE	Change the injection method.	P. 154
CBM PARAMETER	Set the CBM parameters.	P. 155

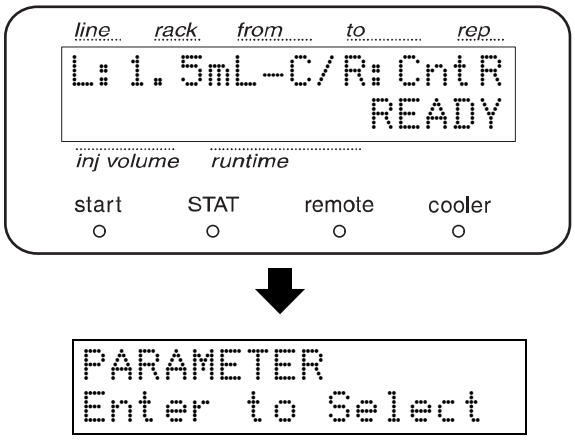
5.2 Parameter in Auxiliary Functions

There are six groups for auxiliary functions: parameter settings, external/internal rinsing parameter settings, control settings, system settings, and changer settings.

5.2.1 Showing the Auxiliary Function Screens

1 Press **CE**.
Initial screen appears.

2 Press **func**.
* Press **back** to return the previous screen.



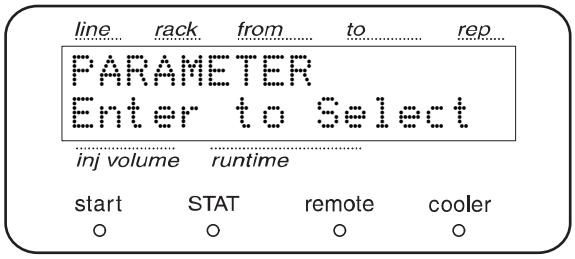
3 After selecting the desired parameter, follow the direction of each parameter described in the next section.

4 To make more setting, press **func** or **back** repeatedly to select the desired parameter.

5 Press **CE** to return to the initial screen.

5.2.2 Parameter Settings Group

This group is for setting and selecting the autosampler's parameters.



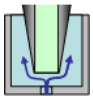
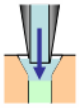
■ [RINSE TYPE]

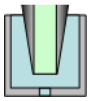
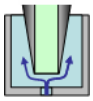
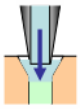
The needle rinsing condition of the instrument is determined by the setting at [RINSE TYPE] in the parameter settings group.
The default set value is "1"; the following needle rinsing conditions are set.

- Only one kind of rinse solution (rinse solution R0) is used for external rinsing of the needle.
- Internal rinsing of the needle and rinsing of the injection port are not available.

With the default setting ([RINSE TYPE] = 1), the rinse solution filled in the rinsing port is fixed to rinse solution "R0".
Rinse solutions "R1" and "R2" are not used for sample aspiration and analysis; however, without filling the flow line with these solutions, air bubbles may be trapped in the sample measuring flow line, resulting in decreasing the measurement accuracy. Accordingly, perform manual priming for rinse solutions "R1" and "R2", set "2" at [RINSE TYPE] and purge three rinse solutions for 10 minutes, respectively, to fill the same solvent as rinse solution "R0" in these flow lines.
The instrument, by adding rinse solutions "R1" and "R2", can use up to three kinds of rinse solutions (except for the rinsing pump) to rinse the external and internal surfaces of the needle. In this case, set "2" at [RINSE TYPE].
The rinse solution used for sample aspiration is fixed to rinse solution "R0" regardless of the set value at [RINSE TYPE].

○: usable, ×: not usable

[RINSE TYPE] Set Value	Rinsing				Purge after sample injection and rinsing			
	External rinsing		Internal rinsing		Measuring flow line purge		Rinsing port purge	
	Dip rinsing 	Pump rinsing 	Internal rinsing of the needle 	Injection port rinsing 	Solution	Purge volume	Solution	Purge volume
0	×	×	×	×	1 solution (R0 only)	100 μL adjustable (0-2000 μL)	× (No purge)	0 μL fixed
1 (default)	○ (1 solution)	○ (1 solution)	×	×	1 solution (R0 only)	100 μL adjustable (0-2000 μL)	○ (R0 only)	500 μL adjustable (0-2000 μL)
2	○ *1) (1 solution)	○ (1 solution)	○ (maximum 3 solutions)	○ (maximum 3 solutions)	1 solution (R0 only)	600 μL adjustable (0-2000 μL)	○ *1 (1 solution)	500 μL adjustable (0-2000 μL)

[RINSE TYPE] Set Value	Rinsing				Purge after sample injection and rinsing			
	External rinsing		Internal rinsing		Measuring flow line purge		Rinsing port purge	
	Dip rinsing 	Pump rinsing 	Internal rinsing of the needle 	Injection port rinsing 	Solution	Purge volume	Solution	Purge volume
3	×	×	×	×	1 solution (R0 only)	100 µL fixed	× (No purge)	0 µL fixed

*1 Select 1 solution from 3 solutions.

*) When performing external / internal rinsing of the needle or rinsing of the injection port, you need to set other items as well.

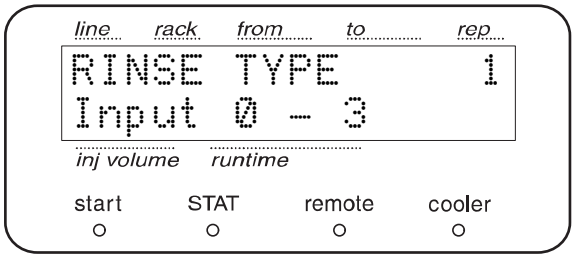
"5.2.3 External Rinsing Parameter Settings Group" P. 93, "5.2.4 Internal Rinsing Parameter Settings Group" P. 96

*) Rinse solution "R0" (mobile phase) must be included in "(maximum 3 solutions)" shown in the list above.

*) Rinse solution "R0" must be used as mobile phase (initial concentration for gradient analysis).

Select the needle rinsing method. Select the rinsing method using the numeric keypad and press **enter**.

Set Value	Function
0	No rinsing of the needle
1	Perform external rinsing of the needle. (default)
2	Perform internal/external rinsing of the needle.
3	No rinsing of the needle (high-speed)



NOTE

- When "0", "1" or "3" is set, rinse solutions R1 and R2 are not used. However, perform manual priming in the low-pressure flow line of rinse solutions "R1" and "R2" in advance to fill the line with the same solvent as rinse solution "R0", set "2" at [RINSE TYPE], and purge three rinse solutions for 10 minutes or longer, respectively. If not filled, air may be trapped in the low-pressure flow line, which may decrease analysis accuracy.
- When "2" is set, use rinse solution R0 as the mobile phase (initial concentration in the case of gradient analysis). After internal rinsing of the needle, the volume set at [ML PURGE VOL] is purged with rinse solution R0 (mobile phase) in the needle and the sample loop.

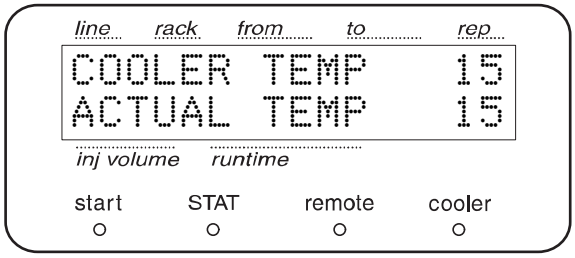
NOTE

- When "3" is set, internal and external rinsing of the needle is not performed as well as the following parameters are automatically fixed to the minimum required level to shorten the analysis cycle. Note that these parameters are not displayed on the screen of the instrument.

Parameter	Value at [RINSE TYPE: 3] (Fixed)	Remark
ML PURGE VOL	100 μL	Purge volume setting for measuring flow line
RINSE SPEED	35 μL/s	Speed setting for purging a rinse solution
SAMPLE SPEED	5 μL/s	Speed setting for measuring the sample
DIP SPEED	1.0 μL/s	Speed setting for filling the sample in the sample loop * For loop injection method only
DIP-R VOL	100 μL	Purge volume setting of the rinse solution in the rinsing port
USE SMALL VIAL	0: OFF (Not usable)	Double-motion setting when using small-capacity vials

■ [COOLER TEMP]

Sets the temperature when using a sample cooler. The current cooler temperature is displayed on the second line.



NOTE

The rack changer (if used) is also set to the same temperature.

Enter the temperature by the numeric keypad, and press (enter). By entering [-1], operation of the sample cooler is stopped.

Set Range	Default value
4-40 °C	15 °C

■ [PURGE TM RP, PURGE TM ML, PURGE TM R0, PURGE TM R1, PURGE TM R2]

Set the time to purge the flow line.

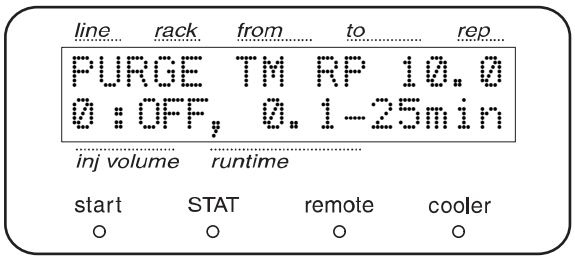
NOTE

The menu indication changes according to the setting at [RINSE TYPE].

Purge Menu	OPERATION	Menu Indication	
		RINSE TYPE: 0, 1, 3	RINSE TYPE: 2
PURGE TM RP	Purge the rinsing port with rinse solution R0.	Displayed	Not displayed
PURGE TM ML	Purge the measuring flow line with rinse solution R0.		
PURGE TM R0	Purge the measuring flow line and the rinsing port with rinse solution R0. (When [DIP-R SOL] = 0)	Not displayed	Displayed
	Purge the rinsing port with rinse solution R0. (When [DIP-R SOL] = 1 or 2)		
PURGE TM R1	Purge the rinsing port with rinse solution R1.		
PURGE TM R2	Purge the rinsing port with rinse solution R2.		

Enter the purge time using the numeric keypad, and press **enter**.

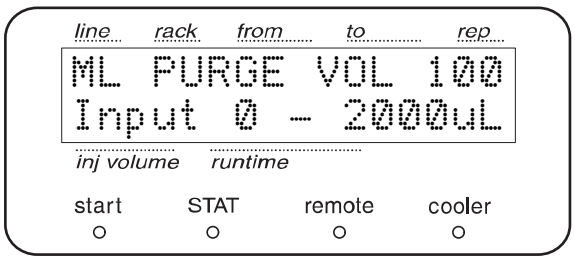
Set range	Default value
0: OFF 0.1 to 25 min.	10.0 min.



■ [ML PURGE VOL]

Set the volume of the solution to be replaced in the measuring flow line and in the internal rinsing flow line of the needle. Enter the purge volume using the numeric keypad, and press **enter**.

Set range	Default value
0 to 2000 μL	100 μL

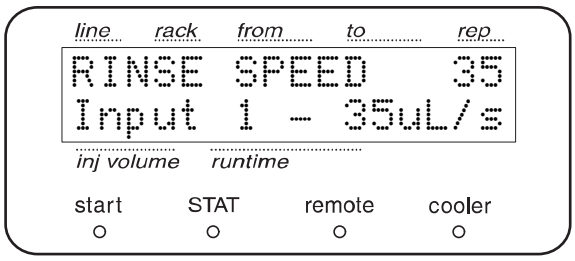


NOTE

As shown in "5.8.2 Example 2: When Performing Internal Rinsing of the Needle After Analysis (without Sample Loop Equilibration)" P. 168, if the high-pressure valve is set to the INJECT position using [LOOP S. TM] and [LOOP HOLD TIME] after internal rinsing of the needle without equilibration of the sample loop by pumping the mobile phase, it is recommended that a value greater than 600 µL be set for the purge volume to replace the rinse solution in the flow line with the mobile phase.

■ [RINSE SPEED]

Sets the discharge rate of the rinse solution during rinsing in µL/sec.



NOTE

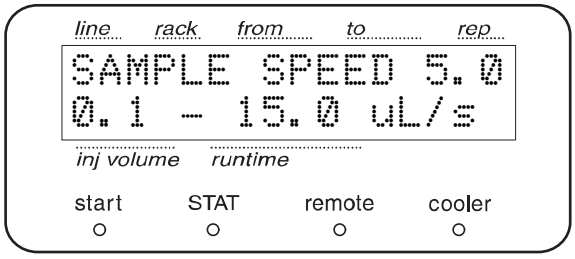
Set a small value when using a rinse solution with a high viscosity that is likely to cause bubbles during pumping.

Enter the value with numeric keypad and **enter**.

Set Range	Default value
1-35 µL/sec	35 µL/sec

■ [SAMPLE SPEED]

Sets the rate at which the sample is aspirated by the needle in µL/sec.
Enter the value with numeric keypad and **enter**.



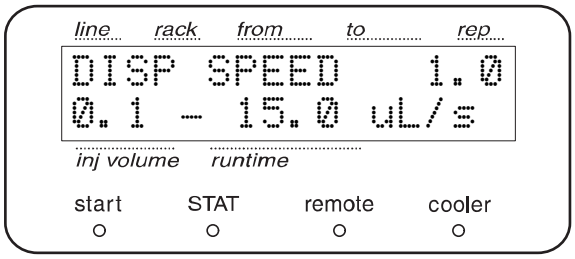
Set Range	Default value
0.1-15.0 µL/sec (in increments of 0.1)	5.0 µL/sec

NOTE

5.0 µL/sec is set for the default value as a general guide. For a viscous sample, set a smaller value.

■ [DISP SPEED]

Set the speed in the units of μL/sec for dispensing the sample from the needle to the sample loop.
Enter the aspiration speed using the numeric keypad and press (**enter**).



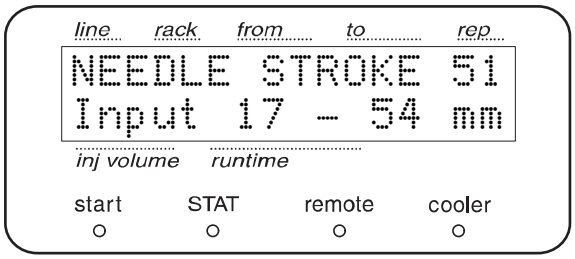
Set range	Default value
0.1 - 15.0 μL/sec (in increments of 0.1)	1.0 μL/sec

NOTE

- 1.0 μL/sec is set for the default value as generally.
- When using a viscous sample or sample loop having a small I.D., set a smaller value.

■ [NEEDLE STROKE]

Sets the distance that the needle descends into the sample vial. When the rack is changed, this parameter is automatically reset to the default value (the needle goes to a depth of 2 mm from the bottom of the sample vial). This parameter needs to be adjusted for each of the different rack types.
Enter the value with numeric keypad and (**enter**).
The setting ranges for different types of rack are shown in the following table.



Rack type	Set Range	Default value (Increment : mm)
Sample vial rack for 1 mL vials	17-54	51
Sample vial rack for 1.5 mL vials	17-54	52
1.5 mL sample vial cooling rack	17-54	52
Sample vial rack for 4 mL vials	17-54	51
Microtiter plate rack	10-52	45
Deep-well plate rack	10-52	40

NOTE

For rack-changer racks, set the needle stroke in the changer settings group.

⚠CAUTION

When using one of the following vials or another thin-tipped small-capacity vial, set the needle stroke to 50 mm or less.
If a value greater than 50 mm is set for these vials, the tip of the needle may touch the vial bottom since it is higher than other glass vials. This may result in varied aspiration amounts or damage to the needle tip.

Part Name	Part No.
1.1 mL vial	S228-21283-91
1 mL vial	S228-31600-91
0.3 mL vial	S228-16847-92
0.2 mL vial	S228-35217-91

☞ "Sample Vial Racks, Microtiter Plate Racks" P. 23

NOTE

The needle stroke must be set so that there will be at least 1 mm left between the needle tip and the bottom of the vial or well. If there is no space between the needle tip and the bottom of the vial or well, the needle tip will contact the vial or well and may fail in sample aspiration, resulting in false analysis results.
In addition, even if the same kinds of microtiter plates or deep-well plates are used with the same needle stroke setting, the distance between the needle tip and the well bottom may vary 3 mm maximum due to the rack structure depending on whether the autosampler is used independently or is used in combination with a rack changer.

[When the autosampler is used independently (microtiter plate rack or deep-well plate rack)]
When the well bottom thickness at the microtiter plate is 1 mm and the needle stroke setting is 45 mm (default), the needle tip will move down to the depth 6 mm from the well bottom surface (as shown in Fig. 5.1).
When the default value is used for needle stroke setting:

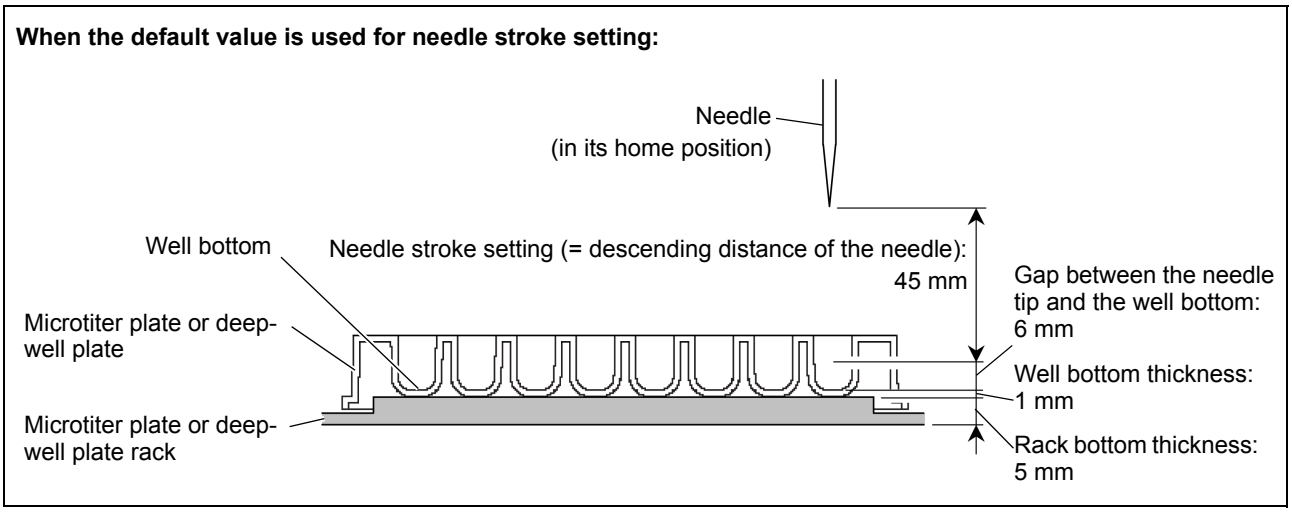


Fig. 5.1

[When the autosampler is used with a rack changer (rack-changer rack)]
When the well bottom thickness at the microtiter plate is 1 mm and the needle stroke setting is 45 mm (default), the needle tip will move down to the depth 3 mm from the well bottom surface (as shown in Fig. 5.2).
When the default value is used for needle stroke setting:

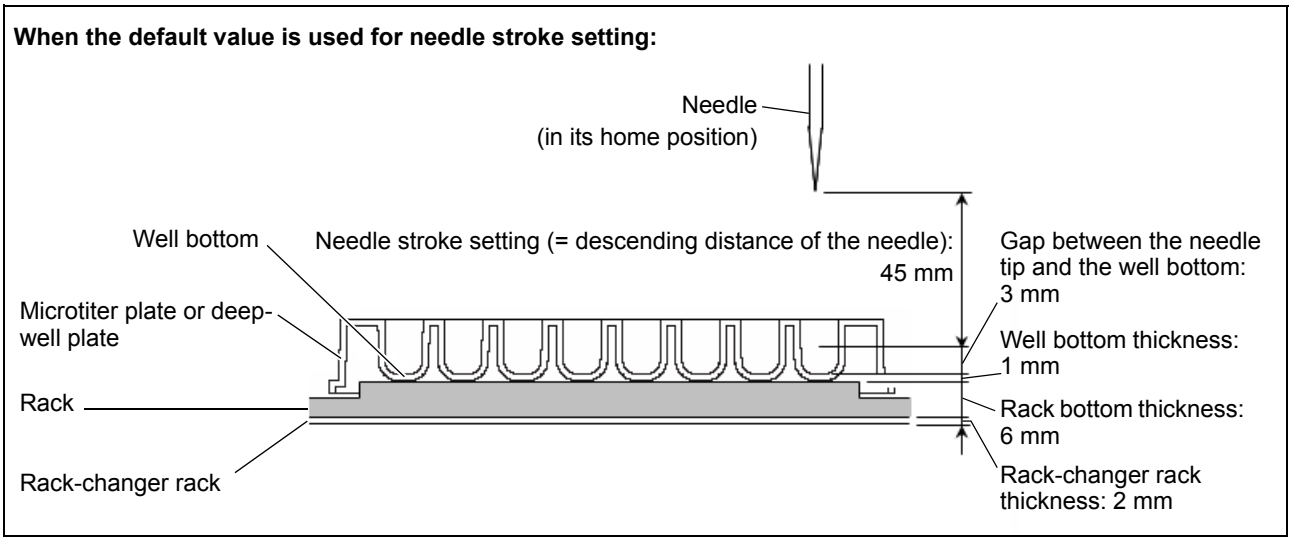


Fig. 5.2

NOTE

The needle stroke setting range and default value for each plate type in combination with a rack changer is given below.

Stack code set value	Plate used	Needle stroke set range (mm)	Needle stroke default value (mm)
0	96-well microtiter plate	10-52	45
1	96-well deep-well plate	10-52	45
2	1.5 mL sample vial plate *1	10-46	44
3	384-well microtiter plate *1	10-52	45
4	384-well deep-well plate *1	10-52	45

*1 When using these plates on the Rack Changer/c (previous model) not equipped with a window on the front panel, be sure to use the Extended Plate Kit for Rack Changer (part No.: S228-50829-91).

⚠CAUTION

When using plastic vials or small-capacity vials with narrow tips, set "42" (mm) or less for the needle stroke.
If a value greater than "42" (mm) is set, since the bottom position of these vials is higher than that of glass vials, there may be cases where the needle hits the bottom, resulting in variations in the amount of sample injection or breakage of the needle tip.

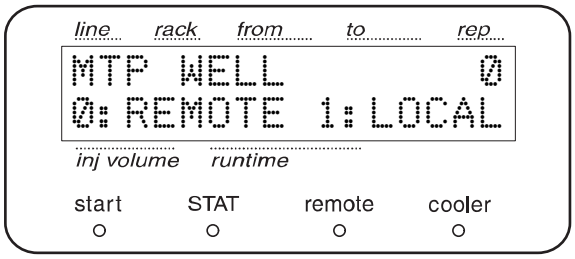
Part Name	Part No.
1.1 mL sample vial	S228-21283-91
1 mL sample vial	S228-31600-91
0.2 mL sample vial	S228-35217-91

Amount of liquid left in the bottom 2 mm of the sample vials.

Part Name	Capacity	Part No.	Amount of liquid 2 mm left at the vial bottom
1 mL vial	1 mL	S228-39699-91	Approx. 65 µL
1.5 mL vial	1.5 mL	S228-15652-92	Approx. 150 µL
1.1 mL vial	1.1 mL	S228-21283-91	Approx. 5 µL
0.3 mL vial	300 µL	S228-16847-92	Approx. 5 µL
1 mL vial	1 mL	S228-31600-91	Approx. 25 µL
0.2 mL sample vial	200 µL	S228-35217-91	Approx. 5 µL
4 mL vial	4 mL	S228-21287-91	Approx. 400 µL
4 mL vial	4 mL	S228-31537-91	Approx. 400 µL
0.3 mL vial	300 µL	S228-21284-91	Approx. 5 µL

■ [MTP WELL]

Set the well type of microtiter plates to be loaded in a rack when microtiter plates are used.
Select whether the well type setting is to be downloaded from the CBM-20A/20Alite or to be set on the instrument.
Enter the set value using the numeric keypad and press **enter**.



Set value	Function
0	Download the selection between 96 wells and 384 wells from the CBM-20A/20Alite.
1	Set up the selection between 96 wells and 384 wells on the instrument.

When setting up the well selection on the instrument, the screen on the right will be displayed next.
Enter the set value using the numeric keypad and press **enter**.

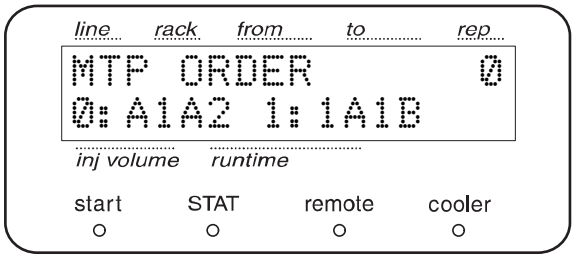
Set value	Function
0	96-well microtiter plates or deep-well plates are used.
1	384-well microtiter plates or deep-well plates are used.

NOTE

When the instrument is connected to the CBM-20A/20Alite via RS-232C cable, well selection cannot be made on the web controller screen. Set "1" at [MTP WELL] and set up the well section on the instrument.

■ [MTP ORDER]

When injecting samples from a microtiter plate, set whether priority is given to columns or rows. Enter the column priority mode (0:A1A2) or the row priority mode (1:1A1B) with the numeric keypad and press **enter**.



Set value	Type of well continuous processing direction
0	Column priority mode (default value). Injection operation is performed in the order A1, A2...
1	Row priority mode Injection operation is performed in the order 1A, 1B...

Sample No. setting in the Column priority mode

When injecting the sample in the Column priority mode, well positions are indicated by entering alphabetical Column letter first, followed by the Row number (e.g.A12).
As a setting example, the setting method when a 96-well microtiter plate is set to Rack 1 position (front of the left rack) and the B2 to B12, C1 to C12, and D1 to D4 wells are analyzed in the order shown in the following figure is described.

To enter the well position "B2" in the sample table, press ABC twice to display "B" (The character on the screen changes as A → B → C → A every time you press the key). Then press enter, and press 0 2 enter with the numeric keypad (The number portion should be entered with two digits. Press 0 2 for 2, and 1 2 for 12).

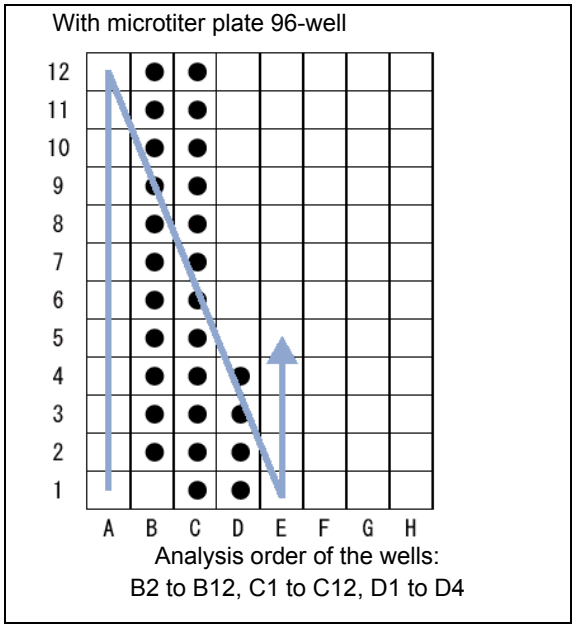


Fig. 5.3

Sample No. setting in the Row priority mode

When injecting the sample in the Row priority mode, well positions are indicated by entering Row number first, followed by the Column letter (e.g.12A).
This section describes an example of the sequence setting method when a 96-well microtiter plate is set to Rack 1 position (front of the left rack) and 2B to 2H, 3A to 3H, 4A to 4D wells are going to be analyzed in the order shown in the following figure.

To enter the well position "2B" in the sample table, press 0 2 enter with the numeric keypad (The number portion should be entered with two digits. Press 0 2 for 2, and 1 2 for 12). Then, press ABC twice to display "B" (The character on the screen changes as A → B → C → A every time you press the key), and press enter.

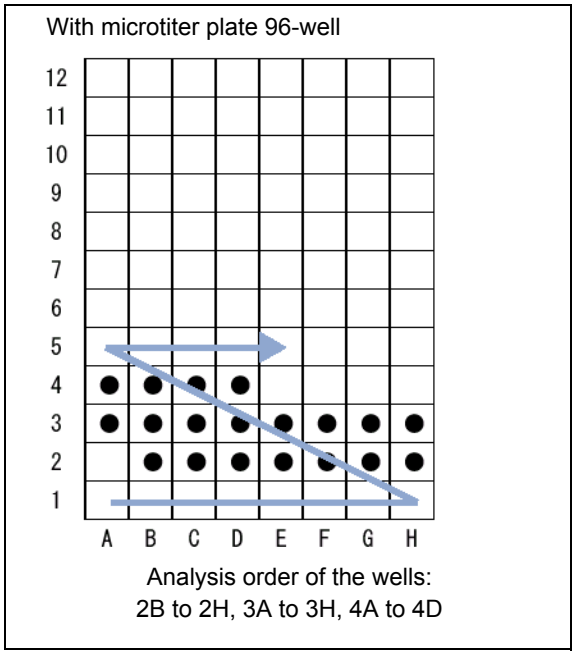
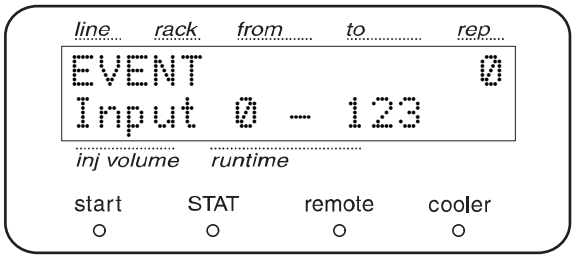


Fig. 5.4

■ [EVENT]

Sets the contact output for external output terminals. It is used, for example, to switch flow-line switching valves. Set the connector number for which the contacts close.



Set value	Function
0	All terminals are opened.
1	Connector 1's terminals are closed.
2	Connector 2's terminals are closed.
3	Connector 3's terminals are closed.
12	Connector 1, 2's terminals are closed.
13	Connector 1, 3's terminals are closed.
23	Connector 2, 3's terminals are closed.
123	Connector 1, 2, 3's terminals are closed.

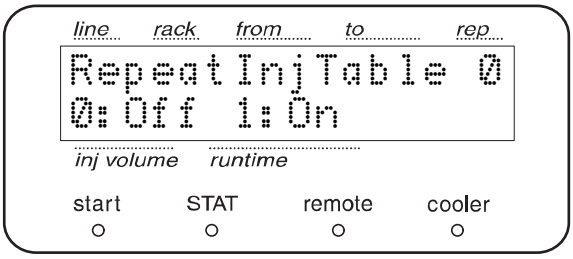
NOTE

When using terminals of connector 1 for the EVENT function, select [EVENT] for [SELECT EVENT 1]. This is the case with terminals of connectors 2 and 3.
No value can be set for terminals if [EVENT] is not selected.

"[SELECT EVENT1]" P. 109, "[SELECT EVENT2]" P. 109, "[SELECT EVENT3]" P. 110

■ [Repeat Inj Table]

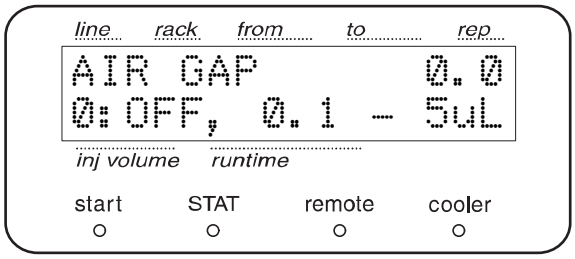
Sets whether or not [Repeat injection] is used.
[Repeat injection] consists of repeatedly executing a repeat injection table at specified line intervals in the analysis sequence table.
Enter the set value by the numeric keypad, and press enter.



Set value	Function
0	Not used (default value).
1	Used.

■ [AIR GAP]

Set the air volume to be trapped before and after the aspirated sample. Enter the air volume using the numeric keypad and press **enter**.
When "0" is input, no air is aspirated.



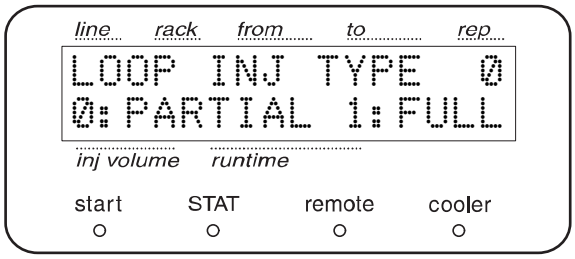
Set range	Default value
0 (OFF) 0.1 to 5 μL	0.0 (default) for total injection method (INJECTION TYPE: 0)
	0.1 (default) for loop injection method (INJECTION TYPE: 1)

■ [LOOP INJ TYPE]

NOTE

This item is displayed when loop injection method (INJECTION TYPE: 1) is selected.

Two loop injection methods are available. Select either partial loop method or full loop method. Enter the set value for the injection method using the numeric keypad and press **enter**.

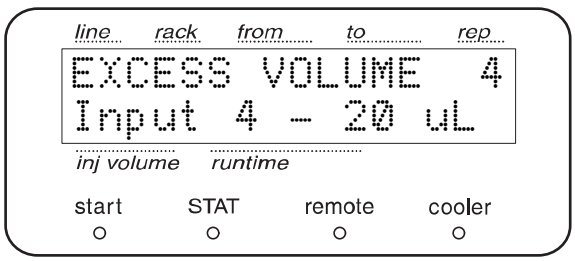


Set value	Function
0	Partial loop method (default)
1	Full loop method

■ [EXCESS VOLUME]

- This item is displayed when loop injection method (INJECTION TYPE: 1) is selected.
- Set a larger value to prevent the sample from being diluted with rinse solution during sample injection.

To change the excess volume of sample to be aspirated, input the volume of aspiration using the numeric keypad, and press **enter**.



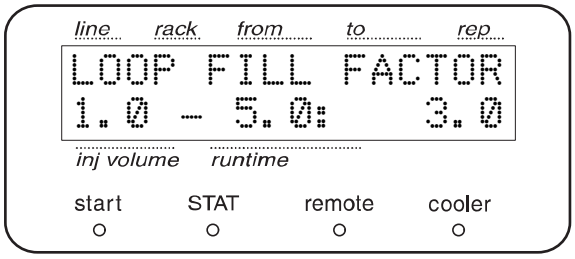
Set range	Default value
4 to 20 μL	10

■ [LOOP FILL FACTOR]

NOTE

This item is displayed when loop injection method (INJECTION TYPE: 1) and full loop method (LOOP INJ TYPE: 1) are selected.

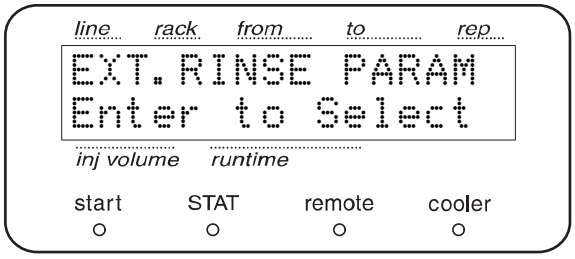
Set how many times of the loop volume the sample should be injected in the case of full loop method.
Enter the number using the numeric keypad and press **enter**.



Set range	Default value
1.0 to 5.0	3.0

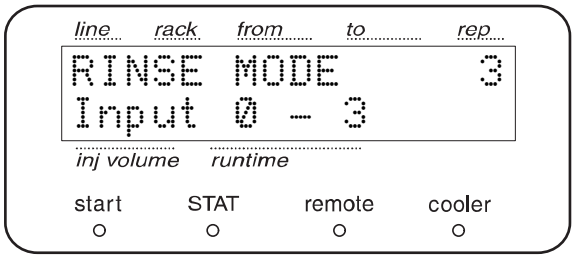
5.2.3 External Rinsing Parameter Settings Group

This is a group of parameters that relate to external rinsing of the needle.



■ [RINSE MODE]

Select the needle rinsing method during sample injection.
Enter the rinse mode number using the numeric keypad and press **enter**.



Set value	Function
0	No rinsing
1	Rinsing before sample aspiration
2	Rinsing after sample aspiration
3	Rinsing before and after sample aspiration (default)

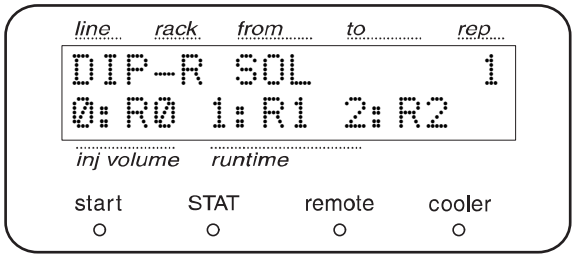
NOTE

When the rinse mode is set to a value other than "0", it is recommended that 450 μL or above be set for [DIP-R VOL] (rinse volume setting) to purge the rinsing port.
☞ "[DIP-R VOL]" P. 94

■ [DIP-R SOL]

NOTE

This item is displayed when "2" is set at [RINSE TYPE].

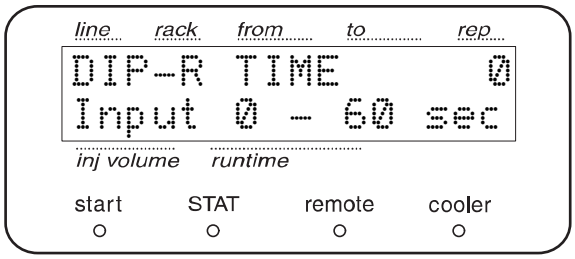


Select the rinse solution type to be used in the rinsing port for external rinsing of the needle. Enter the set value using the numeric keypad and press **enter**.

Set value	Function
0	Rinse solution R0 is filled in the rinsing port.
1	Rinse solution R1 is filled in the rinsing port. (default)
2	Rinse solution R2 is filled in the rinsing port.

■ [DIP-R TIME]

Set the dip time of the needle in the rinsing port for rinsing the needle. Enter the desired time using the numeric keypad, and press **enter**.



Set range	Default value
0 to 60 sec	0 sec

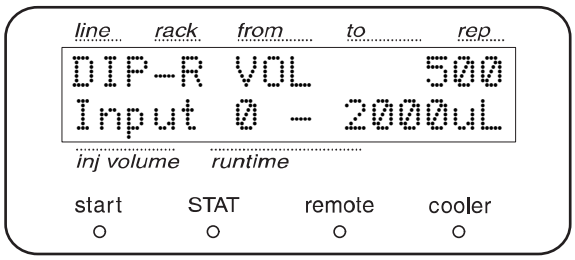
NOTE

When 0 sec is set, the needle is dipped in the rinsing port and immediately moved up from the rinsing port.

■ [DIP-R VOL]

NOTE

This item is displayed when "1" or "2" is set at [RINSE TYPE].



Set the volume of rinse solution to be replaced in the rinsing port, which is contaminated during external rinsing of the needle. Enter the rinse volume using the numeric keypad, and press **enter**.

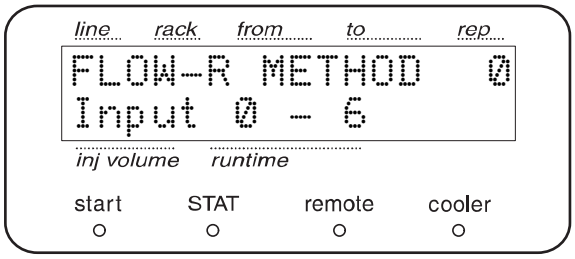
Set range	Default value
0 to 2000 μL	500 μL

NOTE

When the rinse mode is set to a value other than "0", it is recommended that 450 μL or above be set for the rinse volume to be replaced in the rinsing port.

■ [FLOW-R METHOD]

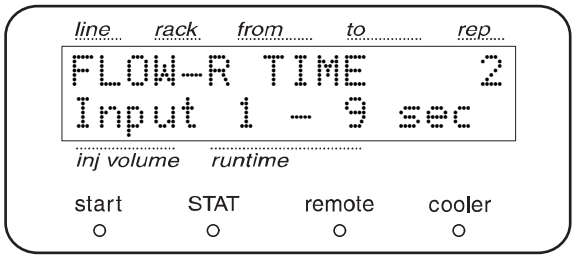
Set the needle rinsing method when using a rinsing pump.
Enter the set value using the numeric keypad and press **enter**.



Set value	Function
0	Not used (default)
1	Rinse the needle by using the rinsing pump only.
2	Rinse the needle by using the rinsing port and a rinsing pump in combination. (The needle is rinsed by pumping and is dip-rinsed in the rinsing port.)
3	Rinse the needle by using the rinsing port and a rinsing pump in combination. (The needle is dip-rinsed in the rinsing port and is rinsed by pumping.)
4	According to the setting at [RINSE MODE], the needle is dip-rinsed in the rinsing port during sample injection and is rinsed by pumping with a rinsing pump at the end of the analysis time. (Example) [RINSE MODE] = 3 [RINSE METHOD] = 4 With the settings shown above: 1) The needle is dip-rinsed in the rinsing port before and after measuring the sample during sample injection. 2) The sample is injected and analysis starts. 3) The analysis time elapses. 4) The needle is rinsed by pumping with the rinsing pump. 5) If analysis continues, steps 1) to 4) will be repeated.
5	Reserved for expansion (cannot be used)
6	Reserved for expansion (cannot be used)

■ [FLOW-R TIME]

Set the rinsing time with a rinsing pump (option).
Enter the rinsing time using the numeric keypad and press **enter**.



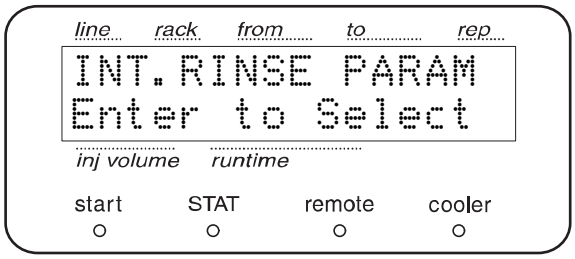
Set range	Default value
1 to 9 sec	2 sec

NOTE

About 1.5 mL rinse solution is used per second.

5.2.4 Internal Rinsing Parameter Settings Group

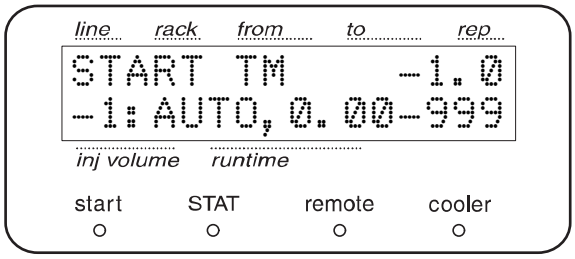
This is a group of parameters that relate to internal rinsing of the needle.



■ [START TM]

NOTE

This item is displayed when total injection method (INJECTION TYPE: 0) is selected and a value other than "2" is set at [RINSE TYPE] .



Set the start time of internal rinsing of the needle during analysis. Select the rinse start time using the numeric keypad, and press **enter**.

Set range	Default value
−1: AUTO 0.00 to 999 min.	−1: Start internal rinsing of the needle on completion of analysis.

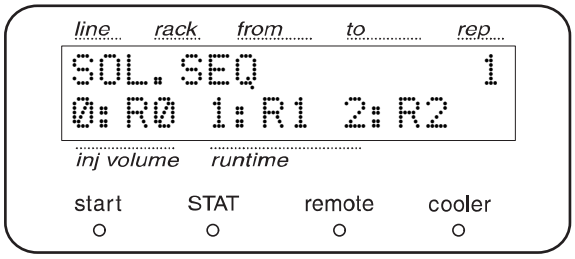
NOTE

- Set the time that counts from the start of analysis.
- The high-pressure valve is switched from the load position to the injection position at the set time, and internal rinsing of the needle starts.
- Set a time that is longer than the time of sample elution from the high-pressure valve. If the setting time is too short, some sample may remain in the sample loop and the needle, and correct analysis results may not be obtained.

■ [SOL. SEQ]

NOTE

This item is displayed when "2" is set at [RINSE TYPE].



Set the type and order of rinse solutions to be used for internal rinsing of the needle. Select the solutions (up to 4) to be used for rinsing using the numeric keypad, and press **enter**.

Set value	Function	Default value
0	Internal rinsing of the needle with rinse solution R0	1
1	Internal rinsing of the needle with rinse solution R1	
2	Internal rinsing of the needle with rinse solution R2	

Example 1: When "1" is set at [SOL. SEQ]:
Internal rinsing of the needle is performed with rinse solution R1.

Rinse solution	R1	➡	R0 *1)	➡	[DIP-R SOL] *1)
Rinse volume	Set value at [SOL. VOLUME]		Set value at [ML PURGE VOL]		Set value at [DIP-R VOL]

Example 2: When "121" is set at [SOL. SEQ]:
Internal rinsing of the needle is performed with rinse solution R1→ R2 → R1.

Rinse solution	R1	➡	R2	➡	R1	➡	R0 *1)	➡	[DIP-R SOL] *1)
Rinse volume	Set value at [SOL. VOLUME]		Set value at [SOL. VOLUME]		Set value at [SOL. VOLUME]		Set value at [ML PURGE VOL]		Set value at [DIP-R VOL]

NOTE

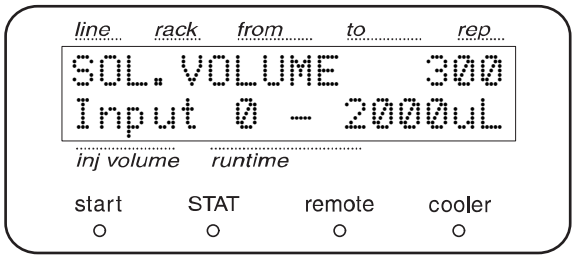
*1) Immediately after the internal rinsing of the needle, replacement with rinse solution R0 (mobile phase) in the sample loop as well as rinsing port will start.

■ [SOL. VOLUME]

NOTE

This item is displayed when "2" is set at [RINSE TYPE].

Set the rinse volume to be used for internal rinsing of the needle. Enter the rinse volume using the numeric keypad and press **enter**.



Set range	Default value
0 to 2000 μL	300 μL

NOTE

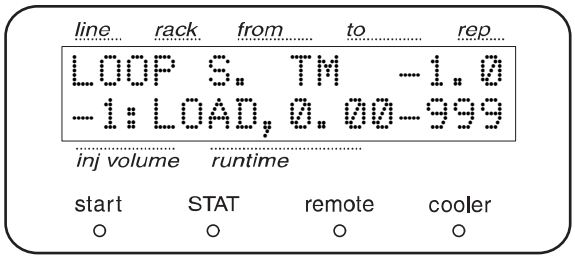
If multiple rinse solutions are selected at [SOL. SEQ], the same volume is used for these rinse solutions. It is not possible to set different rinse volumes for respective rinse solutions.

■ [LOOP S. TM]

NOTE

This item is displayed when total injection method (INJECTION TYPE: 0) is selected and a value other than "2" is set at [RINSE TYPE] .

Set the time to switch the high-pressure valve, which was switched to the load position for internal rinsing of the needle, back to the injection position. Enter the equilibration start time using the numeric keypad, and press **enter**.



Set range	Default value
−1: LOAD 0.00 to 999 min.	−1: The valve position remains in the load position until the next analysis starts.

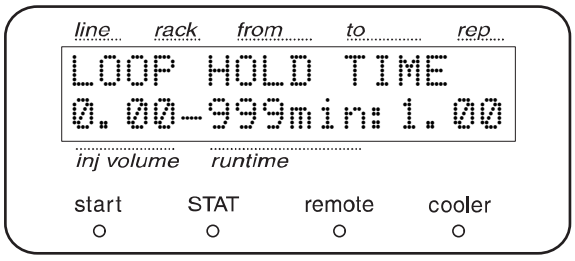
NOTE

- Set the time that counts from the start of analysis.
- If internal rinsing of the needle has not finished by the set time, this function will not start until it is finished.
- After internal rinsing of the needle, the solution in the sample loop and the needle is replaced with rinse solution R0 (mobile phase); however, this is not performed by high-pressure pumping so that rinse solution R1 or R2 may remain in the flow line in trace amounts. Consequently, in the case of internal rinsing of the needle, you are recommended to use the function of switching the high-pressure valve to the injection position during analysis in order to replace the solution in the sample loop and the needle completely. See "5.8.1 Example 1: When Performing the Internal Rinsing of the Needle and Sample Loop Equilibration After Sample Peak Detection (Recommended)" P. 167 in this instruction manual.
- When [-1] is set, the high-pressure valve is switched from the load position to the injection position at the start of the next analysis.

■ [LOOP HOLD TIME]

NOTE

- This item is displayed when total injection method (INJECTION TYPE: 0) is selected and a value other than "2" is set at [RINSE TYPE] and a value other than "–1" is set at [LOOP S. TM].
- If the analysis time has expired during the equilibration of the sample loop, the next analysis will not start until the time set for this function has elapsed.
- The set time here is not the time counted from the start of analysis but the time to hold the high-pressure valve in the injection position.

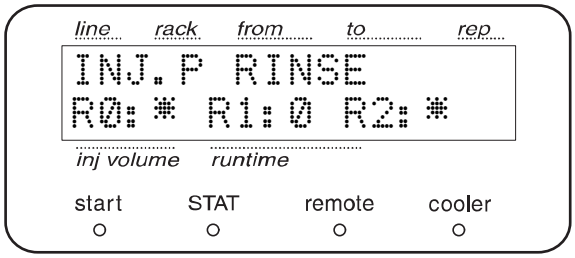


Set the time to keep the high-pressure valve in the injection position for the equilibration of the sample loop. Enter the length of time to keep equilibration using the numeric keypad, and press **enter**.

■ [INJ.P RINSE]

NOTE

This item is displayed when "2" is set at [RINSE TYPE].



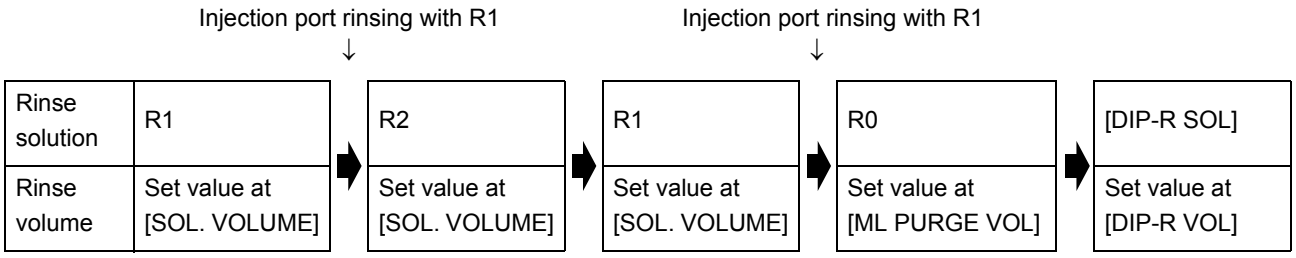
The injection port is rinsed after the internal rinsing of the needle. Enter the set value using the numeric keypad and press (**enter**).

Set value	Function	Default value
0	No rinsing of the injection port after the internal rinsing of the needle	R0: * R1: 0 R2: *
1	Perform rinsing of the injection port after internal rinsing of the needle.	

NOTE

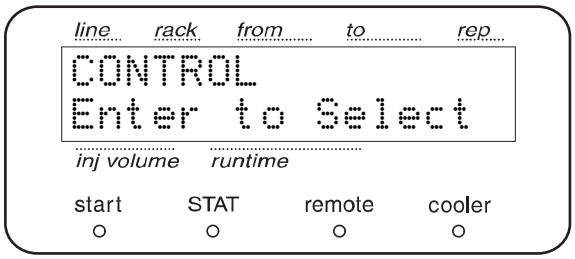
Only the rinse solutions selected at [SOL. SEQ] can be set. "*" is shown for the rinse solution that is not selected.

Example: When "121" is set at [SOL. SEQ] and "R0: * R1: 1 R2: 0" is set at [INJ.P RINSE]:



5.2.5 Control Settings Group

This is the group for system control.

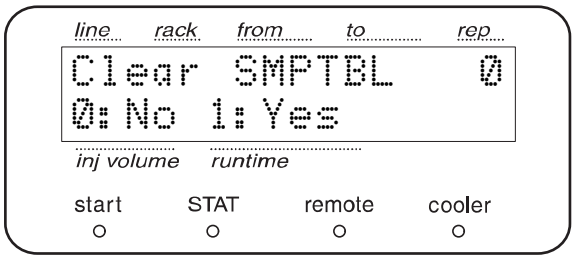


■ [Clear SMPTBL]

Clears the analysis sequence table.

1 From the control setting screen, press **enter**. The [Clear SMPTBL] screen will be displayed.

2 Enter the set value by the numeric keypad and press **enter**.



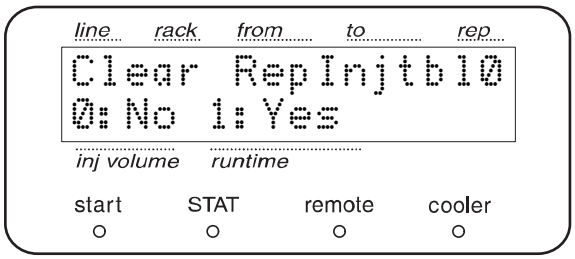
Set value	Function
0	Do not clear the analysis sequence table. (default)
1	Clear the analysis sequence table.

■ [Clear RepInjtbl]

Clears the repeat injection table.

1 From the control setting screen, press **enter**, and press **func** repeatedly until the [Clear RepInjtbl] screen is displayed.

2 Enter the set value by the numeric keypad and press **enter**.



Set value	Function
0	Do not clear the repeat injection table. (default)
1	Clear the repeat injection table.

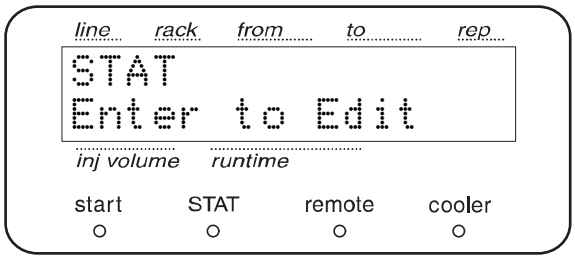
■ [STAT]

Interrupts analysis based on a sample table to perform immediate analysis for one sample.

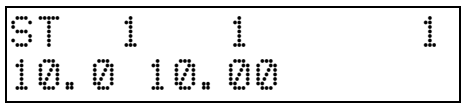
NOTE

- Once a priority analysis has been started, it cannot be canceled.
- When a system controller is connected, this setting is not possible.

1 Press (enter) in the control setting screen and press (func) repeatedly until the [STAT] screen appears.

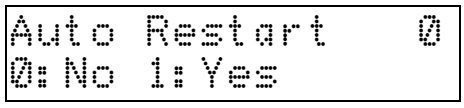


2 Enter the vial number by the numeric keypad and (enter).



3 Enter the injection volume by the numeric keypad and press (enter).

4 Enter the analysis time by the numeric keypad and press (enter).
The next screen specifies how operation is to proceed after the priority analysis ends.



NOTE

To stop priority analysis, press the (CE) to return to the control setting screen.

5 Enter the setting determining the operation after completion of interruption analysis, and press (enter).

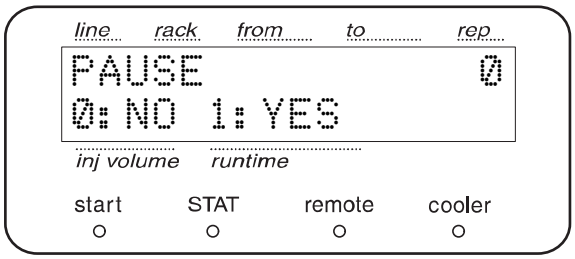
Set value	Function
0	Analysis sequence pauses when priority analysis ends.
1	Analysis sequence resumes when priority analysis ends.

6 The priority analysis will begin as soon as the current sample of the analysis sequence table is analyzed.
The [STAT] LED is illuminated when analysis is being interrupted.

■ [PAUSE]

Stops analysis temporarily.

1 From the control setting screen, press **enter**, and press **func** repeatedly until the [PAUSE] screen is displayed.



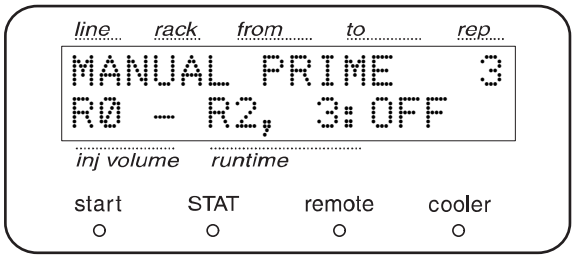
2 Enter the set value by the numeric keypad, and press **enter**.
If [1] is selected, execution is stopped after completion of the analysis currently being executed. The [start] LED is illuminated when [1] is selected.

Set value	Function
0	Analysis is not paused. (default)
1	Analysis is paused.

3 Press **start** to restart analysis.

■ [MANUAL PRIME]

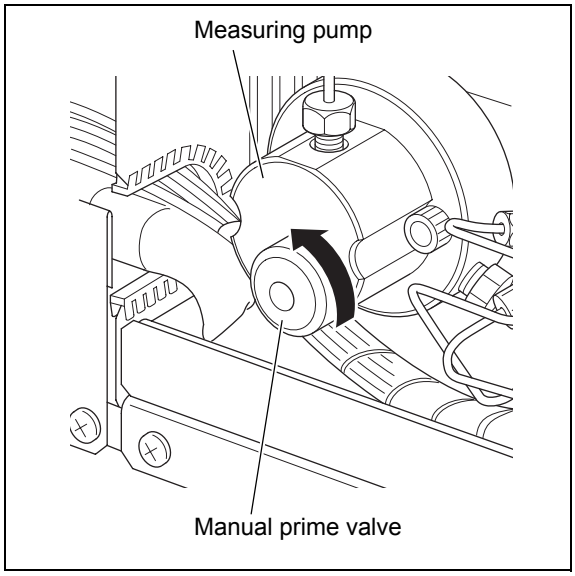
Draws in the rinse solution using the manual syringe. Use the following procedure to perform manual purging.



Set value	Function
0	The low-pressure valve position is switched to the flow line that can draw rinse solution R0.
1	The low-pressure valve position is switched to the flow line that can draw rinse solution R1.
2	The low-pressure valve position is switched to the flow line that can draw rinse solution R2.
3	The drawing of the rinse solution is finished (the low-pressure valve position is reset to the initial state). (default)

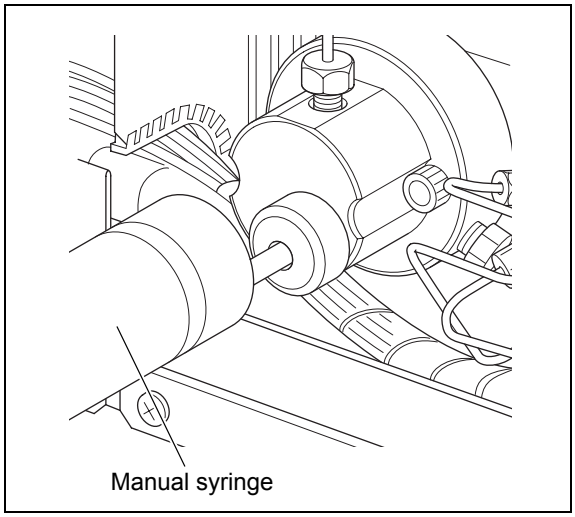
NOTE

This screen is also displayed when (⏚) is pressed on the initial screen.



1 Loosen the manual prime valve at the measuring pump by making a one half to one full turn.

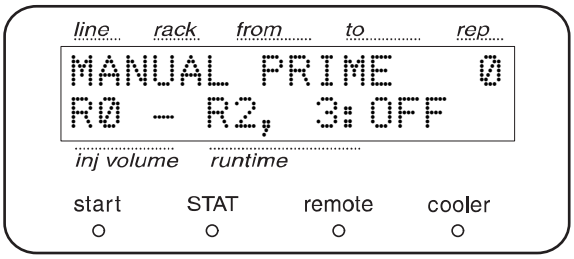
2 Insert a manual syringe into the center hole at the manual prime valve.



NOTE

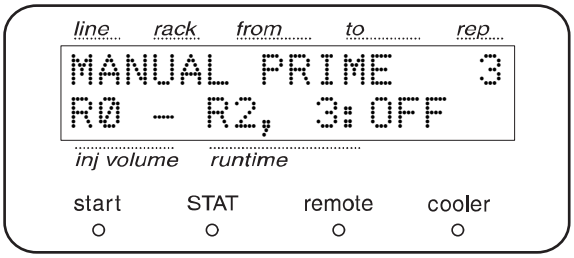
It is not necessary to attach an adapter to the tip of the manual syringe.

3 To draw rinse solution R0, enter the set value [0] and press (**enter**).



4 Draw the rinse solution using the manual syringe until it comes into the syringe.

5 Enter the set value "3" and press (**enter**).



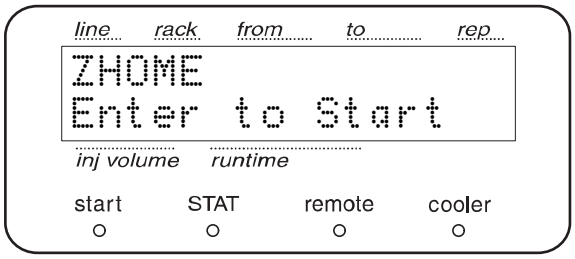
6 Remove the manual syringe and close the manual prime valve by turning it clockwise.

NOTE

If the rinse solution remains at the syringe port at the manual prime valve, wipe it off with wiper paper. In the case of rinse solution R0 (mobile phase), precipitation may result.

■ [Z HOME]

Used when the instrument is not to be used for a long time or before moving the instrument.
Press (enter) to move the needle.
The needle is raised to the highest point and moved to the center of the equipment.



NOTE

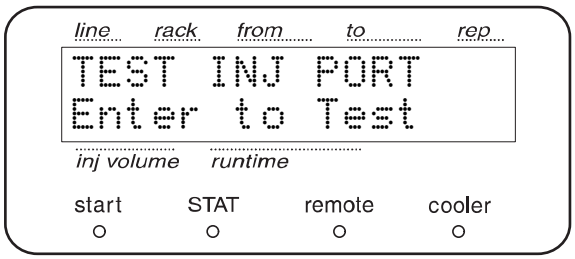
- Press (enter) again to return the needle to the injection port.
- Pressing (⇑) directly moves from the initial screen to the (Z HOME) screen.

■ [TEST INJ PORT]

Checks that the needle is lowered correctly into the injection port.

1 Stop the pump.

2 Press (enter).
The tip of the needle moves up and down twice approximately 2 mm at the position where the injection port is closed. Check that the needle is lowered into the injection port without swaying.



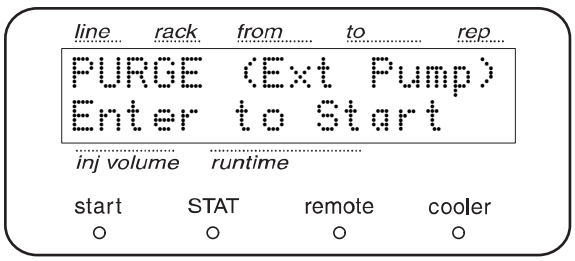
NOTE

Make fine adjustments if the position is incorrect.
☞ "[ADJUST INJ PORT]" P. 151

■ [PURGE (Ext Pump)]

Used to draw in rinse solution with the rinsing pump and rinse the outside of the needle.
Press (enter) to purge.
The following operations are executed.

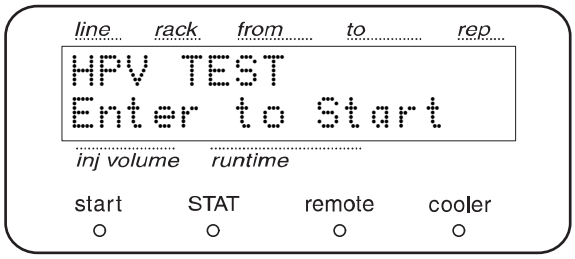
1) The rinsing pump operates for approximately 10 s, and rinse solution is pumped to the rinsing port.
2) The needle moves to the rinsing port.
3) Rinse solution is pumped by the rinsing pump, and the outside of the needle is rinsed for approximately 10 s.
4) The needle returns to the injection port.



■ [HPV TEST]

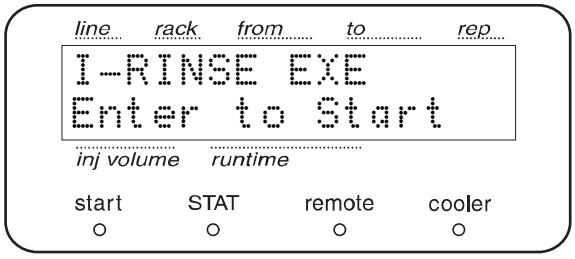
Check the high pressure valve.
After rotating the high pressure valve, send a start signal to the event terminal.
When **enter** is pressed, the high pressure valve is switched from INJECT to LOAD, and is switched back to INJECT immediately.

Use this function to check the high pressure valve, if ghost peaks appear or carry over occurs.
☞ "7.6 Replacing and Inspecting High-Pressure Valve Rotor and Stator" P. 247



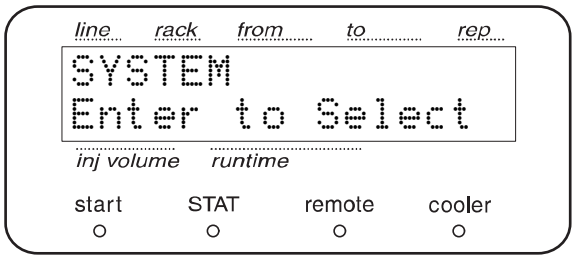
■ [I-RINSE EXE]

Pressing **enter** starts internal and external rinsing of the needle according to the parameter settings including external rinsing parameters and internal rinsing parameters.



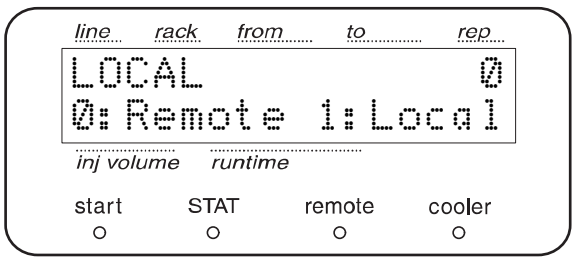
5.2.6 System Settings Group

This is the group for system settings.



■ [LOCAL]

Sets whether this instrument is operated by system controller or the instrument operates independently when system controller is connected.
Enter the desired value, and press **enter**.



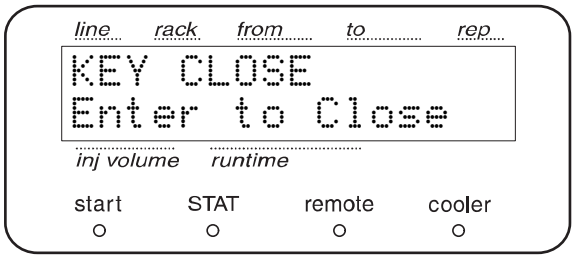
Set value	Mode	Function
0	Remote	Operate via system controller (initial setting).
1	Local	Operate independently (in local mode).

■ [KEY CLOSE]

Press (enter) to prohibit key entry.
After this, key operation is not available.

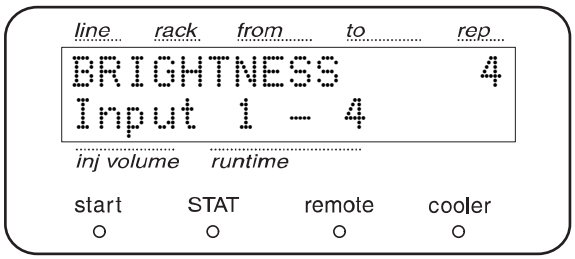
NOTE

To release this function, press (shift) while pressing (del).



■ [BRIGHTNESS]

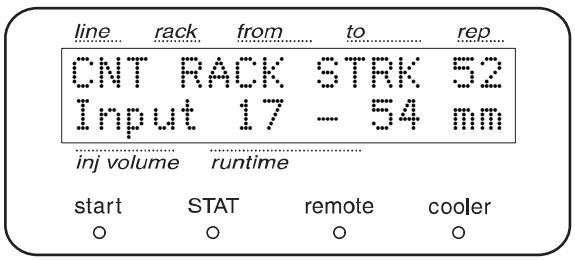
Sets the brightness of display screen.
Enter the set value and press (enter).
Value range is 1 to 4 and 4 is the brightest.



■ [CNT RACK STRK]

Sets the distance that the needle is lowered to the control vial rack.
Enter the stroke value with the numeric keypad, and press (enter).

Set range	Default value
17-54	52



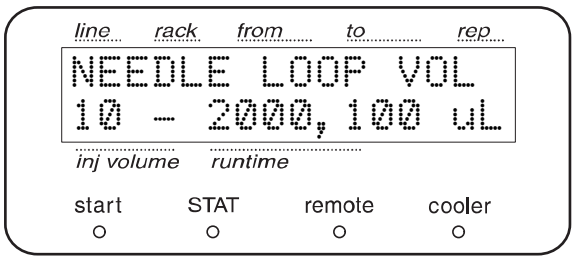
■ [NEEDLE LOOP VOL]

NOTE

This item is displayed when loop injection method (INJECTION TYPE: 1) is selected.

Set the maximum volume that can be aspirated at one time when the loop injection method is selected.
In the partial loop method, ensure that the volume of "sample injection volume" + "air gap" × 2 + "excess volume" is not greater than the setting value.
In the full loop method, ensure that the volume of "sample injection volume" × "preload factor" + "air gap" × 2 + "excess volume" is not greater than the setting value.

Set Range	Default value
0 - 2000 μL	100 μL



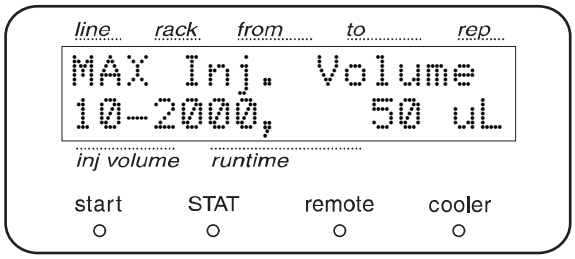
NOTE

Set a value that is smaller than the total volume (214.3 μL) of the needle (11.7 μL), movable sample loop (84.8 μL), and extension tubing (117.8 μL).

■ [MAX INJ. VOLUME]

NOTE

This item is displayed when total injection method (INJECTION TYPE: 0) is selected.



Sets the maximum injection volume. When creating a sample table, specify a value smaller than value for the injection volume.
Enter the maximum injection volume using the numeric keypad and press **enter**.

Set range	Default value
10 to 2000 μL	50 μL

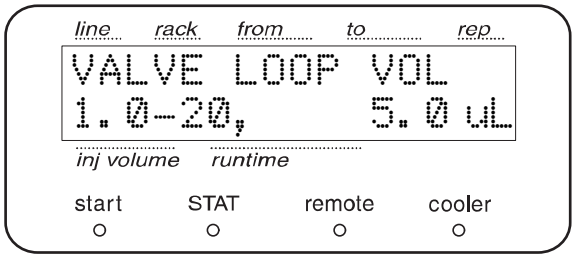
NOTE

With the system controller CBM-20A/20Alite or workstation LCsolution/LabSolutions, the set value for the maximum volume of injection can be displayed but the setting is not possible.

■ [VALVE LOOP VOL]

NOTE

This item is displayed when loop injection method (INJECTION TYPE: 1) is selected.



Set the capacity of sample loop (option).
The volume of injection ([Inj. volume]) used for creating an analysis sequence table must not exceed 70 % of the loop capacity for the partial loop method or 100 % for the full loop method.
Enter the sample loop volume using the numeric keypad and press **enter**.

Set range	Default value
1.0 to 20	5.0 μL

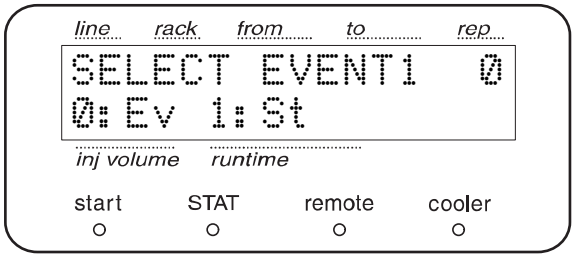
NOTE

The volume of sample aspiration for full loop injection is determined based on the set values here and at [LOOP FILL FACTOR].

"5.10.5 Parameters" P. 205

■ [SELECT EVENT1]

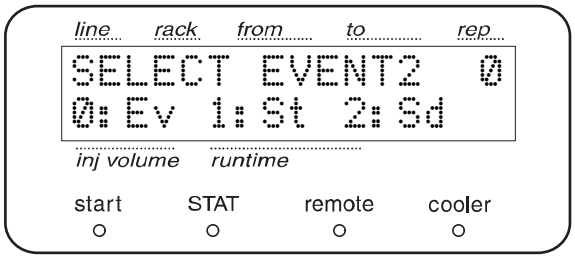
Selects the output of No. 1 external output terminals. This is used, for example, to switch flow-line valves. Enter the set value using the numeric keypad and press **enter**.



Set value	Function
0	Used as event output (default setting).
1	Short-circuit output for six seconds at the start of analysis

■ [SELECT EVENT2]

Selects the output of No. 2 external output terminals. This is used, for example, to switch flow-line valves. Enter the set value using the numeric keypad and press **enter**.



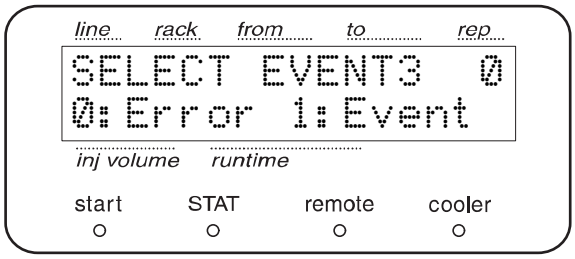
Set value	Function
0	Used as event output (default setting).
1	Short-circuit output for six seconds at the start of analysis
2	Contacts close on completion for six seconds of all lines in the sample table.

NOTE

The set value "2" is valid only when the instrument is used on a standalone basis without connecting the system controller.

■ [SELECT EVENT3]

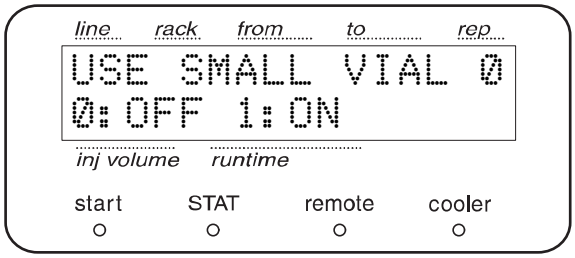
Selects the output of No.3 external output terminals. This is used, for example, to switch external valves.
Enter the set value by the numeric keypad and press **enter**.



Set value	Function
0	Short-circuit output for six seconds at the occurrence of an error (default setting).
1	Used as event output.

■ [USE SMALL VIAL]

When samples vials with a small capacity are used, atmospheric changes in the sample vials during sample measurement can cause incorrect measurements. This function can be used to reduce the influence of these changes in atmospheric pressure.
Select [1: On] when using small-capacity vials.



Set value	Function
0	Small-capacity vials are not used (default setting).
1	Small-capacity vials are used.

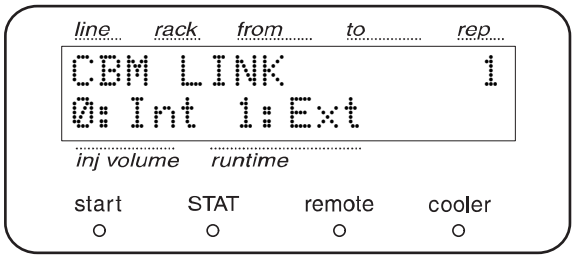
When "1: On" is selected, the screen on the right is displayed. Set the stroke of moving down the needle at the first time.

NOTE

- If correct measurement is not possible because air is trapped inside the needle, such as when a microtiter plate mat is used, select [1: On]. The condition may be improved.
- It would be effective to set the first needle stroke so that the needle will pass through the septum or mat by a few millimeters.

■ [CBM LINK]

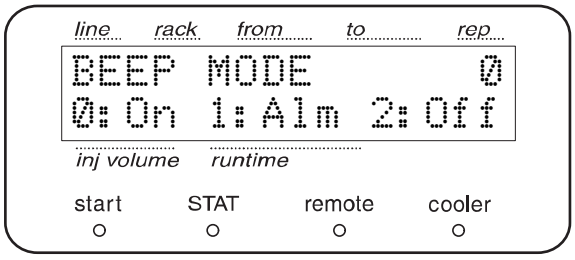
Sets the link destination of system controller.
Enter the desired value, and press **enter**.
After setting, turn off the power and then turn it on again.



Set value	Function
0	Link with CBM-20Alite (option) inside of the instrument.
1	Link with external system controller by optical cable connected to [REMOTE] connector.

■ [BEEP MODE]

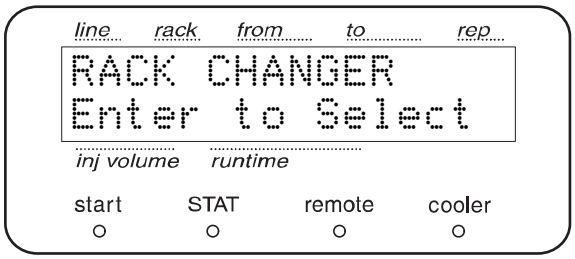
Sets operation of buzzer.
Enter a set value and press **enter**.



Set value	Function
0	Alarm sound when error occurs and key entry sound are enabled. (default setting)
1	Alarm sounds when an error occurs. Key entry sounds are disabled.
2	All sounds are disabled.

5.2.7 Changer Settings Group

Displayed when using an optional rack changer.

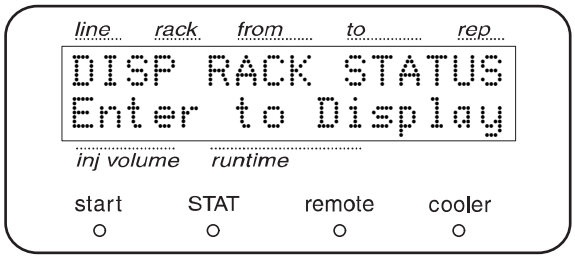


NOTE

When using the plates marked with "*1)" (384-well microtiter plates, 384-well deep-well plates, 1.5 mL sample vial plates) on the Rack Changer/c (previous model) not equipped with a window on the front panel, be sure to use the Extended Plate Kit for Rack Changer (part No.: S228-50829-91).

■ [DISP RACK STATUS]

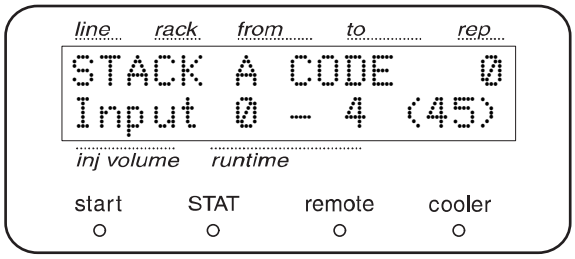
1 From the changer setting screen, press **enter**.
The [DISP RACK STATUS] screen is displayed.



2 Press **enter** to display the status of racks 1 to 12 in order.
The rack number and type are displayed on the first line and the rack status is displayed on the second line.

■ [STACK A CODE]

Enter the set value by the numeric keypad and press **enter**.



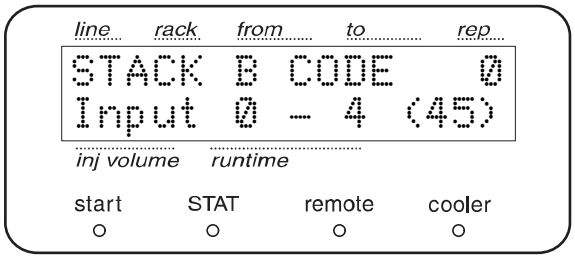
Set value	Function
0	96-well microtiter plate is used.
1	96-well deep-well plate is used.
2	1.5 mL sample vial plate is used. *1)
3	384-well microtiter plate is used. *1)
4	384-well deep-well plate is used. *1)

NOTE

The figure in parentheses at the bottom right corner of the display screen shows the set needle stroke value.

■ [STACK B CODE]

Enter the set value by the numeric keypad and press **enter**.



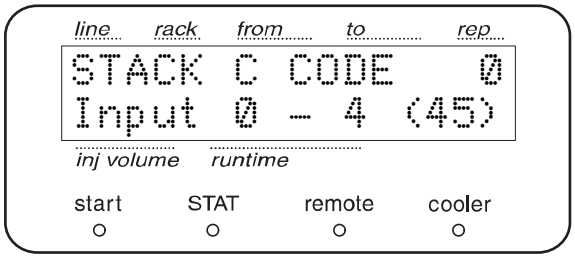
Set value	Function
0	96-well microtiter plate is used.
1	96-well deep-well plate is used.
2	1.5 mL sample vial plate is used. *1)
3	384-well microtiter plate is used. *1)
4	384-well deep-well plate is used. *1)

NOTE

The figure in parentheses at the bottom right corner of the display screen shows the set needle stroke value.

■ [STACK C CODE]

Enter the set value by the numeric keypad and press **enter**.



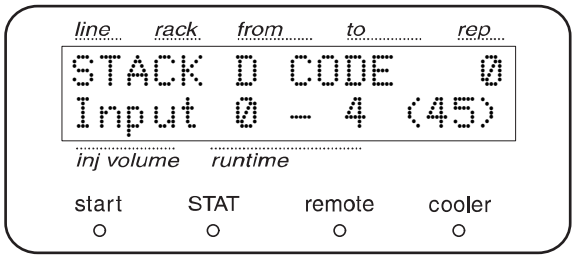
Set value	Function
0	96-well microtiter plate is used.
1	96-well deep-well plate is used.
2	1.5 mL sample vial plate is used. *1)
3	384-well microtiter plate is used. *1)
4	384-well deep-well plate is used. *1)

NOTE

The figure in parentheses at the bottom right corner of the display screen shows the set needle stroke value.

■ [STACK D CODE]

Enter the set value by the numeric keypad and press **enter**.



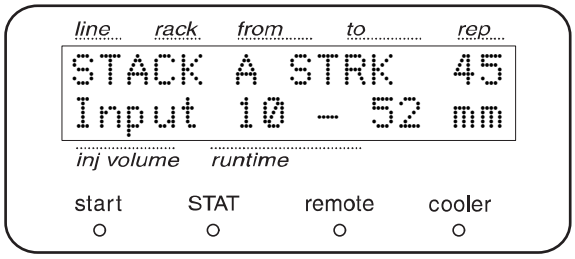
Set value	Function
0	96-well microtiter plate is used.
1	96-well deep-well plate is used.
2	1.5 mL sample vial plate is used. *1)
3	384-well microtiter plate is used. *1)
4	384-well deep-well plate is used. *1)

NOTE

The figure in parentheses at the bottom right corner of the display screen shows the set needle stroke value.

■ [STACK A STRK]

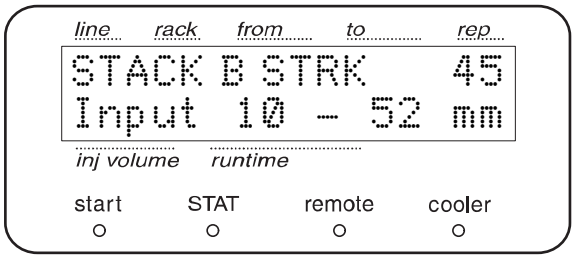
Sets the distance of moving down the needle for stack A. Depending on the stack code used, the default value and setting range differ. Enter the stroke value using the numeric keypad and press **enter**. The default value and setting range for each stack code is given in the table below.



Stack code set value	Function	Set range	Default value (Units: mm)
0	96-well microtiter plate is used.	10-52	45
1	96-well deep-well plate is used.	10-52	45
2	1.5 mL sample vial plate is used. *1)	10-46	44
3	384-well microtiter plate is used. *1)	10-52	45
4	384-well deep-well plate is used. *1)	10-52	45

■ [STACK B STRK]

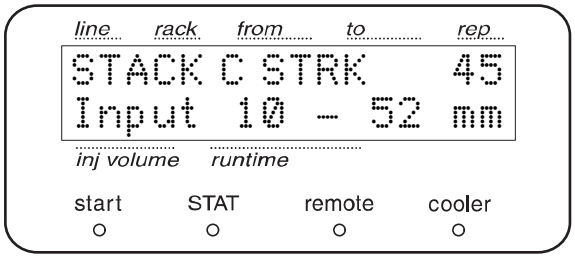
Sets the distance of moving down the needle for stack B. Depending on the stack code used, the default value and setting range differ. Enter the stroke value using the numeric keypad and press (**enter**). The default value and setting range for each stack code is given in the table below.



Stack code set value	Function	Set range	Default value (Units: mm)
0	96-well microtiter plate is used.	10-52	45
1	96-well deep-well plate is used.	10-52	45
2	1.5 mL sample vial plate is used. *1)	10-46	44
3	384-well microtiter plate is used. *1)	10-52	45
4	384-well deep-well plate is used. *1)	10-52	45

■ [STACK C STRK]

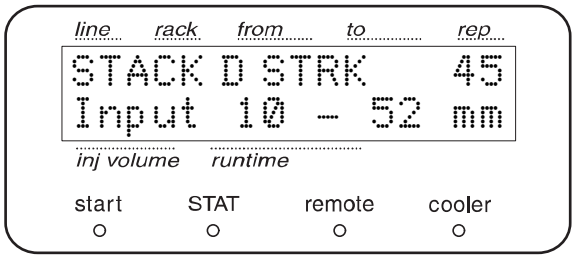
Sets the distance of moving down the needle for stack C. Depending on the stack code used, the default value and setting range differ. Enter the stroke value using the numeric keypad and press (**enter**). The default value and setting range for each stack code is given in the table below.



Stack code set value	Function	Set range	Default value (Units: mm)
0	96-well microtiter plate is used.	10-52	45
1	96-well deep-well plate is used.	10-52	45
2	1.5 mL sample vial plate is used. *1)	10-46	44
3	384-well microtiter plate is used. *1)	10-52	45
4	384-well deep-well plate is used. *1)	10-52	45

■ [STACK D STRK]

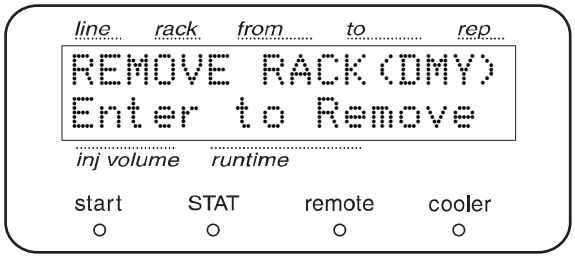
Sets the distance of moving down the needle for stack D. Depending on the stack code used, the default value and setting range differ. Enter the stroke value using the numeric keypad and press **enter**. The default value and setting range for each stack code is given in the table below.



Stack code set value	Function	Set range	Default value (Units: mm)
0	96-well microtiter plate is used.	10-52	45
1	96-well deep-well plate is used.	10-52	45
2	1.5 mL sample vial plate is used. *1)	10-46	44
3	384-well microtiter plate is used. *1)	10-52	45
4	384-well deep-well plate is used. *1)	10-52	45

■ [REMOVE RACK]

Use this function when removing the rack changer from the autosampler. Press **enter** to return the rack remaining inside the autosampler to the rack changer or to return the dummy rack to the neutral position.

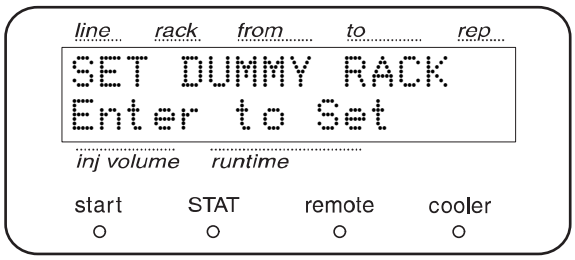


NOTE

- This screen is also displayed when ← is pressed on the initial screen.
- If any figure is displayed in parentheses at the top right corner of the display screen, it indicates the rack number remaining inside the autosampler. If "(DMY)" is displayed, it indicates that the dummy rack remains inside the autosampler.

■ [SET DUMMY RACK]

Use this function when setting the dummy rack in the rack-changer rack. Press **enter** to set the dummy rack in the neutral position to the rack-changer rack.

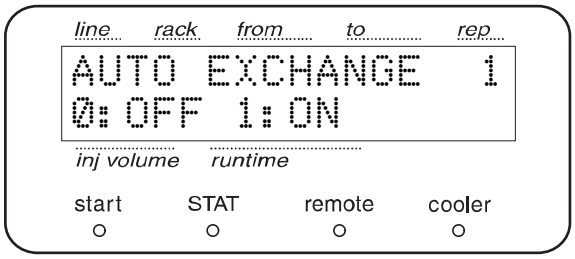


NOTE

If "(DUMMY IN SIL)" is displayed on the screen, it indicates that the dummy rack remains inside the autosampler.

■ [AUTO EXCHANGE]

If it is necessary to change the rack for the next analysis, the rack can be changed to the next one during the current analysis period.
Enter the set value using the numeric keypad and press **enter**.



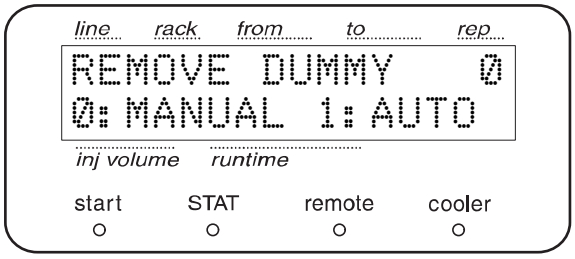
Set value	Function
0	Change the rack to the next one on completion of the current analysis.
1	Change the rack to the next one during the current analysis period. (default)

NOTE

When the pretreatment program of the autosampler is customized, this item is not effective even if "1" is set. The rack will be changed to the next one on completion of the current analysis.

■ [REMOVE DUMMY]

On completion of analysis the dummy rack is automatically moved to the neutral position. This is used when removing the rack-changer rack after analysis.
Enter the set value using the numeric keypad and press **enter**.



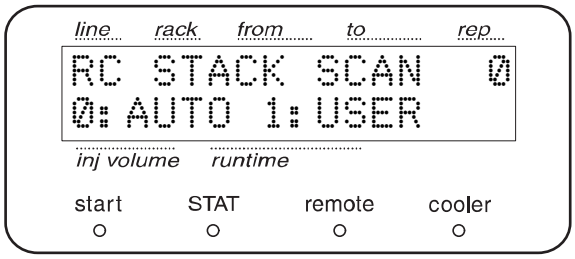
Set value	Function
0	On completion of analysis the dummy rack is not moved to the neutral position. (default)
1	On completion of analysis the dummy rack is moved to the neutral position.

NOTE

When "1" is set on the model equipped with a sample cooler, the sample cooler is automatically turned OFF before moving the dummy rack to the neutral position. (The sample cooler at the rack changer remains ON.) It is automatically turned ON when the rack-changer rack is removed and another rack (rack-changer racks or other racks) is installed.

■ [RC STACK SCAN]

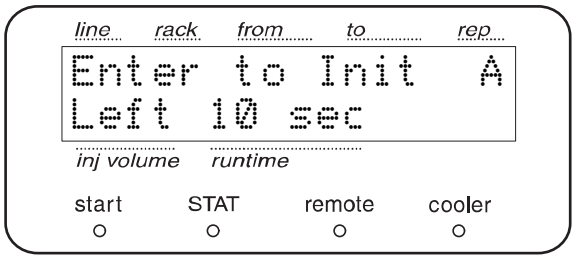
Determine whether or not to perform rack check operation when the stack has been installed.
Enter the set value using the numeric keypad and press **enter**.



Set value	Function
0	Rack check operation is automatically performed when the stack has been installed. (default)
1	Rack check operation is not performed when the stack has been installed.

NOTE

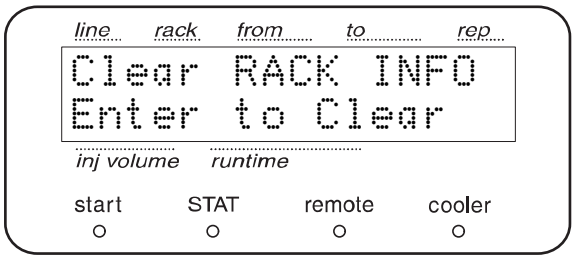
- When "1" is selected, press **enter** within 10 seconds while the screen at the right is displayed after the stack has been installed, to perform rack check operation at the stack.
- The screen at the right is displayed during the rack check operation.
- When "1" is selected, if you remove the rack from the stack and attempt to start analysis with its rack number specified, a rack set warning will occur and stop analysis.
  Within 1 minute after the warning has occurred, set a rack to the rack number position specified for analysis. If you do not set it within 1 minute, a no-rack error will occur and stop analysis.

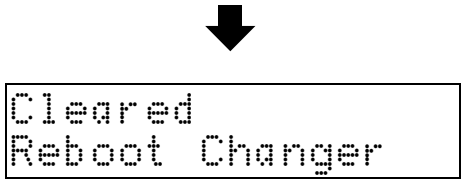


■ [Clear RACK INFO]

The rack changer keeps the information of rack presence or absence at each stack.
Use this function to delete the rack information.
Press (enter), and the rack information will be deleted and the screen at the right will be displayed. Turn the rack changer power OFF and ON.

☞ "4.5 Turning ON the Power" in the Rack Changer Instruction Manual





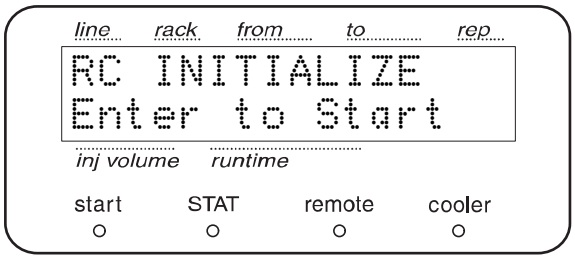
NOTE

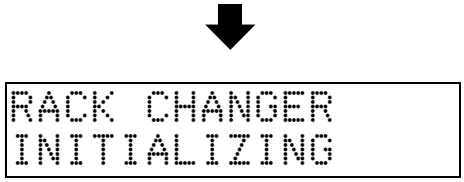
When the rack information has been deleted, turn the rack changer power OFF and ON, or perform initialization (rack check operation for all stacks) from the RC INITIALIZE menu.

☞ "[RC INITIALIZE]" P. 119

■ [RC INITIALIZE]

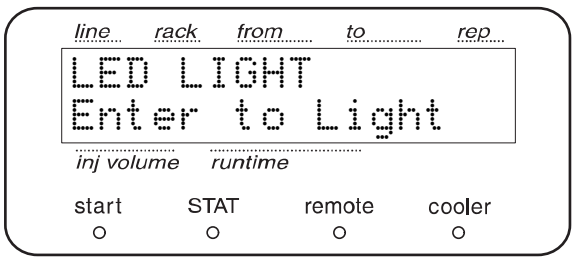
Rack check operation is performed for all stacks.
Press (enter), and the rack changer will perform rack check operation for all stacks and update the display on the rack changer to the current state.
The screen at the right is displayed during the rack check operation.





■ [LED LIGHT]

Press (enter) to turn on the LED inside the Rack Changer II for about 10 seconds.



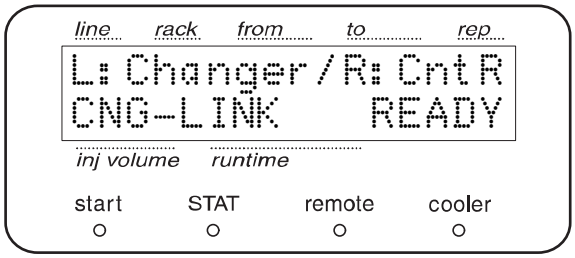
NOTE

This is enabled only when the Rack Changer II with LED illumination is displayed.

5.3 Using a Rack Changer

By using the rack changer, analysis is possible with a maximum of 12 microtiter plates, deep-well plates or 1.5-mL vial plates. Use the rack changer according to the procedure described below.
(☞ Refer to the Rack Changer II Instruction Manual.)

1 Set the changer rack in the autosampler.
Verify the communication between the autosampler and the rack changer.
- The screen shown on the right is displayed.
- The rack changer's [READY] lamp is illuminated.



2 Set the plates in the rack changer.

Stack number	Adapter number (rack number input in the sample table)
Stack A	1, 2, and 3 from the stack front side
Stack B	4, 5, and 6 from the stack front side
Stack C	7, 8, and 9 from the stack front side
Stack D	10, 11, and 12 from the stack front side

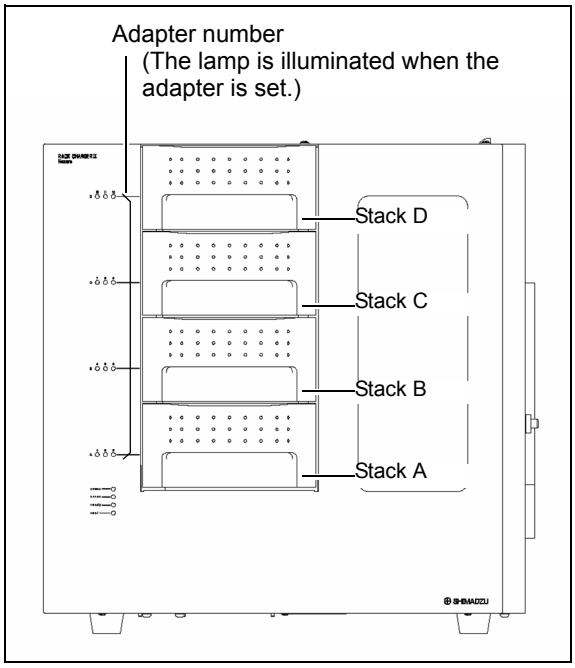


Fig. 5.5

NOTE

When performing injection from a control vial rack, set [0] as the rack number in the sample table.

NOTE

When using an autosampler with a rack changer, analysis is possible using a rack other than the changer rack.
Before removing the changer rack from the autosampler, check that there is no adapter on the rack. If an adapter is left, return it to the rack changer.
☞ "[REMOVE RACK]" P. 116

5.4 Setting Bracket Sequence Analysis

When a specific number of samples is to be injected, this mode can be used to inject samples repeatedly, at periodic intervals. In this mode, a special repeat injection table (in additional to the standard injection mode analysis sequence table) is created.
An example of a repeat injection table is provided below.

Analysis based on the following sample injection table and repeat injection table (interval = 4) is performed in the way described below.

Sample injection table

line	rack	from	to	rep	inj vol	runtime
0	1	0	9	10	10	10
1	1	20	30	10	10	30
2	1	30	39	1	10	30
3	1	50	59	10	10	30
4	1	60	60	1	200	30
5	1	70	79	1	100	30
6	1	71	71	1	100	30
7	1	72	72	1	100	30
8	1	73	73	1	100	30

Repeat injection table

line	rack	from	to	rep	inj vol	runtime
0	1	80	80	1	100	30
1	1	81	81	5	100	30
2	1	82	82	1	100	30

Interval = 4

NOTE

The display cannot be used when CBM-20A/20Alite is connected.

1 First, the repeat injection table (lines 0 through 2) is executed.

2 Next, lines 0 through 3 of the analysis sequence table are executed.
(Since the Interval is 4, the repeat injection table is executed every fourth line of the analysis sequence table.)

3 The repeat injection table (lines 0 through 2) is executed again.

4 Lines 4 through 7 of the analysis sequence table are executed.

5 The repeat injection table (lines 0 through 2) is executed once more.

6 Line 8 of the analysis sequence table is executed. All the analyses have now been completed.

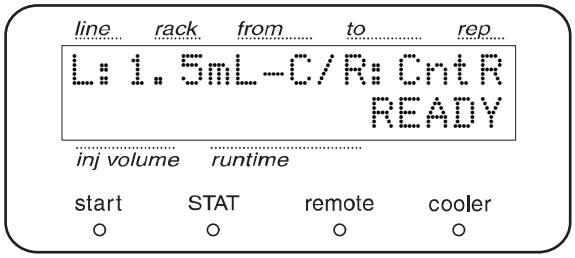
Use the following procedure to create a sample table and repeat injection table for this type of analysis.

5.4.1 Creating a Repeat Injection Table

■ Displaying the Setting Screen

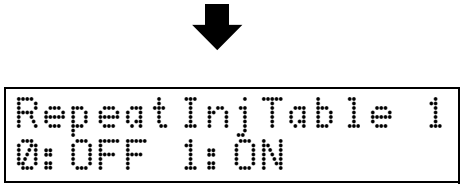
1 Press **CE** to return to the initial screen.

2 Press **func** to display the parameter setting screen.

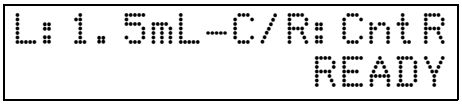


3 From the parameter setting screen, press **enter**, and press **func** repeatedly until the [RepeatInjTable] screen is displayed.

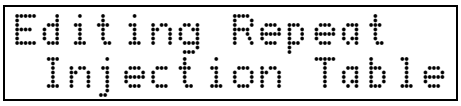
4 Enter **1** and press **enter**.



5 Press **CE** two times to return to the initial screen.

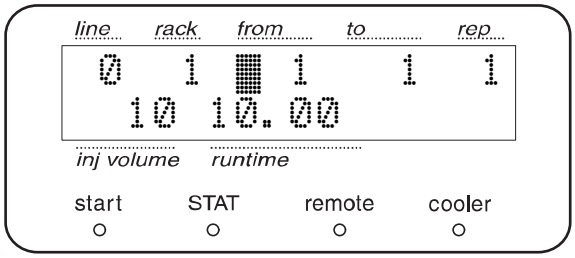


6 Press **edit**.
The screen on the right will be briefly displayed, to be replaced by the parameter setting screen.

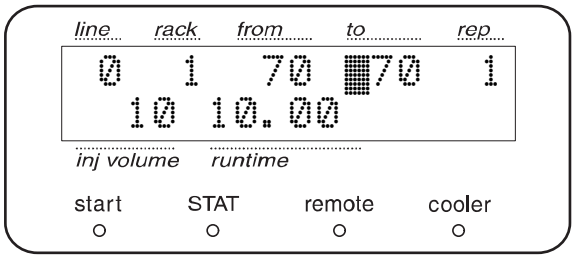


■ Setting Repeat Injection Table

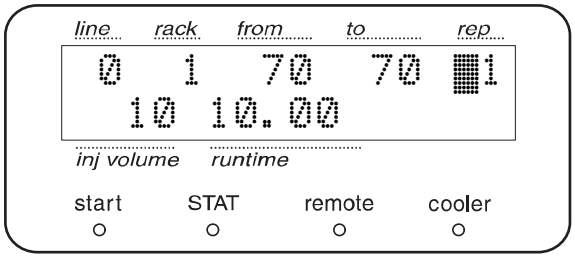
1 Use numeric keypad to enter the rack number, and press **enter**.The cursor will move to the [from] field.



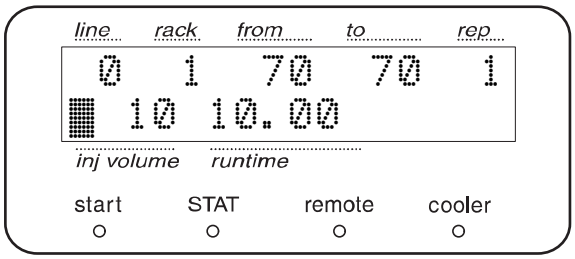
2 Use numeric keypad to enter the number of the first vial to be injected, and press **enter**.The cursor will move to the [to] field.



3 Enter the number of the last vial to be injected, and press **enter**.The cursor will move to the [rep] field.



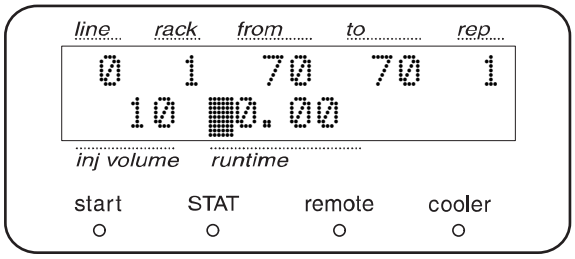
4 Enter the number of injections to be made from each sample vial, and press **enter**. The cursor will move to the [inj volume] field on the second line.



NOTE

The maximum setting for the number of injections is 30.

5 Enter the injection volume by the numeric keypad, in µL and press **enter**.The cursor will move to the [runtime] field.



NOTE

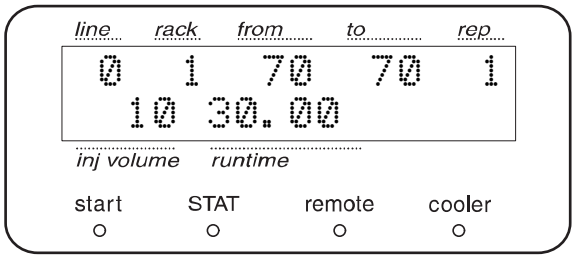
Setting is possible in increments of 0.1 µL from 0.1 to 9.9 µL or in increments of 1 µL from 10 to 50 µL. (20 µL maximum for loop injection method) Set a value smaller than the maximum injection volume.

With the system controller CBM-20A/20Alite or workstation LCsolution/LabSolution, the set value for the maximum volume of injection can be displayed but the setting is not possible.

For the total injection method, see "[MAX INJ. VOLUME]" P. 108

For the loop injection method, see "[VALVE LOOP VOL]" P. 108

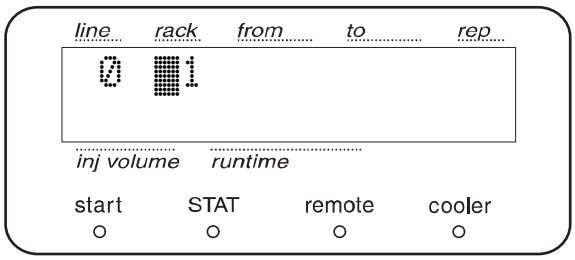
6 Enter the analysis time, in minutes, and press **enter**.



NOTE

Any value between 0.01 and 9999 minutes can be set. Values higher than 100 minutes must be in 1 minute increments.

7 Press **enter**.
The screen for setting the next line appears.



8 Repeat steps 1 to 7 and set the next line.

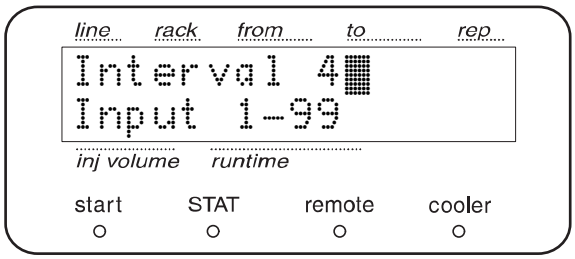
NOTE

- Up to 10 lines can be set in the repeat injection table.
- If no further lines are required, press **CE**.

9 Press **CE** when the table settings have been completed.

■ Setting Interval Conditions

1 Press **enter**.
The [Interval] screen is displayed.

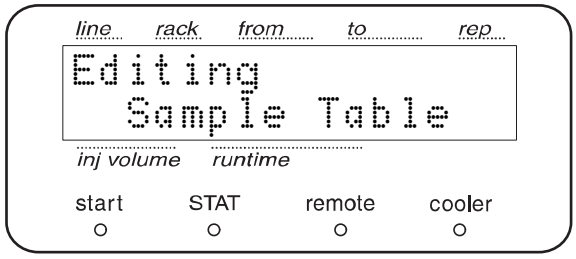


2 Using the numeric keypad, set the interval between insertions in terms of the number of lines in the analysis sequence created with the sample table. In this case, enter **4**. The setting range is from 1 to 99 lines.

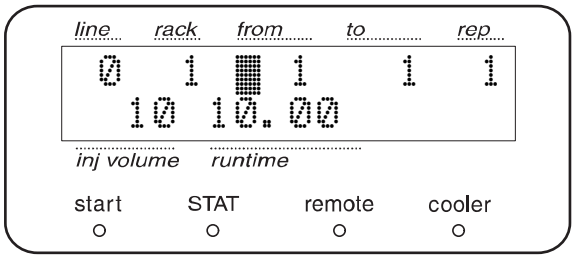
3 Press **enter**.
The input value is validated.

■ Setting Sample Table

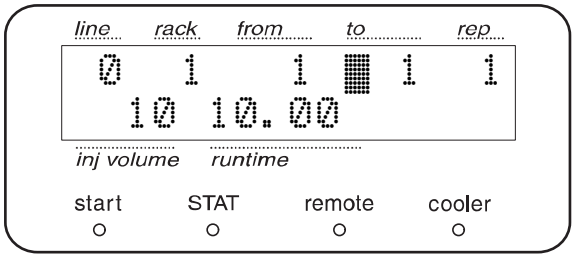
1 The sample table screen will be briefly displayed, and then replaced by the parameter setting screen.



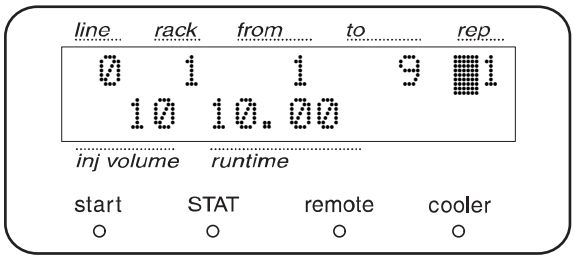
2 Use the numeric keypad to enter the first rack number, and press **enter**.The cursor will move to the [from] field.



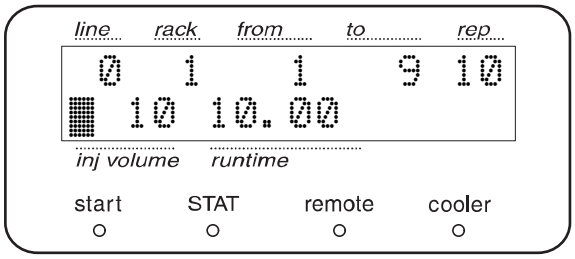
3 Use numeric keypad to enter the number [0] of the first vial to be injected, and press **enter**.
The cursor will move to the [to] field.



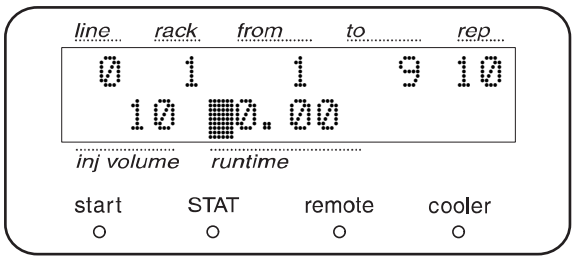
4 Enter the number of the last vial to be injected, and press **enter**.The cursor will move to the [rep] field.



5 Enter the number of injections to be made from each sample vial, and press **enter**.
The cursor will move to the [inj volume] field on the second line.



6 Enter the injection volume with numeric keypad, in µL and press **enter**.The cursor will move to the [runtime] field.



NOTE

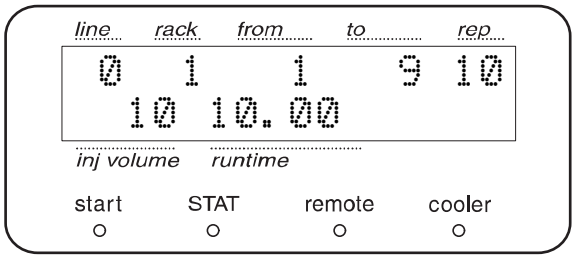
Setting is possible in increments of 0.1 µL from 0.1 to 9.9 µL or in increments of 10 µL from 1 to 50 µL. (20 µL maximum for loop injection method) Set a value smaller than the maximum injection volume.

With the system controller CBM-20A/20Alite or workstation LCsolution/LabSolution, the set value for the maximum volume of injection can be displayed but the setting is not possible.

For the total injection method, see "[MAX INJ. VOLUME]" P. 108

For the loop injection method, see "[VALVE LOOP VOL]" P. 108

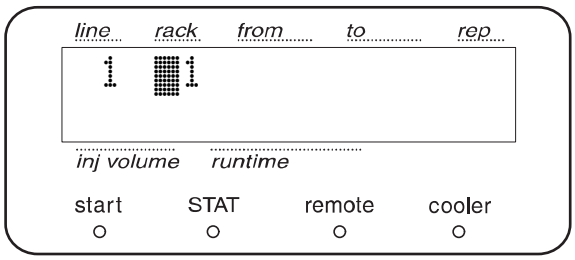
7 Enter the analysis time, in minutes, and press **enter**.



NOTE

Any value between 0.01 and 9999 minutes can be set. Values higher than 100 minutes must be in 1 minute increments.

8 Press **enter**.
The next line is displayed.



9 Repeat steps 1 to 7 and set the next line.

NOTE

- Up to 99 lines can be set in the sample table. (If a repeat injection table is created, up to 99 lines including the number of repetitions can be set.)
- If no further lines are required, press **CE**.

10 Press **CE** to go back to the initial screen.

5.4.2 Creating an Analysis Sequence Table during the Analysis

When the repeat injection table is used, a repeat injection table or an analysis sequence table cannot be changed or added during the analysis.
When the repeat injection table is not used, the parameters can be changed or added from the second line under the current analysis. Under the pause state, the parameters can be changed or added from the next line.

5.4.3 Analysis in Repeat Injection Mode

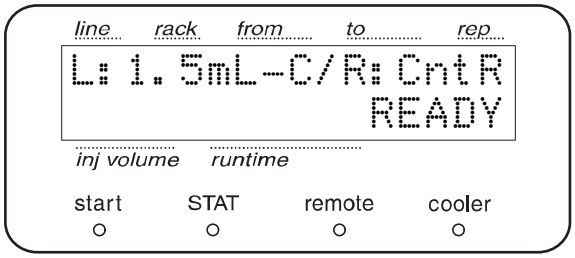
The procedures for sample preparation and configuration of settings are the same as those for standard injection mode. The procedures for performing analysis are the same as those for standard injection mode.

5.5 VP Functions

VP functions support the validation of the instrument by check functions or displaying the instrument information.
There are four groups for VP functions: Product Information, Maintenance Information, Validation Support, and Calibration Support.

5.5.1 Displaying the VP Functions

1 Press **CE**.
The initial screen appears.



2 Press **VP** to select the desired Group.

3 Press **func** until the desired function appears.
∗ To return to the previous screen, press **back**.

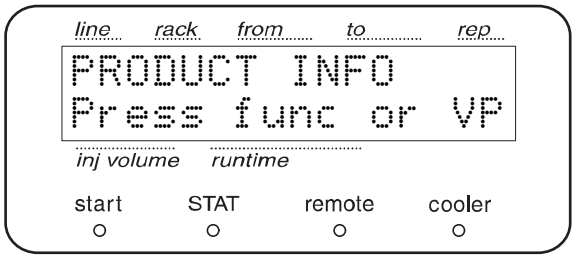
4 Follow the further instructions of the selected function.

5 To select a different VP Function Group, press **VP** repeatedly. To select the desired function, press **func** or **back**.

6 To return to the initial screen, press **CE**.

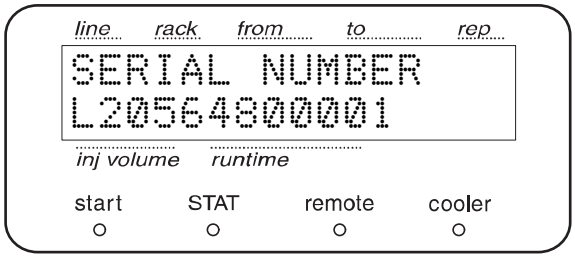
5.5.2 Product Information Group

This group provides the information about the instrument.



■ [SERIAL NUMBER]

Shows the serial number of this instrument.

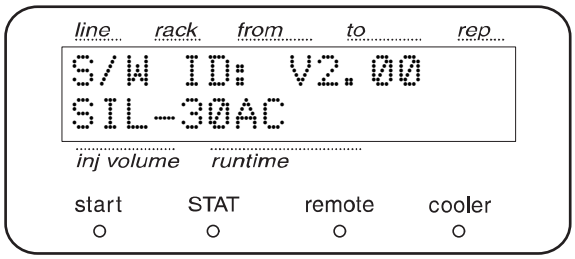


NOTE

The serial number and software version number can only be displayed. They cannot be changed.

■ [S/W ID]

Shows the name of software (same as the model name) and version number.

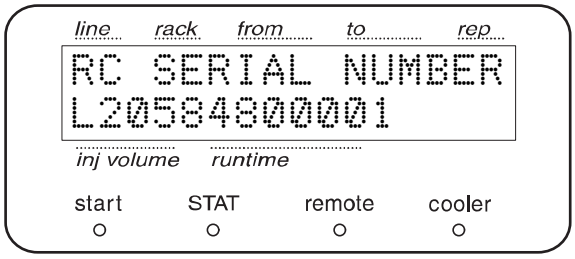


NOTE

The serial number and software version can only be displayed. They cannot be changed.

■ [RC SERIAL NUMBER]

Press **func** again to display the rack changer's serial number.

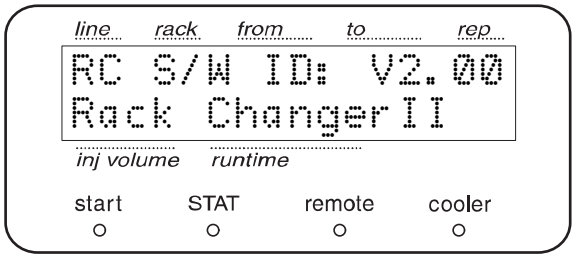


NOTE

Only displayed if a rack changer is connected.
The rack changer's serial number and software version can only be displayed. They cannot be changed.

■ [RC S/W ID]

Press **func** again to display the rack changer's software version.

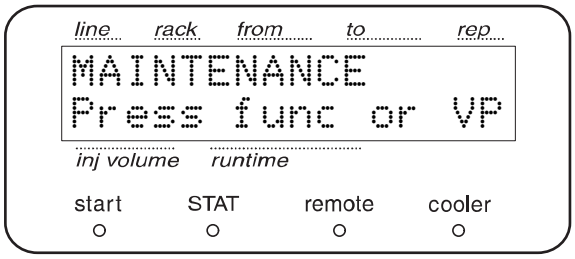


NOTE

Only displayed if a rack changer is connected.
The rack changer's serial number and software version can only be displayed. They cannot be changed.

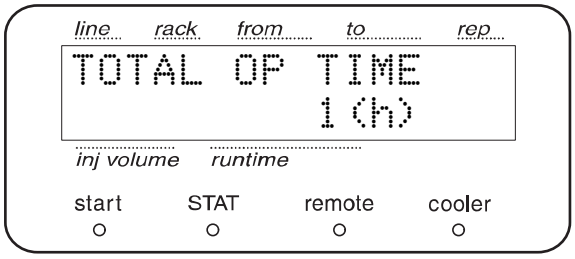
5.5.3 Maintenance Information Group

This group provides the maintenance-related information.



■ [TOTAL OP TIME]

Shows the total operating time of the instrument.



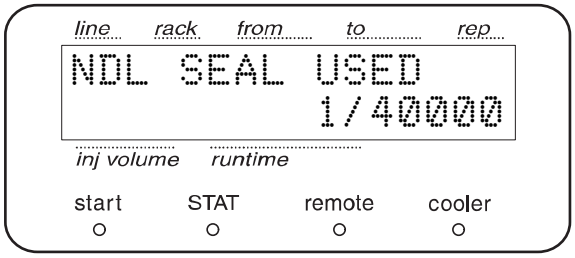
■ [NDL SEAL USED]

Displays the usage frequency and replacement alert value for the needle seal.

NOTE

After replacing the needle seal, reset the counter to [0] by pressing **0** and **enter**.

"7.2 Replacing Needle Seal" P. 229



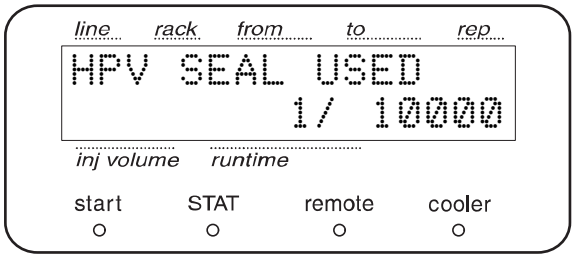
■ [HPV SEAL USED]

Displays the usage frequency and replacement alert value for the high-pressure valve's rotor seal.

NOTE

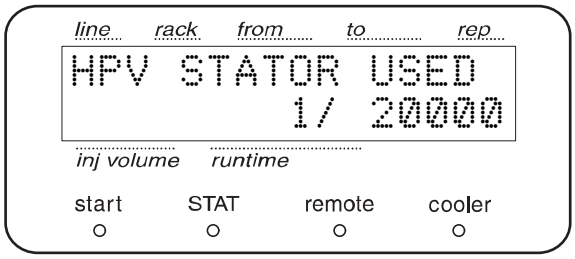
After replacing the rotor seal, reset the counter to [0] by pressing **0** and **enter**.

"7.6 Replacing and Inspecting High-Pressure Valve Rotor and Stator" P. 247



■ [HPV STATOR USED]

Displays the usage frequency and replacement alert value for the high-pressure valve stator.



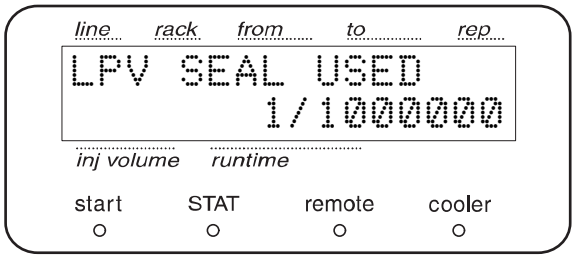
NOTE

After replacing the stator, reset the counter to [0] by pressing 0 and enter.

"7.6 Replacing and Inspecting High-Pressure Valve Rotor and Stator" P. 247

■ [LPV SEAL USED]

Displays the usage frequency and replacement alert value for the low-pressure valve's rotor seal.



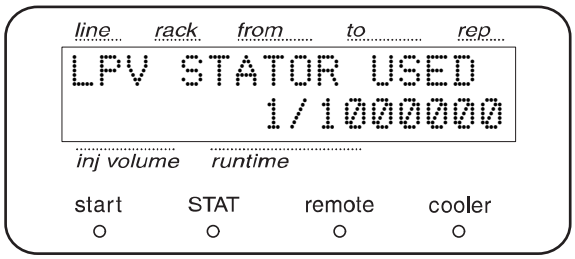
NOTE

After replacing the rotor seal, reset the counter to [0] by pressing 0 and enter.

"7.5 Replacing and Inspecting Low-Pressure Valve Rotor and Stator" P. 242

■ [LPV STATOR USED]

Displays the usage frequency and replacement alert value for the low-pressure valve stator.



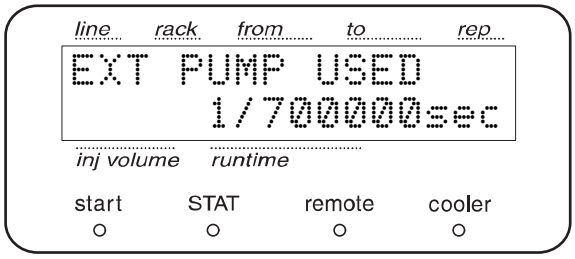
NOTE

After replacing the stator, reset the counter to [0] by pressing 0 and enter.

"7.5 Replacing and Inspecting Low-Pressure Valve Rotor and Stator" P. 242

■ [EXT PUMP USED]

Displays the rinsing pump's usage frequency and replacement alert value.



NOTE

After replacing the rinsing pump, reset the counter to [0] by pressing 0 and enter.

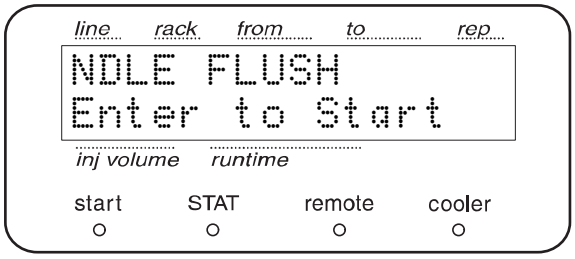
■ [NDLE FLUSH]

NOTE

This item is displayed when total injection method (INJECTION TYPE: 0) is selected.

The screen on the right can be used to rinse the inside of the needle with mobile phase when it is clogged.

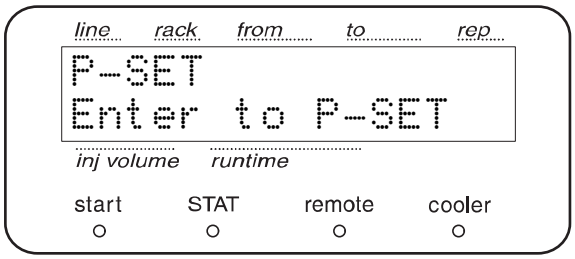
"7.13.1 Rinsing Needle and Sample Loop" P. 268



■ [P-SET]

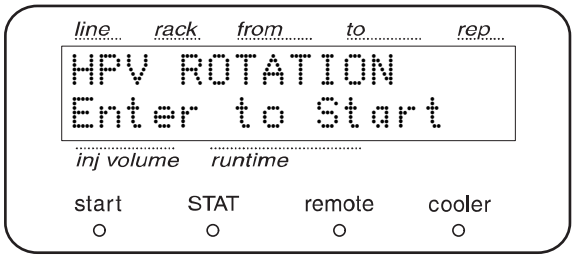
This screen can be used to move the plunger position when replacing the plunger seal and plunger.

"7.3 Replacing Plunger Seal" P. 233, "7.4 Replacing and Inspecting Measuring Pump Plunger" P. 238



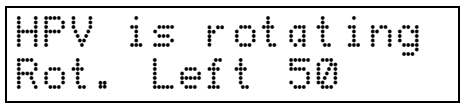
■ [HPV ROTATION]

This screen is used when replacing the high-pressure valve's rotor seal. When this menu is executed, the high-pressure valve rotates 50 times automatically.
It takes approx. 20 minutes to finish the operation.



1 Remove the column and, with the pump connected to the plumbing using, for example, a coupling, pump 2-propanol or methanol at a rate of 2 mL/min.

2 Press **enter**.
The high-pressure valve rotates every several seconds 60° at a time automatically. The left number of rotation is displayed in the second line.



NOTE

If the operation starts once, it cannot be canceled until the 50 times-rotation has been carried out.

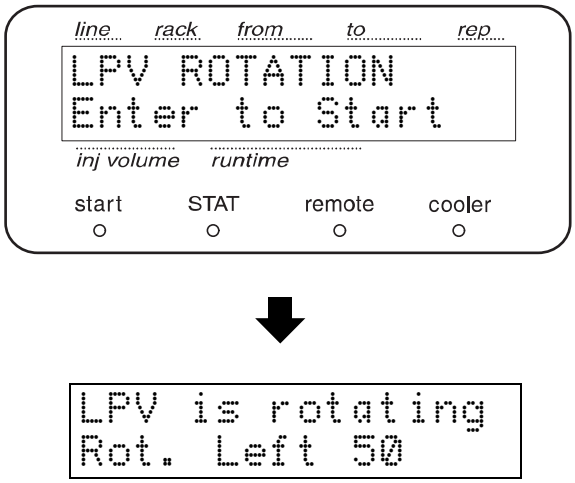
■ [LPV ROTATION]

This screen is used when replacing the low-pressure valve's rotor seal. When this menu is executed, the low-pressure valve rotates 50 times automatically.
It takes approx. 20 minutes to finish the operation.

Press (enter) to make the low-pressure valve to rotate every several seconds 60° at a time automatically. The left number of rotation is displayed in the second line.

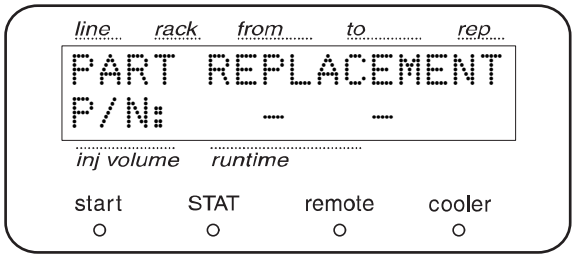
NOTE

If the operation starts once, it cannot be canceled until the 50 times-rotation has been carried out.



■ [PART REPLACEMENT]

Enters the replaced part No.
The part No. is recorded in the maintenance log.



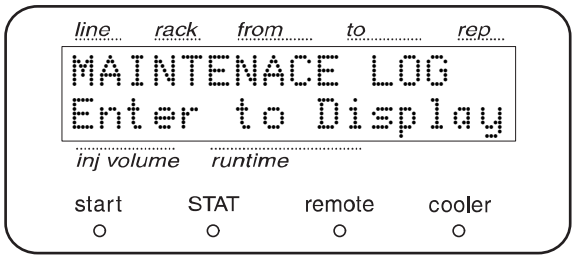
■ [MAINTENANCE LOG]

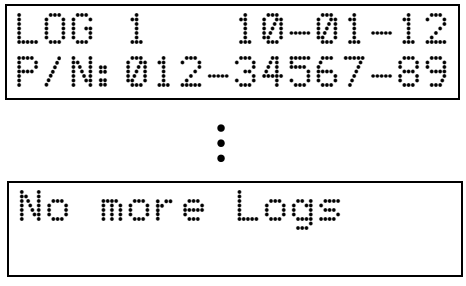
Shows the maintenance log, which contains the most recent parts replacement records (part No. and date) (up to 10).

Press (enter) repeatedly to show Log1 to Log10 in sequence, and return to the title screen.

In the example on the right, the Log1 entry indicates that a part No. S012-34567-89 was replaced on Jan. 12, 2010.

If less than 20 logs are recorded, the screen displays the message as shown on the right.
Press (CE) to return to the title screen.

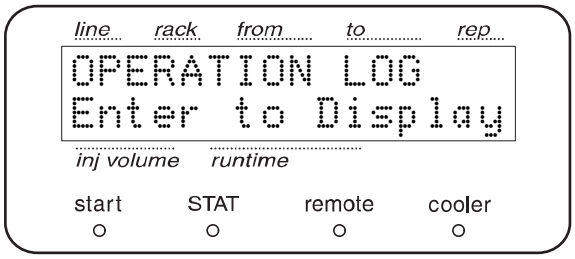




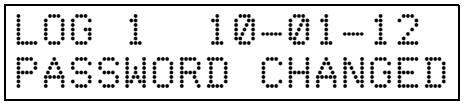
■ [OPERATION LOG]

Shows the operation log, which contains the most recent password settings, parameter initialization, etc. (up to 10).

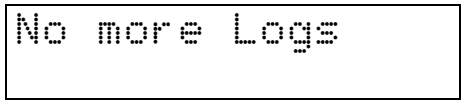
Press **enter** repeatedly to show Log1 to Log10 in sequence, and return to the title screen.



In the example on the right, the Log1 entry indicates that a password setting was made on Jan. 12, 2010.



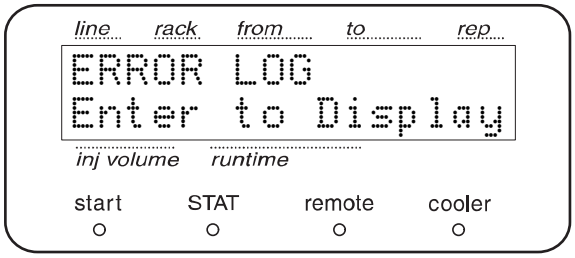
If less than 10 logs are recorded, the screen displays the message as shown on the right.
Press **CE** to return to the title screen.



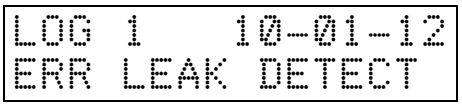
■ [ERROR LOG]

Shows the error log, which contains the most recent errors (up to 10) with their dates.

Press **enter** repeatedly to show Log1 to Log10 in sequence, and return to the title screen.



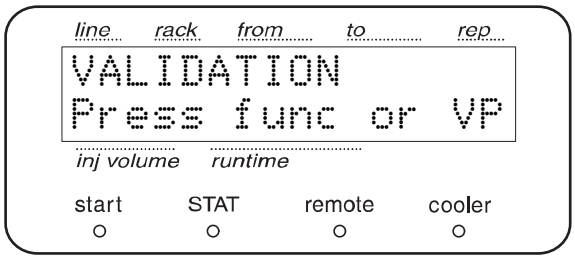
In the example on the right, the Log1 entry indicates that a leak was detected on Jan. 12, 2010.



If less than 10 logs are recorded, the screen displays the message as shown on the right.
Press **CE** to return to the title screen.

5.5.4 Validation Support Group

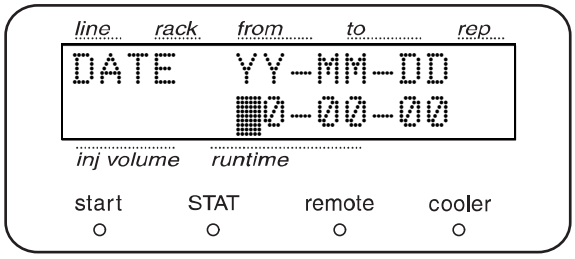
This group checks whether the instrument is running correctly.



■ [DATE]

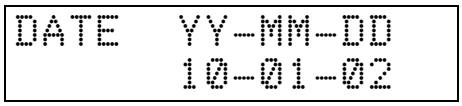
Shows/enters the date.
This value returns to "00-00-00" when the power is turned off. When the instrument is controlled by the system controller, the date is transmitted at the connection.
Example: Setting January 2, 2010

Date



1 Use numeric keypad to first set the year, then the month, then the day. For the year, enter the year of the decade only. For each item, be sure to enter 2 digits (i.e. enter a zero in the tenths column if necessary.).

Input

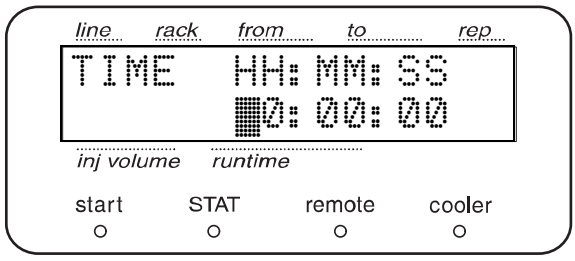


2 When setting is complete, press **enter**.

■ [TIME]

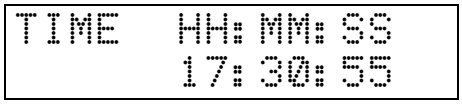
Shows/enters the time.
This value returns to "00:00:00" when the power is turned off. When the instrument is controlled by the system controller, the time is transmitted at the connection.
Example: Setting 5:30:55 p.m.

Time



1 Use numeric keypad to first set the hours, then the minutes, then the seconds. The display uses a 24-hour clock. For each item, be sure to enter 2 digits (i.e. enter a zero in the tenths column if necessary.).

Input

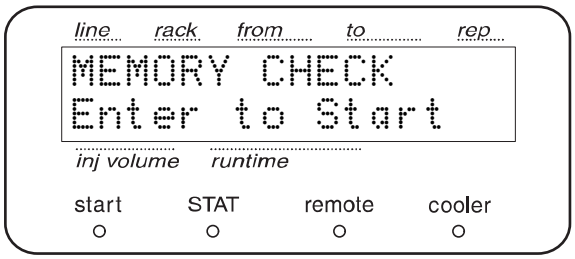


2 When setting is complete, press **enter**.

■ [MEMORY CHECK]

Runs the memory check on ROM and RAM.

Press **enter** to start.
Results are shown when checking is completed.



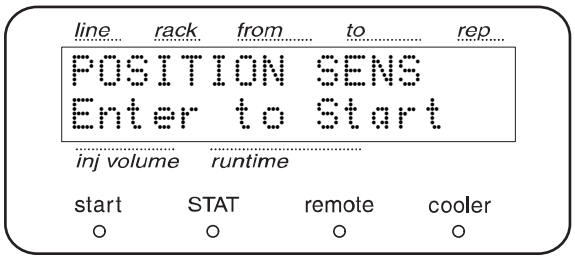
NOTE

If a rack changer is connected, the rack changer's memory check result is also displayed.

Result

■ [POSITION SENS]

When **enter** is pressed, the sensors inside the autosampler (i.e., for the needle's X direction, the rack's Y direction, the needle's Z direction, the high-pressure valve, the low-pressure valve, and the pump) perform automatic operation checks.



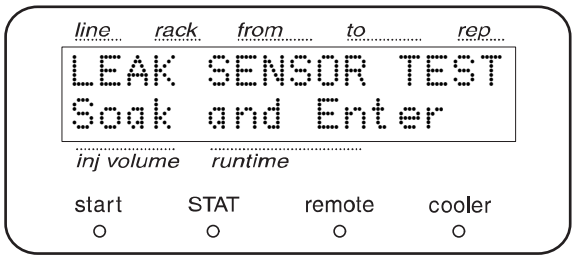
During the checks, the message on the right is displayed.

The results of the checks are displayed upon completion. If there is an error, an error message for the place with the error is displayed.

■ [LEAK SENSOR TEST]

Carries on the operation test for leak sensor.

1 Use a syringe filled with water to wet the thermosensor at the bottom of the leak sensor.



2 Wait about 10 seconds. Then press (enter). If the sensor detects a leakage, [GOOD] will be shown. If not, [NO GOOD] will be shown.

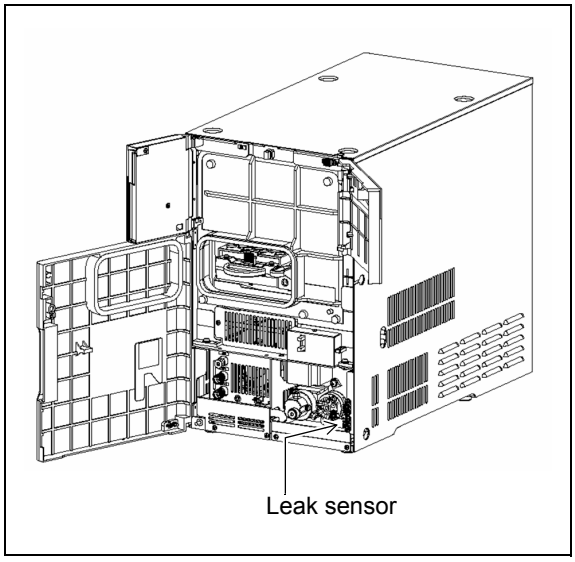


Fig. 5.6

3 Carefully wipe the thermosensor body at the lower side of the leak sensor until it is completely dry.

> **⚠ CAUTION**
>
> The sensor must be dry when exiting this screen, or leak sensor errors may occur.

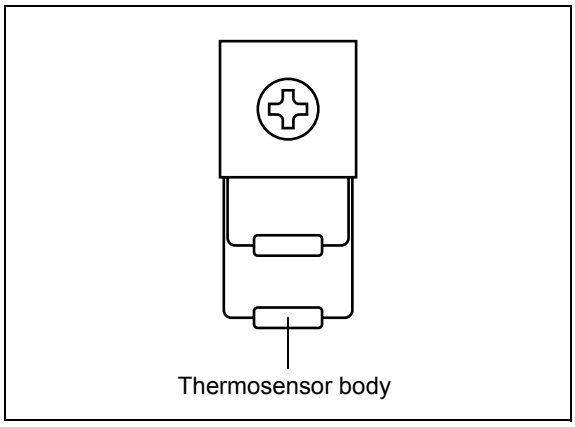
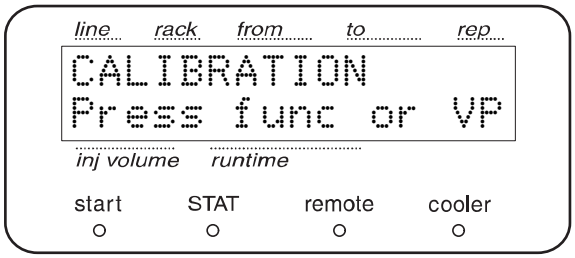


Fig. 5.7

4 Press (func) to return to the [DATE] screen.
Press (back) to return to the previous screen.
Press (CE) to return to the initial screen.

5.5.5 Calibration Support Group

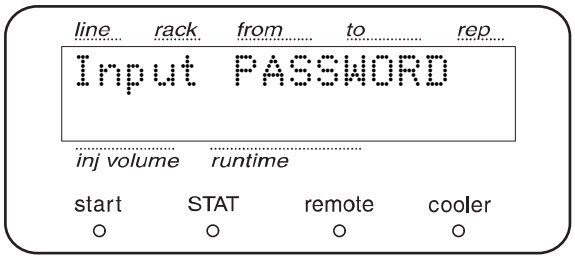
This group calibrates the instrument.



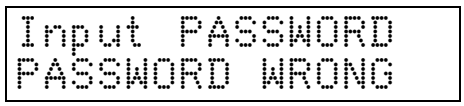
■ [Input PASSWORD]

Password should be registered by a system manager.
Press (**func**), input five numbers and press (**enter**).

∗ Be sure to input five numbers. The default password is [00000].
If the password is input correctly, [ADJUST MTP] function (subsequent function) appears.



If the password is not input correctly, the subsequent functions cannot be accessed.



■ [ADJUST MTP]

NOTE

- When using the microtiter plate for the first time or changing its type, the needle position against the well needs to be adjusted.
- For details about needle position adjustment with rack-changer racks, refer to "4.6 Adjusting Plate Positions" in the Rack Changer Instruction Manual.
- When the tip of the needle is less visible, use the jig, rack teaching provided with the instrument.
  Install and remove the jig, rack teaching as described below.

(1) Installation of the jig, rack teaching

1 Press ↑ on the initial screen. The [Z HOME] screen will be displayed.

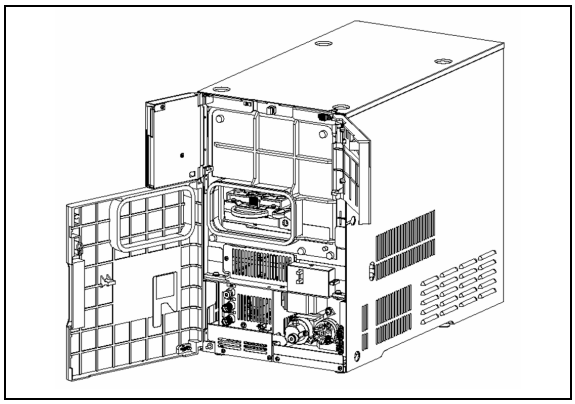


Fig. 5.8

2 Press **enter**. The needle rises to the highest position and then moves to the center of the instrument.

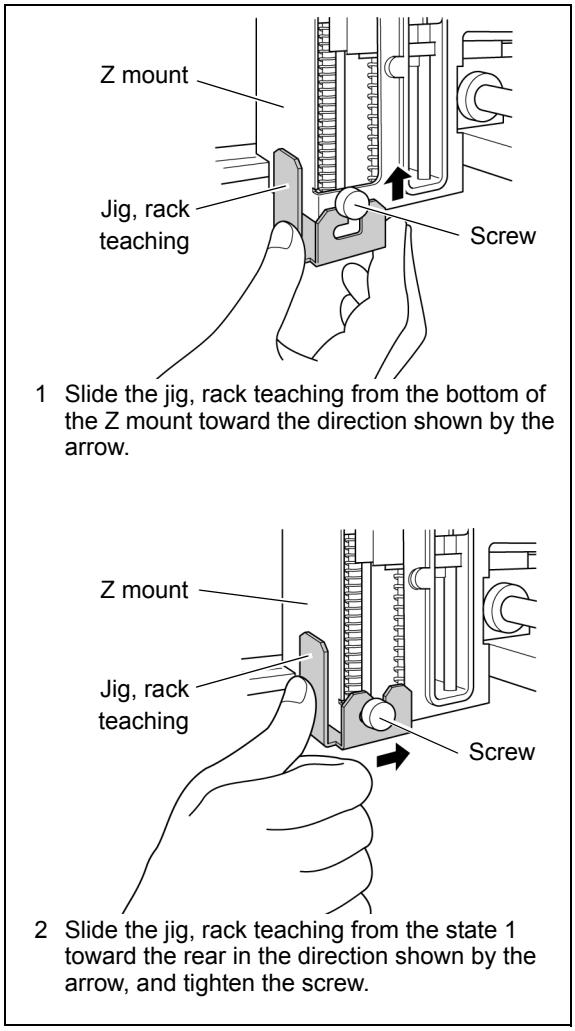
3 Turn OFF the power switch.

4 Loosen the screws (5 positions), slide panel F to the right, and pull it toward you until it is removed.

5 Loosen one screw at the bottom right of the Z mount, and follow steps ① and ② shown in Fig. 5.9 to attach the jig, rack teaching to the Z mount.

NOTE

Install the jig, rack teaching so that it will come into close contact with the Z mount cover.



1 Slide the jig, rack teaching from the bottom of the Z mount toward the direction shown by the arrow.

2 Slide the jig, rack teaching from the state 1 toward the rear in the direction shown by the arrow, and tighten the screw.

Fig. 5.9

(2) Removal of the jig, rack teaching

1 Press ↑ on the initial screen.
The [Z HOME] screen will be displayed.

2 Press **enter**. The needle rises to the highest position and then moves to the center of the instrument.

3 Turn OFF the power switch.
Open the door, loosen one screw at the bottom right of the Z mount, and remove the jig, rack teaching from the Z mount.

NOTE

Be careful not to lose the jig, rack teaching after it is removed.

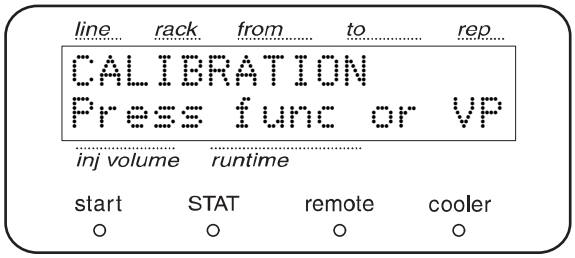
4 Attach panel F so that the autosampler is sealed off, and close the door.

> **⚠ WARNING**
>
> During this adjustment, operation will not stop when the door is open. Do not reach into the auto injector during this procedure; you could be injured.

- Adjust the needle position against the well on the microtiter plate as described below.

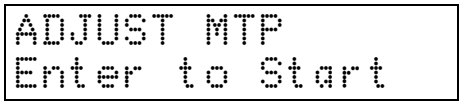
1 Place the microtiter plate on the rack.

2 Press **VP** in the initial screen repeatedly until the screen on the right is displayed.



3 Input the password correctly.

4 Press **enter**.
The [ADJUST MTP] screen is displayed.



5 Press **enter**.
The needle moves close to hole A1 in the near-side microtiter plate (the left position of the rack) and stops.

6 Using the arrow keys, adjust the right or left and front or rear position so that the needle is lowered into the center of hole A1.
(The vertical direction is set with the [NEEDLE STROKE].)

☞ "[NEEDLE STROKE]" P. 85

Arrow key	Direction of needle movement
←	The needle moves 0.1 mm to the left.
→	The needle moves 0.1 mm to the right.
↑	The needle moves all the way up.
↓	The needle moves down 4 mm the first time. The needle moves down 0.2 mm afterwards.
func	The needle moves forwards 0.1 mm.
back	The needle moves backwards 0.1 mm.
ins	The needle moves down 4 mm at first. Then the needle moves down 1 mm.

7 Press (enter).
The position is determined, the finely adjusted position is stored in memory, and the needle moves close to hole A1 of the rear-side microtiter plate (the deep, left position of the rack) and stops.

NOTE

To stop the adjustment partway through, press (CE).
The needle position is not set newly.

8 Move the needle laterally in the way described in step 6. Using the arrow keys, move it to the position where the tip of the needle is aligned with the center of hole A1 of the rear-side microtiter plate.

9 Press (enter).
The position is decided, its fine adjustment position is recorded, and the needle moves forward. The needle moves to H1 (with a 96-well rack) or P1 (with 384-well rack), and stops.

NOTE

To stop the adjustment partway through, press (CE).
The needle position is not set newly.

10 Move the as step 6.
Using (func) and (back), move the edge of needle so that it comes to the center of hole.

11 Press (enter) to confirm the position. The fine adjustment point will be memorized.
The fine adjustment finishes, and the needle moves to the injection port.

12 Press (func) to display the next screen.
Press (back) to return to the previous screen.
Press (CE) to return to the initial screen.

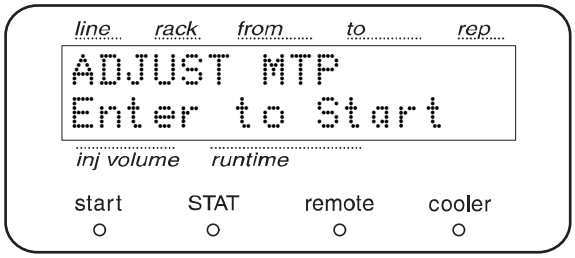
NOTE

- The data set with the teaching function is stored in memory for each type of rack (i.e., rack for microtiter plates or deep-well plates, or changer rack).
- When the jig, rack teaching has been used, remove the jig from the Z mount.

■ [ERASE MTP ADJ]

This screen is used to bring the microtiter plate position data set in [ADJUST MTP] (microtiter plate position fine adjustment) to the initial state.

1 Input the password correctly and press **enter**.
The [ADJUST MTP] screen will be displayed.

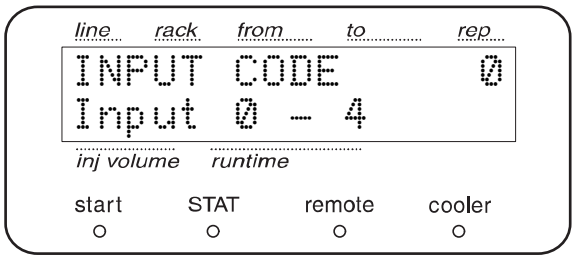


2 Press **func**.
The [ERASE MTP ADJ] screen will be displayed.

3 Press **enter**.
Select the data to be erased, and press **enter**.

NOTE

- When rack-changer racks are used:
  Position data can be reset to the default state for each of stack codes (set value: 0 to 4).
  When step 3 is completed, the screen at the right is displayed. Input the stack code of which position data is to be deleted.



4 Press **func** to display the next screen.
Press **back** to return to the previous screen.
Press **CE** to return to the initial screen.

NOTE

When using a microtiter-plate rack, it is necessary to readjust the position of the microtiter-plate rack with the [ADJUST MTP] screen.
"[ADJUST MTP]" P. 137

- When executing this function, the data of the set microtiter plate is initialized.
  (The data for other microtiter plates and deep-well plates rack is not erased.)
- Before erasing all data for microtiter plates, deep-well plates, and changer racks, remove the rack from the autosampler.

■ [ASP FACTOR]

This screen is used for compensating sample injection precision. There are two compensation methods available: automatically obtaining a factor using the gravimetric method or directly specifying a factor.

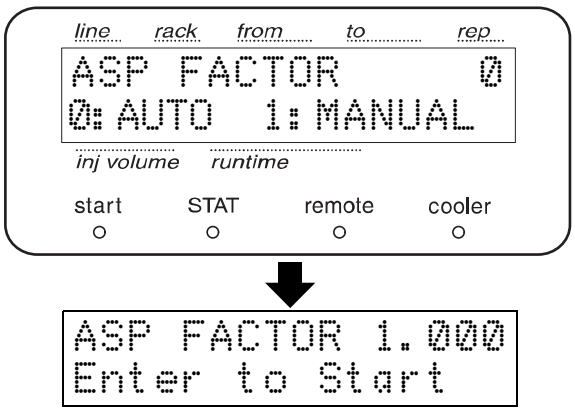
NOTE

Before compensating, the measuring flow line must be purged. If the measuring flow line is not degassed enough, may not aspirate the water correctly.
"Purging Air Bubbles" P. 43

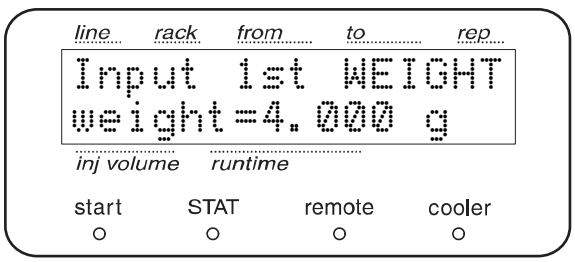
1) [AUTO] (This factor is calculated automatically.)

Determines the sample aspiration volume compensation factor. The factor is calculated as follows: A 1.5 mL vial is filled with at least 1 mL of distilled water, and ten 50 µL volumes of the water are aspirated consecutively from the vial. The vial is then weighed (by the operator) to determine the decrease in weight of the water. This weight is converted into a volume value and used as the compensation factor.

1 Press **0**, **enter** to start the procedure.



2 Using a calibrated scale, weigh a vial filled with distilled water. Then enter the weight (mg) with numeric keypad and press **enter**.

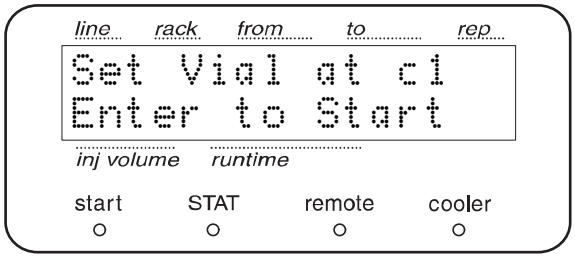


3 Remove the control vial rack.

4 Set a sample vial that contains distilled water in the No.1 position on the control vial rack, and insert the rack into the instrument .

5 Close the door.

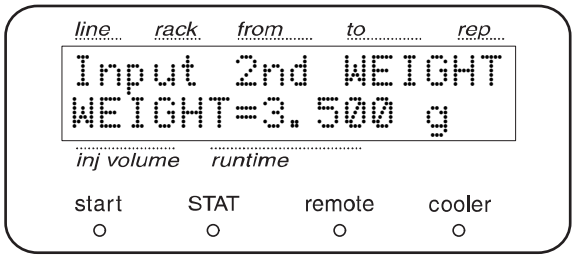
6 Press **enter**.
The sequence of 10 aspirations will begin.



NOTE

- During the aspirations, the operation cannot be canceled.
- The distilled water aspirated from the vial is drained into the rinsing port.

7 When all the aspirations have been completed, the message on the right appears. Open the door and remove the vial.



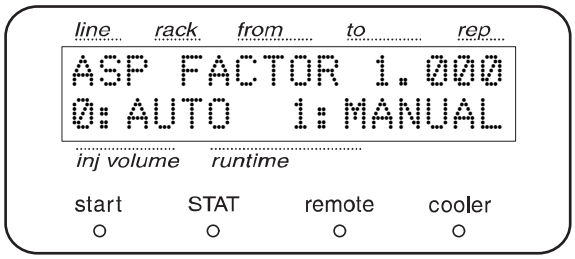
8 Weigh the vial again.

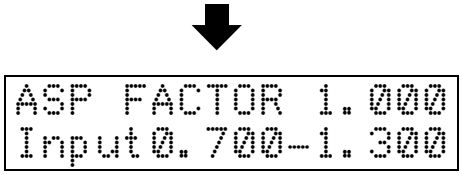
9 Then enter the weight (mg), and press **enter**.
The compensation factor is displayed on the screen.

10 Press **enter**.
The [ASP FACTOR] screen will be displayed.

2) [MANUAL] (Enter the compensation factor directly.)

1 Press **1**, **enter** to start the procedure.





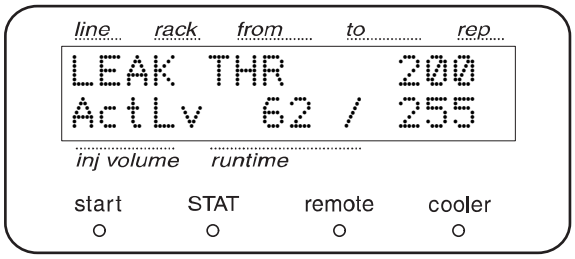
2 Set the compensation factor and press **enter**.
Min: 0.700
Max: 1.300

■ [LEAK THR]

Sets the level (threshold value) at which the leak sensor is actuated. Use numeric keypad to enter the level, and press (enter). The setting range is [0-255].

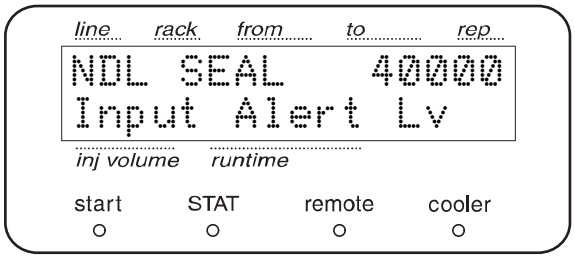
NOTE

The [ActLv] in the bottom line of the display shows the leak sensor's current (actual) value. If this value exceeds the value set for [LEAK THR], the sensor detects a leak.



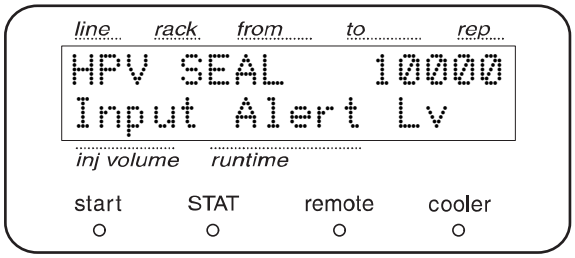
■ [NDL SEAL]

Changes the needle seal replacement alert level (i.e.total number of injections before an alert is issued), indicating that the needle seal needs to be replaced.
Use numeric keypad to enter the new value and press (enter).
Default value: 40000 (times)



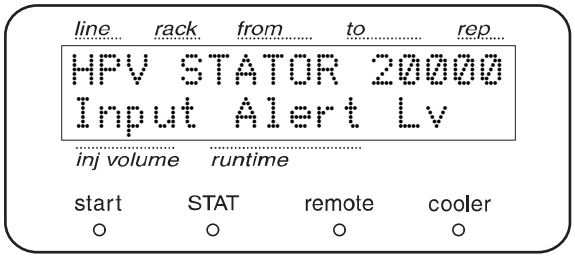
■ [HPV SEAL]

Changes the high-pressure valve rotor seal replacement alert level (i.e.total number of injections before an alert is issued), indicating that the needle seal needs to be replaced.
Use numeric keypad to enter the new value and press (enter).
Default value: 10000 (times)



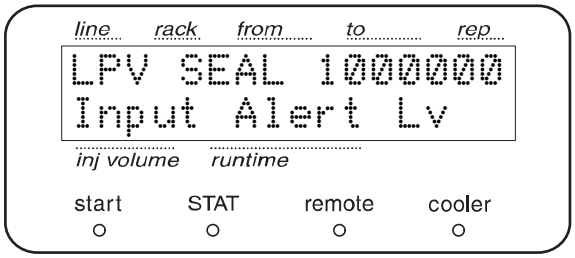
■ [HPV STATOR]

Changes the high-pressure valve stator replacement alert value.
Use the numeric keypad to enter a new value and press (enter).
Default value: 20000 (times)



■ [LPV SEAL]

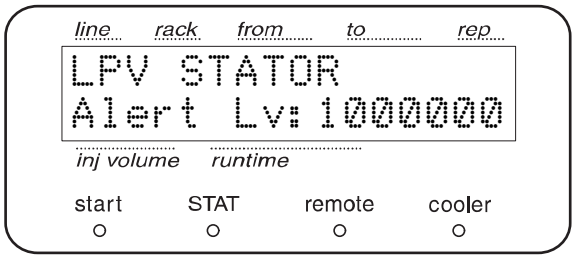
Changes the low-pressure valve seal replacement alert level (i.e.total number of injections before an alert is issued), indicating that the needle seal needs to be replaced.
Use numeric keypad to enter the new value and press (enter).
Default value: 1000000 (times)



■ [LPV STATOR]

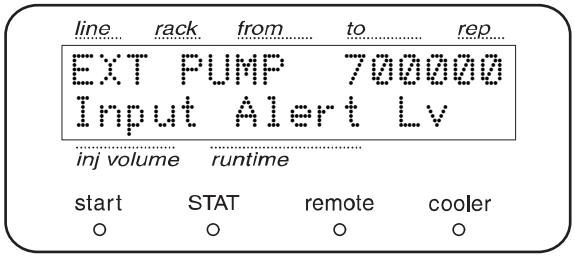
Changes the low-pressure valve stator replacement alert value.

Enter the replacement alert value using the numeric keypad, and press **enter**.
Default value: 1000000 (times)



■ [EXT PUMP]

Changes the rinsing pump (optional) seal replacement alert level (i.e.total number of injections before an alert is issued), indicating that the needle seal needs to be replaced.
Use numeric keypad to enter the new value and press **enter**.
Default value: 700000 (second)



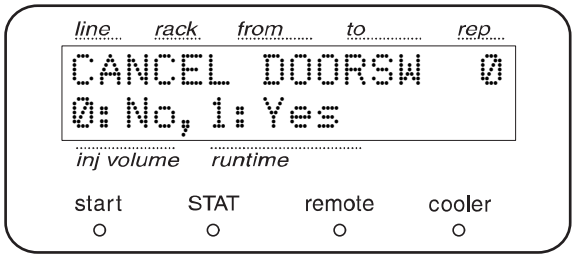
■ [CANCEL DOORSW]

This function cancels open/close door detection.
When this function is enabled (detection is off), opening the door will not stop the injection operation.

> **⚠ CAUTION**
>
> When [1] is set, the autosampler will perform injection even if the door is open or the panel F is removed. Do not change the set value unless it is absolutely required.
> Doing so may cause injuries from the needle.

Enter the set value and press **enter**.

Set value	Function
0	Automatic door open/close detection is enabled. (default setting)
1	Automatic door open/close detection is disabled.

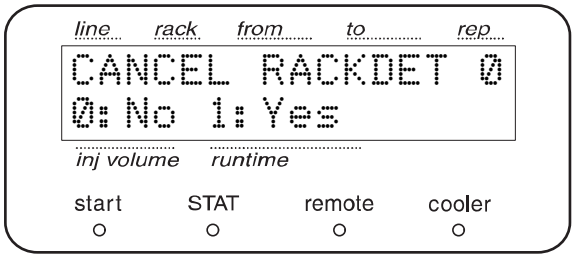


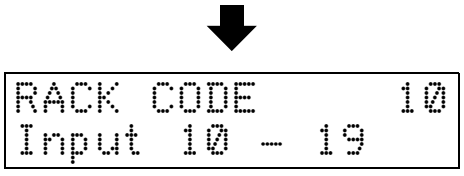
⚠ CAUTION

Never reach into the autosampler in operation with automatic door open/close detection disabled. Doing so may cause injuries from the needle.

■ [CANCEL RACKDET]

If the automatic rack position sensor is not working properly (i.e., due to a broken sensor or sensor detection block), the automatic rack position detection function can be disabled.
Select a numerical key.
Enter the set value and press **enter**.





Set value	Function
0	Automatic rack position detection is enabled. (default setting)
1	Automatic rack position detection is disabled.

NOTE

When [1] is set, enter the rack code number and confirm it with [Enter].

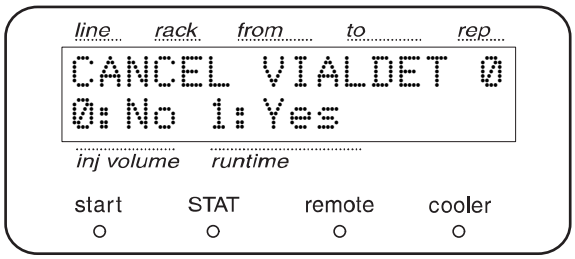
Rack type	Input value
Sample vial rack for 1 mL	12
1.5 mL sample vial rack, 1.5 mL sample vial cooling rack (105 vials)	10
1.5 mL sample vial cooling rack (70 vials)	11
Sample vial rack for 4 mL	13
Microtiter plate rack	14
Deep-well plate rack	16
Rack-changer rack	18
Not used (reserved)	19

⚠ CAUTION

If automatic rack detection is disabled ([1]), the needle may descend below the bottom of the vial or well if another rack is used for analysis. This could result in clogging or bending the needle. Except when adjusting the rack position ("[ADJUST RACK]" P. 150) or when an emergency arises, do not change the set value.

■ [CANCEL VIALDET]

Use numerical keypad to enter the value, and press **enter**.

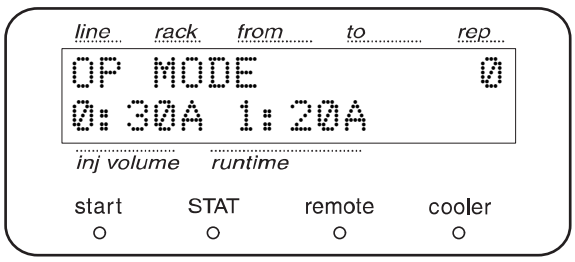


Set value	Function
0	Sample vial automatic detection is enabled. (default setting)
1	Sample vial automatic detection is disabled.

■ [OP MODE]

Select the operation mode according to the type of connected system controller.

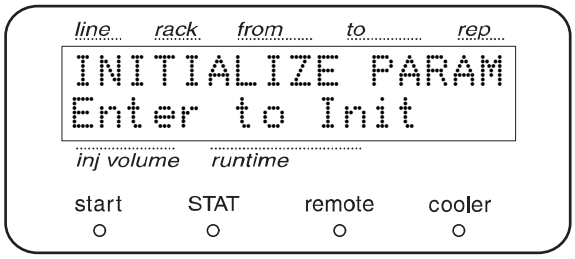
Enter the number and press **enter**.
After setting, turn off the power, and then turn it on again.



Value	System controller
0	CBM-20A/20Alite (Ver. 2.00 and later) The instrument is recognized as SIL-30AC.
1	CBM-20A/20Alite (Ver. 1.30 and earlier) The instrument is recognized as SIL-20AC.

■ [INITIALIZE PARAM]

Initializes the parameters and deletes the time programs.



Press **enter** to return to the default value and to delete the time programs.

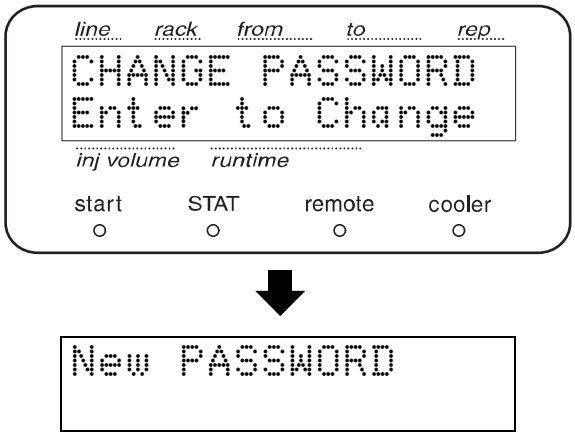
NOTE

The [TOTAL OP TIME], [NDL SEAL USED], and [HPV SEAL] valves are not erased.

☞ "[TOTAL OP TIME]" P. 129, "[NDL SEAL USED]" P. 129, "[HPV SEAL USED]" P. 129

■ [CHANGE PASSWORD]

Changes the password set.



1 Press **enter**.
The input screen appears.

2 Input a new password and press **enter**.
The password must consist of five digits.

3 To confirm, input the same password again.

4 The new password is enabled.

NOTE

If a wrong password is entered, you are returned to the password input screen.
Press **enter** and return to step 2.

■ [ADJUST RACK]

⚠WARNING

During this adjustment, operation will not stop when the door is open. Do not reach into the autosampler during this procedure; Doing so may cause injuries from the needle.

NOTE

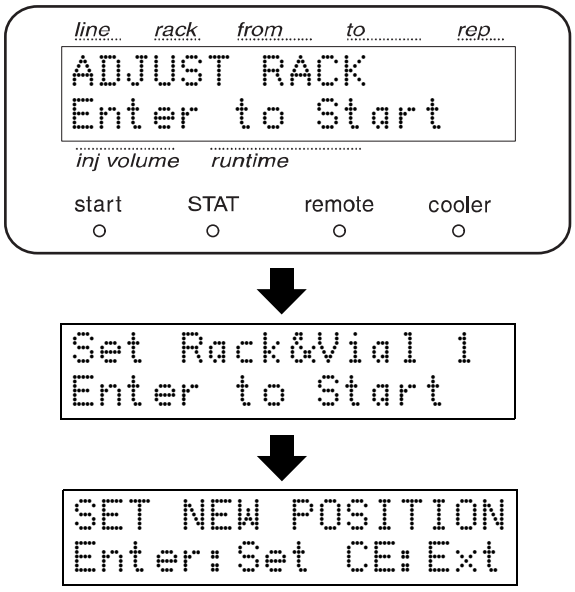
In case of using the cooling rack for 1.5 mL, after removing the rack cover, set the sample rack into autosampler.
When using the 1.5 mL sample vial cooling rack (70 vials), the rack code cannot be recognized automatically. Use the function described in "[CANCEL RACKDET]" P. 147 and enter rack code "11".
After adjusting, set the [0:No] parameter to automatic detection.

1 Press **enter**.
Set a sample vial in position 1 of the rack used (1.5 mL, 1 mL, or 4 mL rack). Do not attach a septum to the sample vial.

2 Press **enter**.
The needle stops above position 1 of the rack.

3 Using the arrow keys, adjust the lateral and longitudinal positions so that the needle is lowered into the center of the sample vial.
∗Adjustment of the vertical position is unnecessary.

Arrow key	Direction of needle movement
←	The needle moves 0.1 mm to the left.
→	The needle moves 0.1 mm to the right.
↑	The needle moves all the way up.
↓	The needle moves down 4 mm at first. Then the needle moves down 0.2 mm.
func	The needle moves forwards 0.1 mm.
back	The needle moves backwards 0.1 mm.
ins	The needle moves down 4 mm at first. Then the needle moves down 1 mm.



NOTE

When the needle position is less visible, use the jig, rack teaching.
"[ADJUST MTP]" P. 137

4 Press **enter**.
Adjustment is completed and the needle moves to the injection port.

NOTE

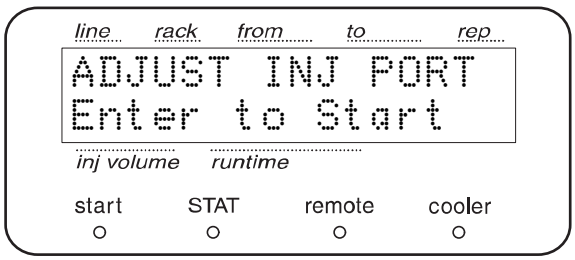
Reattach panel F if it was removed.

■ [ADJUST INJ PORT]

⚠WARNING

During this adjustment, operation will not stop when the door is open. Do not reach into the autosampler during this procedure; Doing so may cause injuries from the needle.

1 Press **enter**.
The needle stops at the front central position of the rack.
Loosen the screws at 5 locations on the front of the autosampler by hand and remove panel F.



2 Loosen the screws in the top part of the Z mount on the left and right and in the bottom part on the right by hand. Pull the cover forwards to remove it.
Press **enter**.

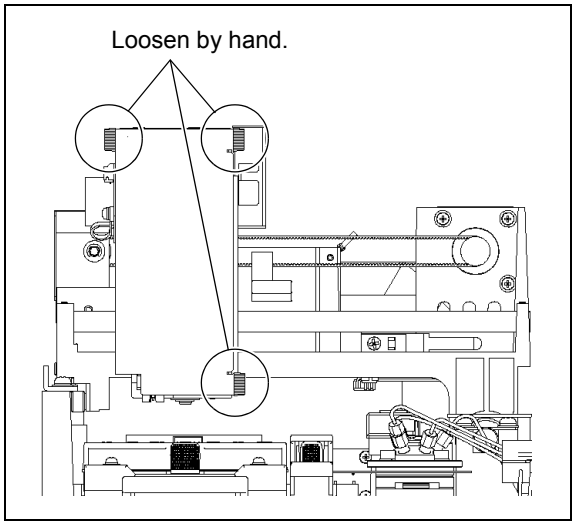
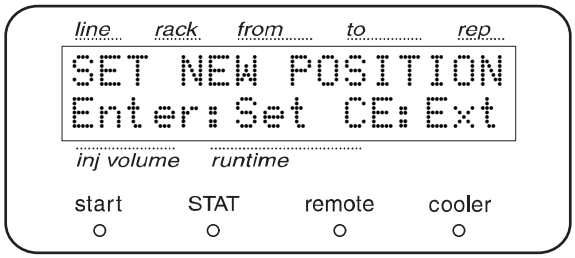


Fig. 5.10

3 Use the arrow keys to adjust the position so that the tip of the needle is at the center of the upper side of the injection port.

Arrow key	Direction of needle movement
←	The needle moves 0.1 mm to the left.
→	The needle moves 0.1 mm to the right.
↑	The needle moves all the way up.
↓	The needle moves down 0.1 mm or approx. 5 mm (only when the needle is at the uppermost part).
func	The needle moves forwards 0.1 mm.
back	The needle moves backwards 0.1 mm.
ins	The needle moves down 1 mm (or approx. 5 mm only when the needle is at the uppermost part).



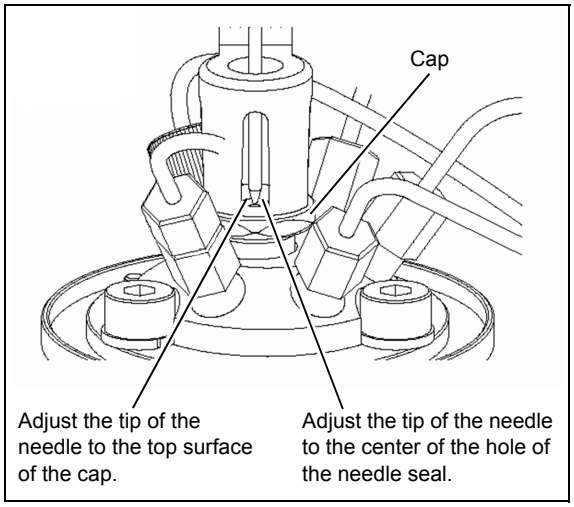


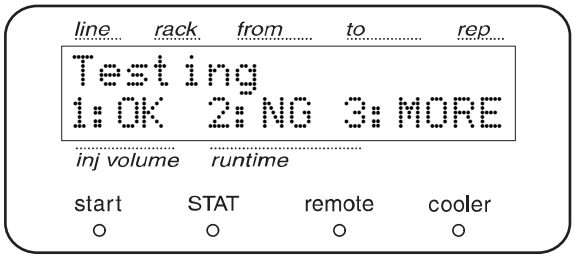
Fig. 5.11

4 Press **enter**.
In order to check that the position has been entered correctly, the tip of the needle moves up and down twice approximately 2 mm at the position where the seal is attached. Check that the needle is lowered smoothly into the injection port as Fig. 5.13.

Set value	Function
1	The needle position is stored in memory.
2	Needle adjustment is performed again.
3	The needle position is checked again.

NOTE

If the needle tip is not aligned with the position where the needle seal is attached, carryover or leaks may be caused.



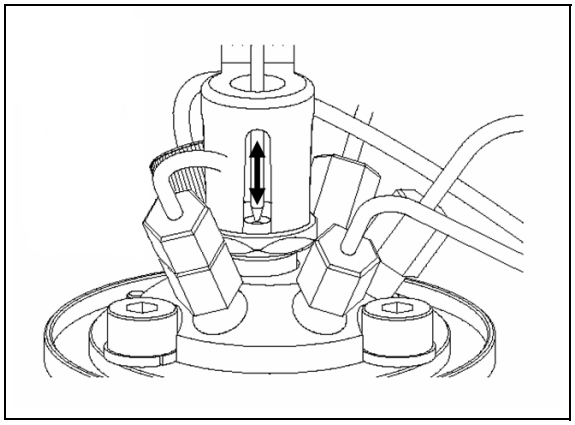


Fig. 5.12

5 Enter the set value [1].
The position is determined, the finely adjusted position is stored in memory.
The needle moves to the injection position.

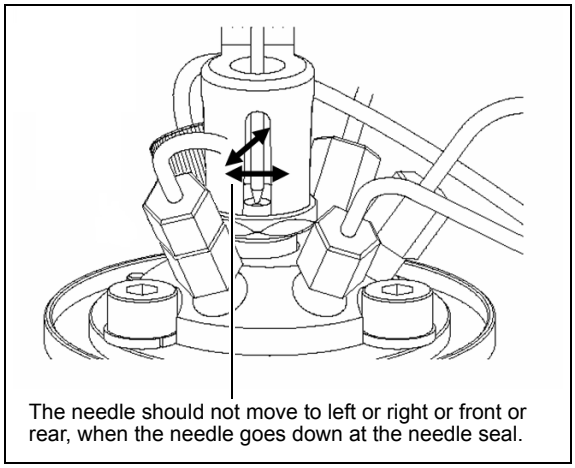
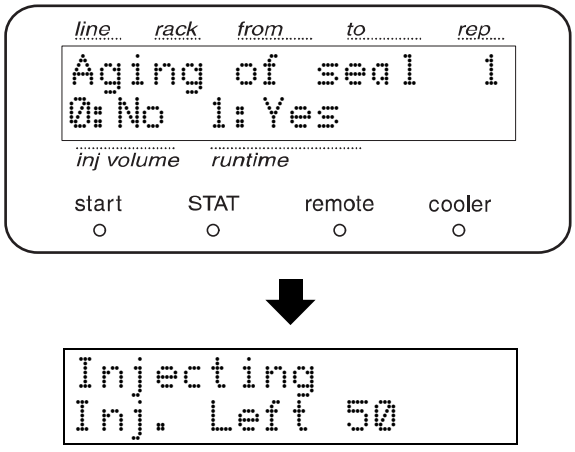


Fig. 5.13

6 The screen is displayed as the right drawing.
If the needle or needle seal has been changed, the seal surface must be aged so that liquid does not leak.
Select [Yes] in the screen on the right.
The needle moves up and down 50 times.
* If the needle or needle seal has not been replaced, aging of the seal surface is not necessary. Select [No].



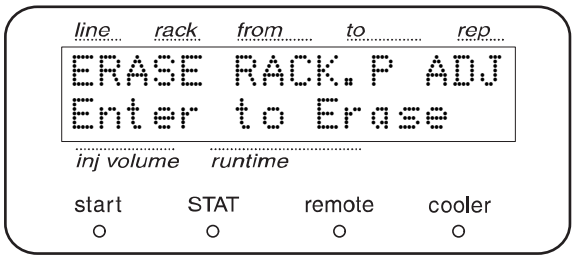
7 Select the [Z HOME] screen, and press (enter).
After installing the cover, press (enter) again.

8 Reinstall the panel F and fixed with screws.

■ [ERASE RACK.P ADJ]

Use this function to initialize the rack position data.

Press **enter**.
The rack position data is erased.



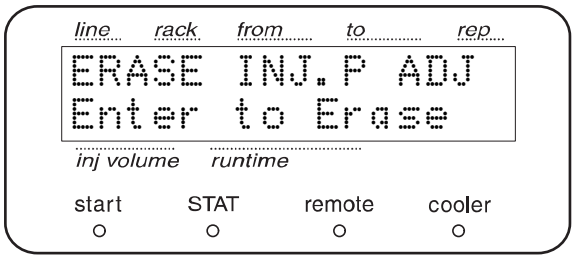
NOTE

When data has been erased, the rack position must be readjusted in the [ADJUST RACK] screen.
☞ "[ADJUST RACK]" P. 150

■ [ERASE INJ.P ADJ]

Use this function before replacing the needle or needle seal.

Press **enter**.
The injection port position data is erased.



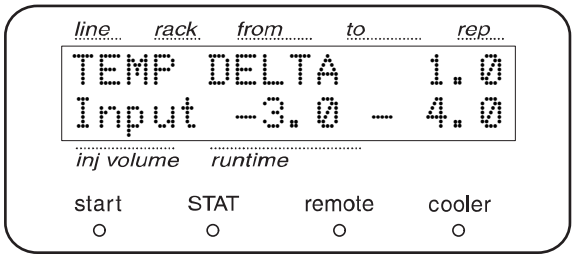
NOTE

When data has been erased, the injection port position must be readjusted in the [ADJUST INJ.P] screen.
☞ "[ADJUST INJ PORT]" P. 151

■ [TEMP DELTA]

Corrects the sample cooler temperature.
Enter the correction value (the difference from the true temperature) using the numeric keypad.

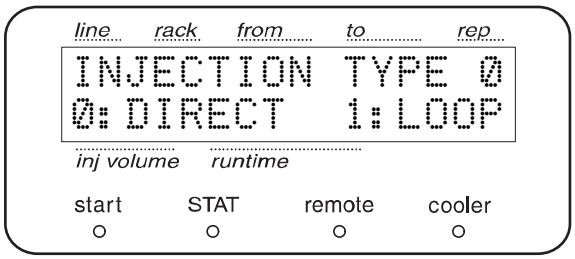
Set range	Step
-3 - 4 °C	0.1 °C



■ [INJECTION TYPE]

Select either total injection method or loop injection method. Enter the set value using the numeric keypad and press **enter**.

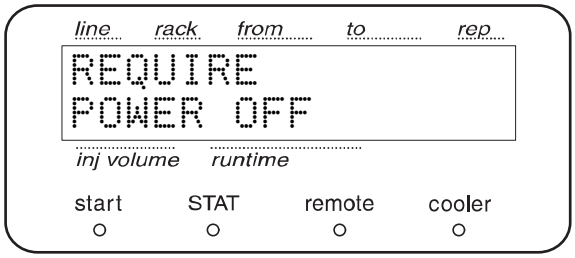
Set value	Function
0	Sample injection with total injection method (default)
1	Sample injection with loop injection method (option)



NOTE

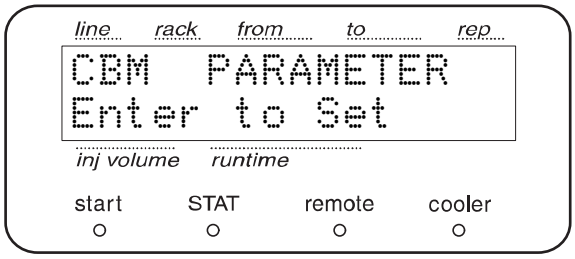
- When the set value is changed, the screen shown at the right is displayed. Turn the power OFF and turn it ON again.
- To perform an analysis with loop injection method, a loop injection base kit (P/N S228-45421-91) and a sample loop for loop injection (P/N S228-52612-41 to 43) are required optionally. Do not set [1] at [INJECTION TYPE] when this kit is not provided. This could disable accurate analysis.

"1.6 Optional Parts" P. 21



5.5.6 CBM Parameter Group

This group is for setting parameters for system controller CBM-20A/20Alite.



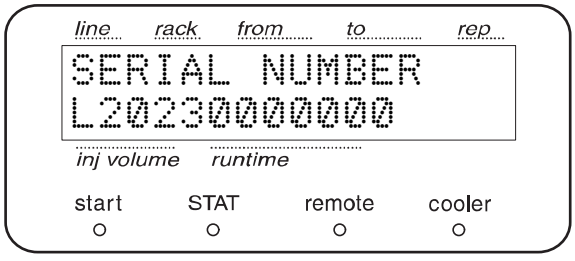
NOTE

CBM parameter group is only displayed when the instrument is connected to a CBM.

Press **enter** and see the Parameter.

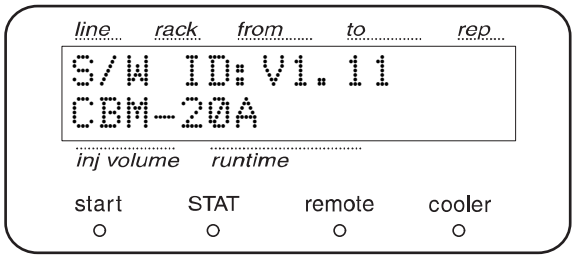
■ Displaying the Serial Number [SERIAL NUMBER]

The CBM-20A's serial number is displayed on the second line.



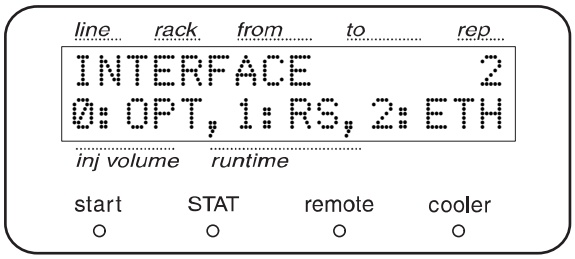
■ Displaying the S/W Version Number [S/W ID]

The program version number is displayed on the first line and the name of the system controller is displayed on the second line.



■ Setting Transmitting Medium between CBM-20A and Data Processing Unit [INTERFACE]

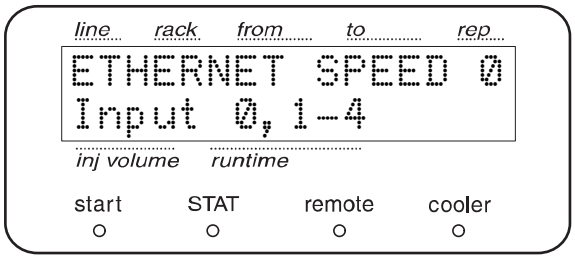
The current setting is displayed on the first line.
Enter the set value by the numeric keypad and press **enter**.



Set value	Function
0	Connects with optical cable.
1	Connects with serial transmission (RS-232C).
2	Connects with Ethernet.

■ Setting Transmitting Speed of Ethernet [ETHERNET SPEED]

The current setting is displayed on the first line.
Enter the set value by the numeric keypad and press **enter**.



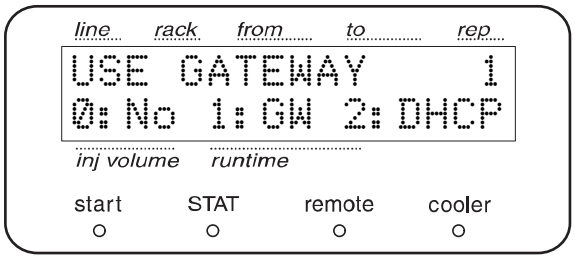
Set value	Function
0	Realizes automatically.
1	Sets to 10 Mbps, Half Duplex.
2	Sets to 10 Mbps, Full Duplex.
3	Sets to 100 Mbps, Half Duplex.
4	Sets to 100 Mbps, Full Duplex.

■ Setting Used/Not used of Default Gateway [USE GATEWAY]

NOTE

Depending on the setting for the system controller, this item may not require setting and so therefore may not be displayed.

The current setting is displayed on the first line.
Enter the set value by the numeric keypad and press **enter**.



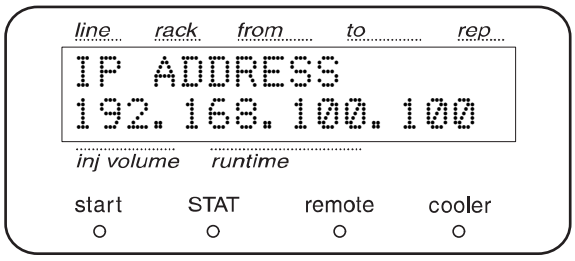
Set value	Function
0	Not used.
1	Used.
2	Acquires IP address automatically.

■ Setting the IP Address [IP ADDRESS]

NOTE

Depending on the setting for the system controller, this item may not require setting and so therefore may not be displayed.

The current setting is displayed on the second line. Enter the set value by the numeric keypad and press **enter**.



NOTE

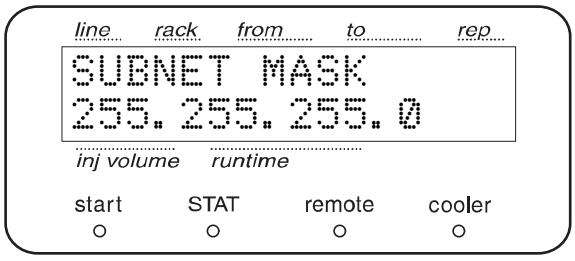
Obtain the setting from your network administrator.

■ Setting the Subnet Mask [SUBNET MASK]

NOTE

Depending on the setting for the system controller, this item may not require setting and so therefore may not be displayed.

The current setting is displayed on the second line. Enter the set value by the numeric keypad and press **enter**.



NOTE

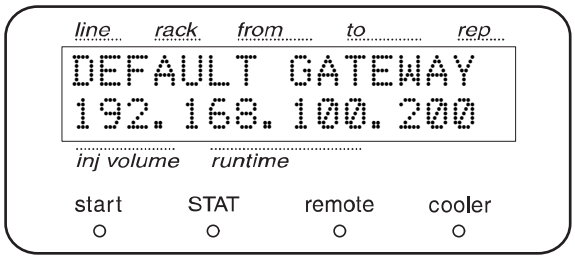
Obtain the setting from your network administrator.

■ Setting the Default Gateway [DEFAULT GATEWAY]

NOTE

Depending on the setting for the system controller, this item may not require setting and so therefore may not be displayed.

The current setting is displayed on the second line. Enter the set value by the numeric keypad and press **enter**.

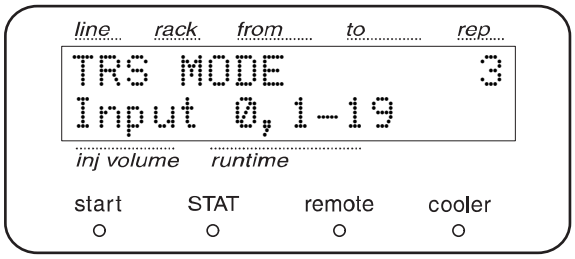


NOTE

Obtain the setting from your network administrator. When using the DHCP function, [---] is displayed as the address setting.

■ Setting the Serial Communication [TRS MODE]

The current setting for serial communications is displayed in the first line.
Enter the set value by the numeric keypad and press **enter**.



Set value	Function
0	The communications setting is not changed.
1	Not used (reserved for expansion).
2	Cannot be used.
3	Connects with LCsolution (default).
4-10	Not used (reserved).
11	Connects with C-R8A.
12	Connects with C-R7A/C-R5A.
13	Connects with C-R4A.
14	Connects with C-R6A (without extended ROM board).
15	Connects with C-R6A (with extended ROM board).
16-19	Not used (reserved for expansion).

5.6 Control by CBM-20A or CBM-20Alite System Controller

5.6.1 Preparation

To control the instrument by the CBM-20A or CBM-20Alite system controller, set the parameters as follows: Set [LOCAL] to [0: Remote], [OP MODE] to [0: 30A].

Command	Set Value	References
LOCAL	0: Remote	"[LOCAL]" P. 106
OP MODE	0: 30A, 1: 20A	"[OP MODE]" P. 148

NOTE

- This instrument cannot be connected to the SCL-10Avp system controller, even if [1: 20A] is set at [OP MODE].
- Select [1: 30A] at [OP MODE], to control the instrument using the CBM-20A/20Alite with software version of 1.11 or earlier.
- To control the instrument using the CBM-20A/20Alite of software version 2.00 and later, select [0: 30A] at [OP MODE]. However, if the workstation is not LabSolutions of version 5.2 and later, select [1: 20A].
- When [1: 20A] is set at [OP MODE], some parameters for auxiliary functions such as internal rinsing of the needle are not downloaded from the CBM-20A/20Alite or workstation and these parameters must be set on the instrument.

"5.1.2 Auxiliary Settings Screens" P. 68

(1) Connection with LabSolutions

Workstation	CBM-20A/20Alite	SIL-30AC	[OP MODE] Setting
LabSolution Ver. 5.2 and later	Ver. 2.00 and later	Ver. 2.00 and later	[0: 30A]
	Ver. 1.21 - 1.30		[1: 20A]

(2) Connection with a workstation other than LabSolutions

Workstation	CBM-20A/20Alite	SIL-30AC	[OP MODE] Setting
LCsolution Ver. 1.24 and later	Ver. 1.21 and later	Ver. 2.00 and later	[1: 20A]
LCMSsolution Ver. 3.50 and later			
Shimadzu LC Driver Ver. 2.00 SP1 and later (Empower1/Empower2)			

5.6.2 Basic Parameters

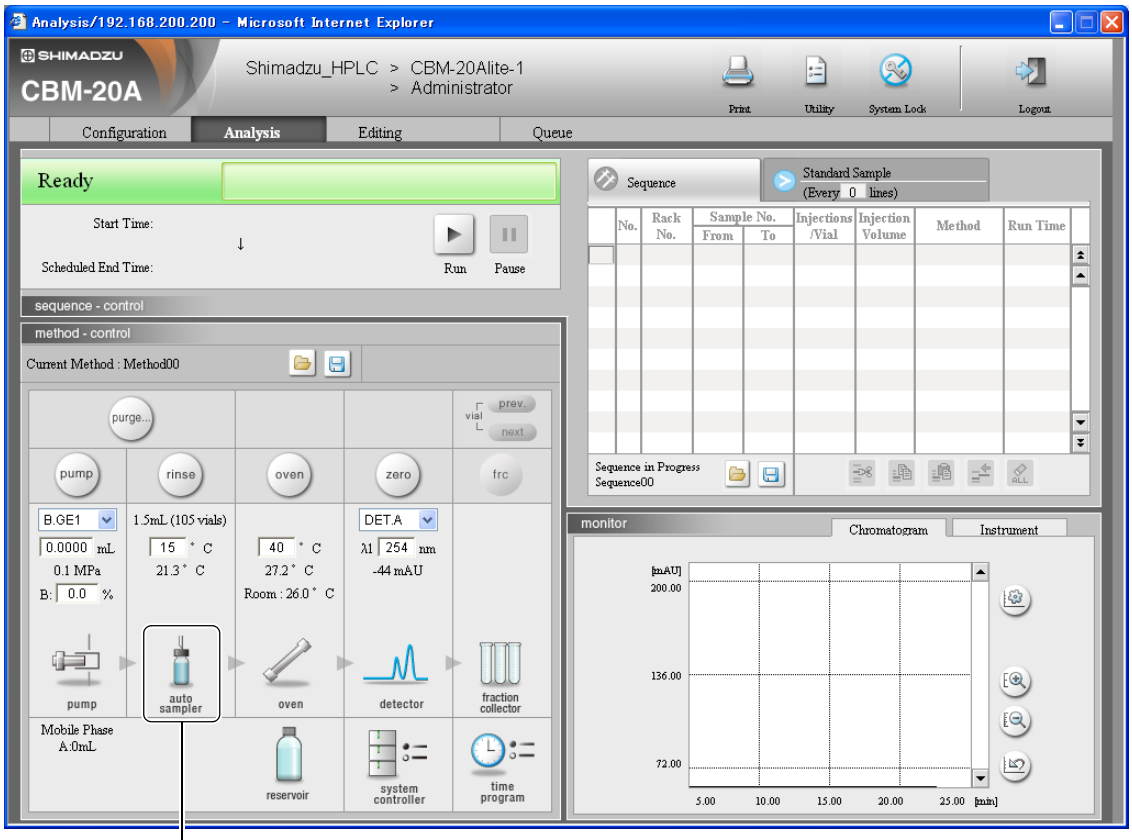
The basic setting conditions and the analysis sequence are set with the CBM-20A/20Alite's analysis sequence screen.
Refer to the CBM-20A/20Alite instruction manual for details.

■ Control by the CBM-20A

When the instrument is controlled by the CBM-20A (or CBM-20Alite), the methods relating to the instrument can be set on the three web controller screens shown below.

■ Instrument Setting Screens at the CBM-20A/20Alite Web Controller

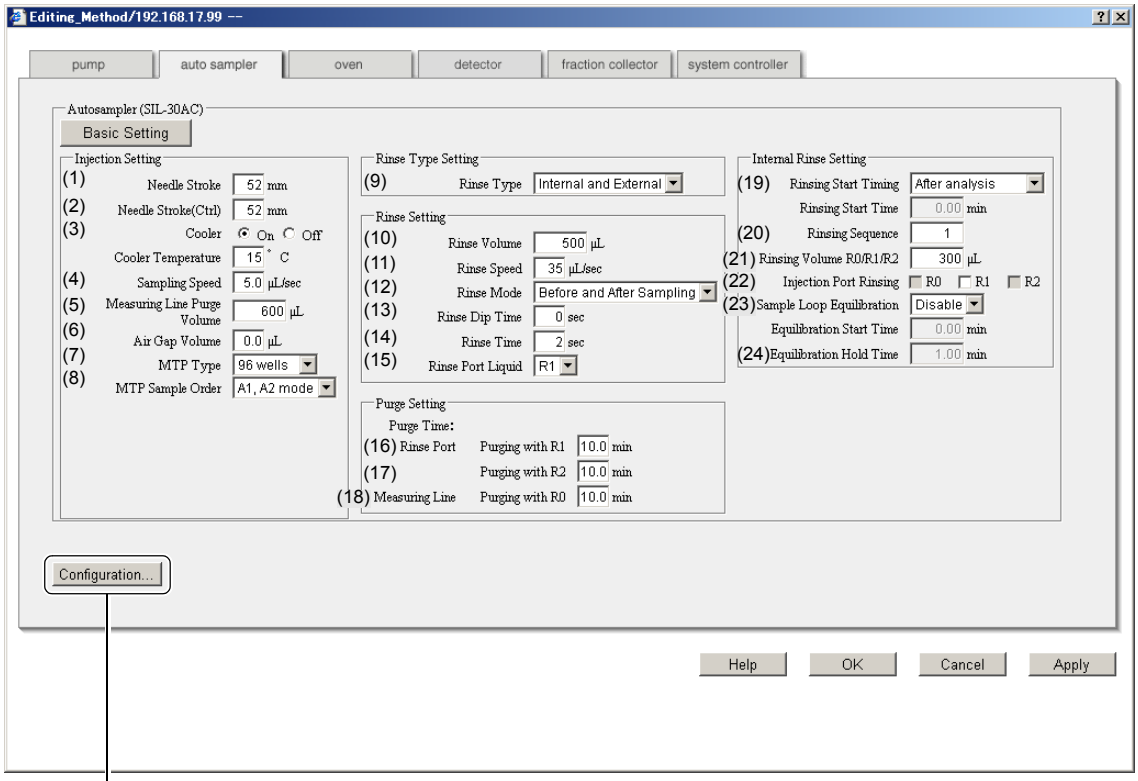
- CBM-20A/20Alite web controller: [Analysis] screen



Autosampler injection setting button

- CBM-20A/20Alite web controller: Autosampler injection detailed settings screen (total injection method)

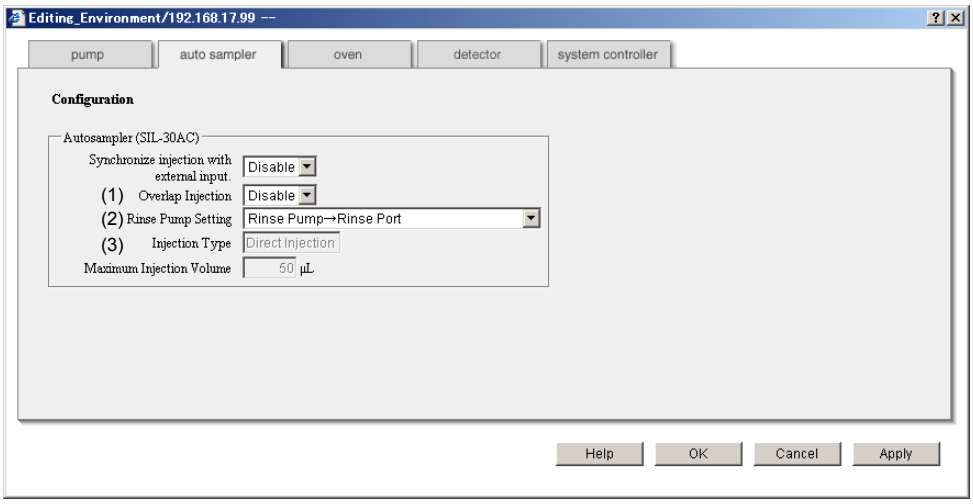
* When RINSE TYPE = 2 (external and internal rinsing of the needle)



[Configuration] button

No.	Indication on the SIL-30AC	See	No.	Indication on the SIL-30AC	See
(1)	NEEDLE STROKE	P. 85	(13)	DIP-R TIME	P. 94
(2)	CNT RACK STRK	P. 107	(14)	FLOW-R TIME	P. 96
(3)	COOLER TEMP	P. 82	(15)	DIP-R SOL	P. 94
(4)	SAMPLE SPEED	P. 84	(16)	PURGE TM R1	P. 83
(5)	ML PURGE VOL	P. 83	(17)	PURGE TM R2	P. 83
(6)	AIR GAP	P. 92	(18)	PURGE TM R0	P. 83
(7)	MTP WELL	P. 89	(19)	START TM	P. 96
(8)	MTP ORDER	P. 89	(20)	SOL. SEQ	P. 97
(9)	RINSE TYPE	P. 80	(21)	SOL. VOLUME	P. 98
(10)	DIP-R VOL	P. 94	(22)	INJ.P RINSE	P. 100
(11)	RINSE SPEED	P. 84	(23)	LOOP S.TM	P. 98
(12)	RINSE MODE	P. 93	(24)	LOOP HOLD TIME	P. 99

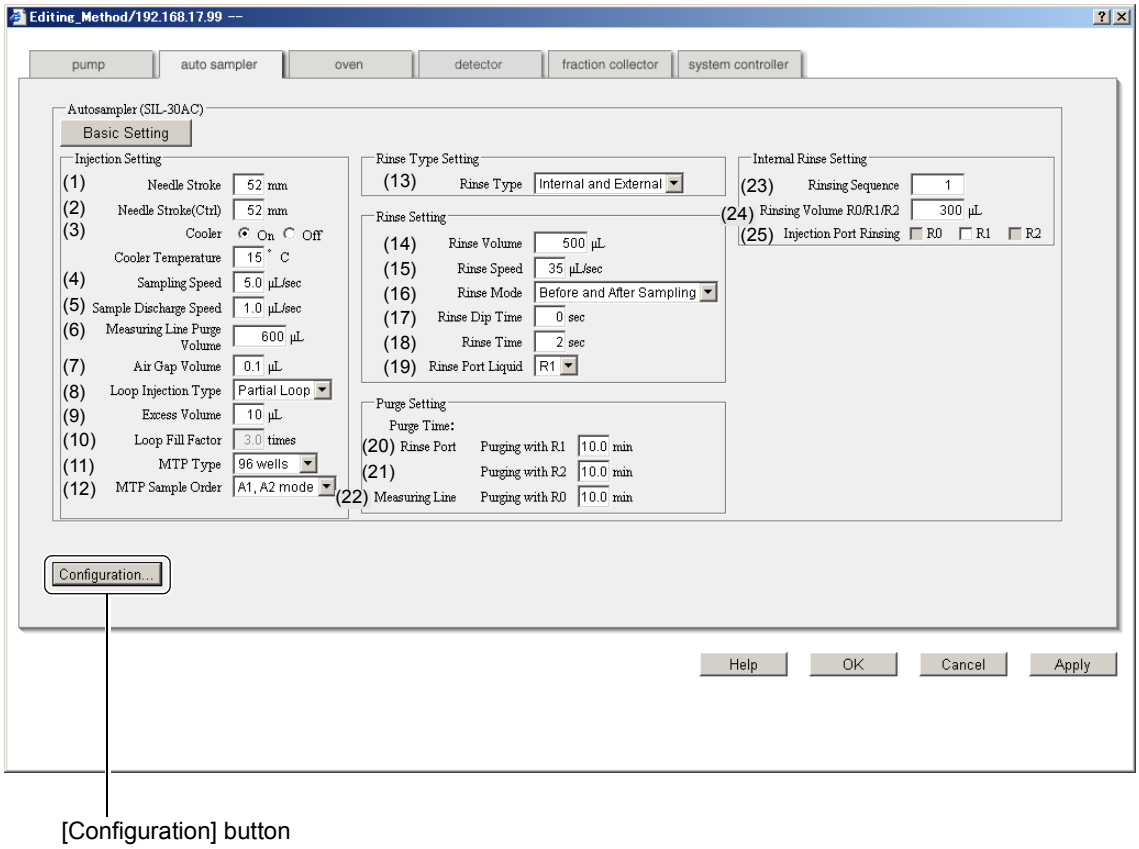
- CBM-20A/20Alite web controller: Autosampler configuration screen (total injection method)



No.	Indication on the SIL-30AC	Remark	See
(1)	FLOW-R METHOD		P. 95
(2)	INJECTION TYPE	Available only at the SIL-30AC	P. 154
(3)	MAX INJ. VOLUME	Available only at the SIL-30AC	P. 108

- CBM-20A/20Alite web controller: Autosampler injection detailed settings screen (loop injection method)

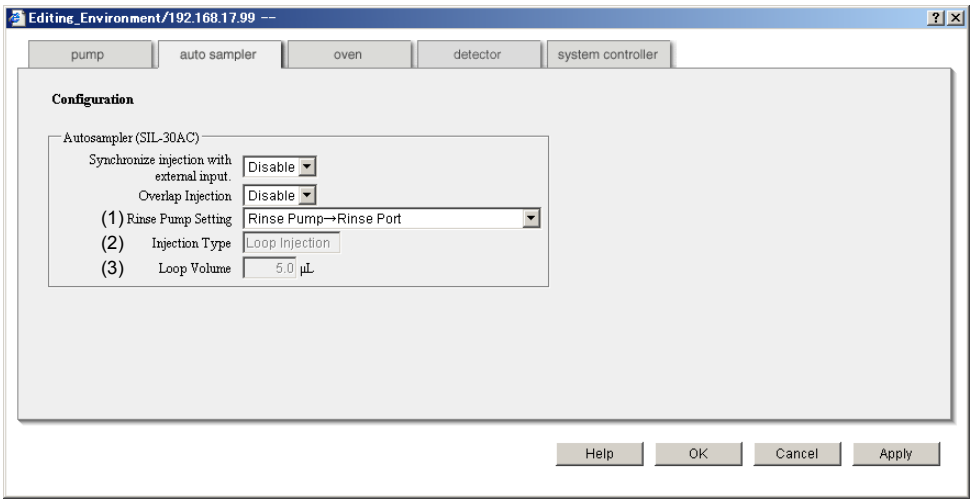
* When RINSE TYPE = 2 (external and internal rinsing of the needle)



[Configuration] button

No.	Indication on the SIL-30AC	See	No.	Indication on the SIL-30AC	See
(1)	NEEDLE STROKE	P. 85	(14)	DIP-R VOL	P. 94
(2)	CNT RACK STRK	P. 107	(15)	RINSE SPEED	P. 84
(3)	COOLER TEMP	P. 82	(16)	RINSE MODE	P. 93
(4)	SAMPLE SPEED	P. 84	(17)	DIP-R TIME	P. 94
(5)	DISP SPEED	P. 85	(18)	FLOW-R TIME	P. 96
(6)	ML PURGE VOL	P. 83	(19)	DIP-R SOL	P. 94
(7)	AIR GAP	P. 92	(20)	PURGE TM R1	P. 83
(8)	LOOP INJ TYPE	P. 92	(21)	PURGE TM R2	P. 83
(9)	EXCESS VOLUME	P. 92	(22)	PURGE TM R0	P. 83
(10)	LOOP FILL FACTOR	P. 93	(23)	SOL. SEQ	P. 97
(11)	MTP WELL	P. 89	(24)	SOL. VOLUME	P. 98
(12)	MTP ORDER	P. 89	(25)	INJ.P RINSE	P. 100
(13)	RINSE TYPE	P. 80			

- CBM-20A/20Alite web controller: Autosampler configuration screen (loop injection method)



No.	Indication on the SIL-30AC	Remark	See
(1)	FLOW-R METHOD		P. 95
(2)	INJECTION TYPE	Available only at the SIL-30AC	P. 154
(3)	VALVE LOOP VOL	Available only at the SIL-30AC	P. 108

5.7 External Input/Output Terminals

The external input/output terminals are connected to an event output device or another external device with a provided event cable.
Details of the terminal and wiring are described as follows.

⚠CAUTION

- Before connecting the cable, turn off the power and unplug the instrument.
- Use only the specified cable.
- Connect as specified.

Otherwise, fire, electric shock or malfunction may occur.

5.7.1 Event Cable

Signals	Description	Remark
START (output) (injected)	Relay contact output. Switches ON/OFF when the autosampler starts analysis.	Contact rating: 30 VDC/1 A
EVENT1 (output)	Relay contact output. Output switched by [EVENT] setting.	
EVENT2 (output)	Relay contact output. Output switched by [EVENT] setting.	
EVENT3/ERROR (output)	Relay contact output or error output. Output switched by [EVENT] setting.	
RSVD	For factory adjustment. Not used.	
READY (input)	When this contact is closed, the autosampler becomes ready to start injection operation. When this contact is opened, the autosampler start injection operation (from pretreatment operation). Used to start injection operation in synchronization with external devices.	
STOP (input)	Injection operation stops when this contact is closed.	
CHANGER	Connected when using a rack changer.	

5.7.2 Connection of Event Cable

1 Peel the cable about 10 mm.
* Note that this is not necessary with the EVENT cable provided.

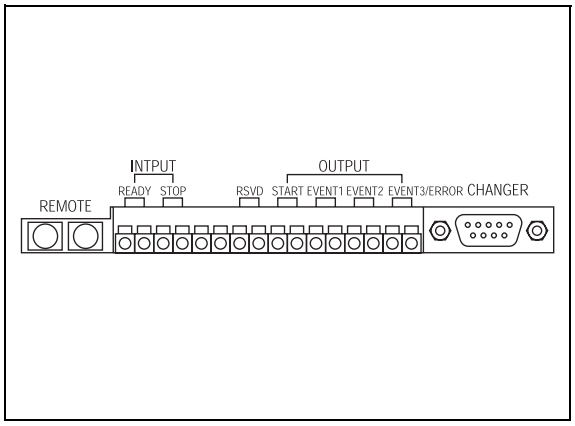


Fig. 5.14

2 Insert the cable.
When the cable has the single core wire, just insert the cable.
When the cable has the stranded wires, strand the wires enough and insert with pressing the button of the terminal.
When removing the cable, remove the cable by pressing the button of the terminal.

NOTE

One EVENT cable is provided with this instrument. When connecting to terminals in two or more circuits, prepare the cables shown below.
The compatible diameter range of the core wire is given below.
- Cable with single wire : ϕ0.4 to ϕ1.2 (AWG26 to 16)
- Cable with stranded wire : 0.3 mm^2 to 1.25 mm^2 (AWG22 to 16), diameter of single wire thicker than ϕ0.18.

The cable with stranded wire is suitable to prevent disconnection.

5.8 Example of Time Settings at Parameters for Internal Rinsing of the Needle

Three examples are shown for time settings at the parameters (START TM, LOOP S. TM, LOOP HOLD TIME) for internal rinsing of the needle. The sample loop, the needle, and the high-pressure valve flow line must be rinsed for sensitive analysis in order to reduce carryover. Although the throughput may be lowered, we recommend the operations shown in Example 1 below as this has less possibility of disturbing the baseline or sample peak shape. When "2" is set at [RINSE TYPE] and the default values are used for other parameters for internal rinsing of the needle, the operations shown in Example 2 will be made.
In either case, perform a preliminary evaluation before the actual analysis operation and ensure that the accurate analysis results can be obtained from the operations.

5.8.1 Example 1: When Performing the Internal Rinsing of the Needle and Sample Loop Equilibration After Sample Peak Detection (Recommended)

Parameter Name	Set Value	See	Remark
START TM	1.3 (min) *1)	*4)	Start internal rinsing of the needle after sample peak detection. The high-pressure valve position is switched to the load position.
LOOP S. TM	3.3 (min) *1) *2)	*5)	The sample loop is equilibrated after the internal rinsing of the needle. The high-pressure valve position is switched to the injection position.
LOOP HOLD TIME	1.0 (min) *3)	*6)	Set the time to keep the equilibration of the sample loop. The high-pressure valve position remains in the injection position.

* In this example, default values are taken for the parameters other than the above.
*1) Set the time that counts from the start of analysis.
*2) If the internal rinsing of the needle has not finished by the set time, the high-pressure valve will be switched to the injection position on completion of internal rinsing of the needle.
*3) The set time here is the time to reset the high-pressure valve to the injection position.
*4) "5.2.4 Internal Rinsing Parameter Settings Group" P. 96 "[START TM]" P. 96
*5) "5.2.4 Internal Rinsing Parameter Settings Group" P. 96 "[LOOP S. TM]" P. 98
*6) "5.2.4 Internal Rinsing Parameter Settings Group" P. 96 "[LOOP HOLD TIME]" P. 99

NOTE

- Advantages

1. Since the high-pressure valve position remains in the injection position until sample peaks are eluted, the baseline stays flat.
2. After rinsing the sample loop, the needle, and the flow line of the high-pressure valve, high-pressure pumping replaces the solution completely with the mobile phase.

• Disadvantages

1. When the analysis time is short, throughput decreases because flow line rinsing and replacement with mobile phase take some time.

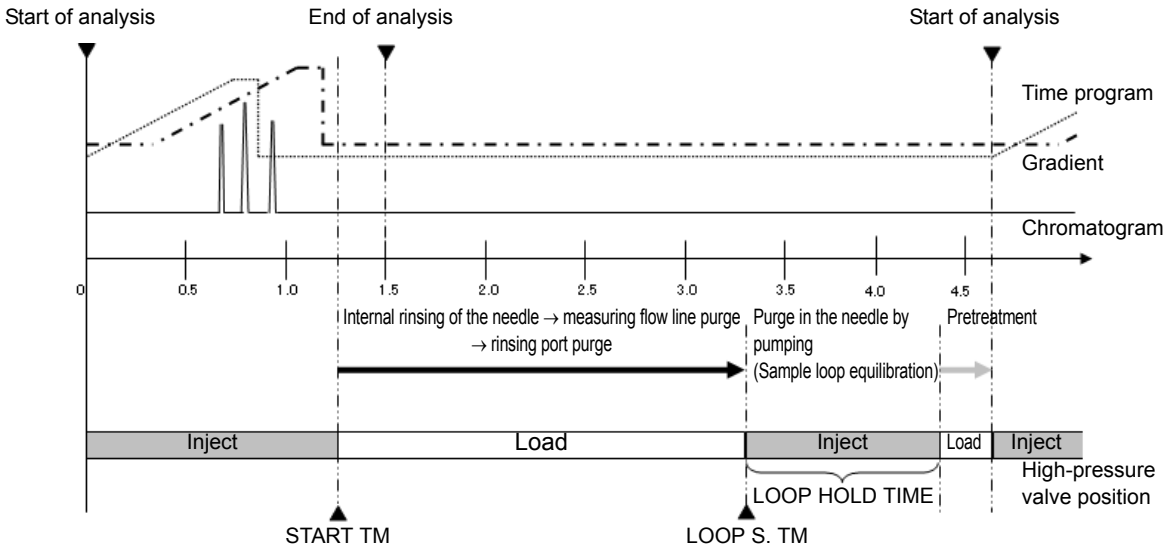


Fig. 5.15

5.8.2 Example 2: When Performing Internal Rinsing of the Needle After Analysis (without Sample Loop Equilibration)

Parameter Name	Set Value	See	Remark
START TM	−1:AUTO	*1)	Internal rinsing of the needle starts immediately after analysis. The high-pressure valve position is switched to the load position.
LOOP S. TM	−1:LOAD	*2)	The sample loop is not equilibrated after internal rinsing of the needle. The high-pressure valve position remains in the load position.
LOOP HOLD TIME	-	*3)	This is not displayed when "-1" is set at [LOOP HOLD TIME].

* In this example, default values are taken for the parameters other than the above.

*1) "5.2.4 Internal Rinsing Parameter Settings Group" P. 96 "[START TM]" P. 96

*2) "5.2.4 Internal Rinsing Parameter Settings Group" P. 96 "[LOOP S. TM]" P. 98

*3) "5.2.4 Internal Rinsing Parameter Settings Group" P. 96 "[LOOP HOLD TIME]" P. 99

NOTE

• Advantages

1. Since the high-pressure valve position remains in the injection position until the analysis ends, the baseline stays flat.

- Disadvantages

1. When the analysis time is short, the throughput decreases because rinsing is started after analysis.
2. Since the rinse solution is replaced with the mobile phase using the measuring pump after rinsing of the sample loop, the needle, and the flow line of the high-pressure valve, some of it may remain in the internal surface of the flow line, which may disturb the baseline or sample peaks for the next analysis.

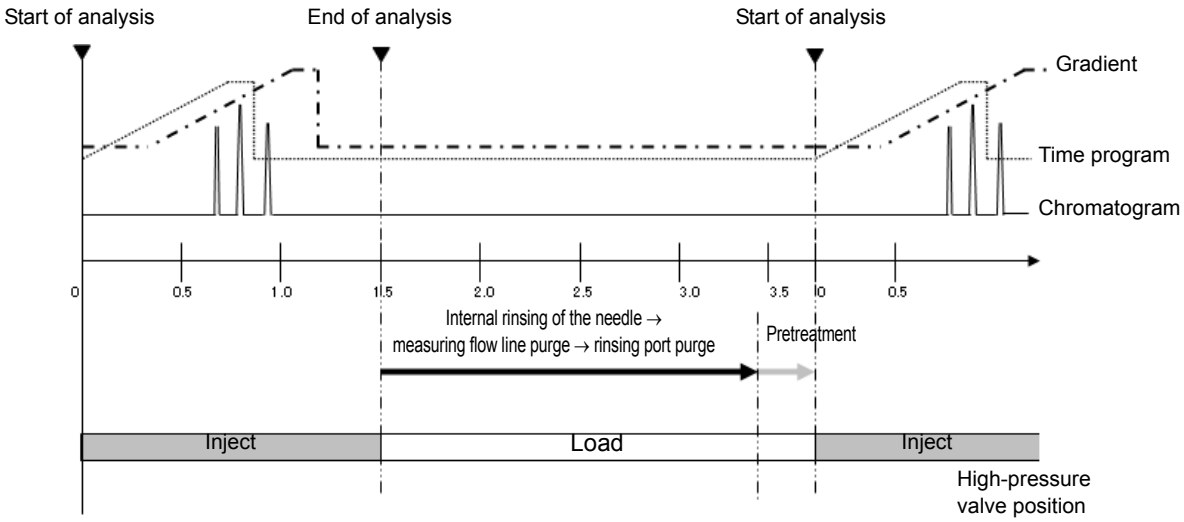


Fig. 5.16

5.8.3 Example 3: When Performing the Internal Rinsing of the Needle and Sample Injection

Parameter Name	Set Value	See	Remark
START TM	0.2 (min) *1)	*4)	Set the amount of time from the sample passing through the high-pressure valve to the gradient-delivered solvent being loaded into the high-pressure valve. The valve position is switched to the load position.
LOOP S. TM	2.2 (min) *1) *2)	*5)	The sample loop is equilibrated after the internal rinsing of the needle. The valve position is switched to the injection position. The valve position is switched to the injection position.
LOOP HOLD TIME	1.0 (min) *3)	*6)	Set the time to keep the equilibration of the sample loop. The valve position remains in the injection position.

* In this example, default values are taken for the parameters other than the above.

*1) Set the time that counts from the start of analysis.

*2) If the internal rinsing of the needle has not finished by the set time, the high-pressure valve will be switched to the injection position on completion of internal rinsing of the needle.

*3) The set time here is the time to reset the high-pressure valve to the injection position.

*4) "5.2.4 Internal Rinsing Parameter Settings Group" P. 96 "[START TM]" P. 96

*5) "5.2.4 Internal Rinsing Parameter Settings Group" P. 96 "[LOOP S. TM]" P. 98

*6) "5.2.4 Internal Rinsing Parameter Settings Group" P. 96 "[LOOP HOLD TIME]" P. 99

NOTE

- Advantages

1. Since rinsing starts immediately after sample injection, it is possible to reduce the decrease in throughput that occurs due to rinsing of the sample loop, the needle, and the flow line of the high-pressure valve and replacement with the mobile phase.
2. It is possible to shorten the time of restoring the mobile phase composition to the initial concentration after gradient analysis.
3. After rinsing the sample loop, the needle, and the flow line of the high-pressure valve, high-pressure pumping replaces the solution completely with the mobile phase.

- Disadvantages

1. Since the high-pressure valve is switched to the load position immediately after the sample injection, the baseline or sample peak shape may be disturbed.

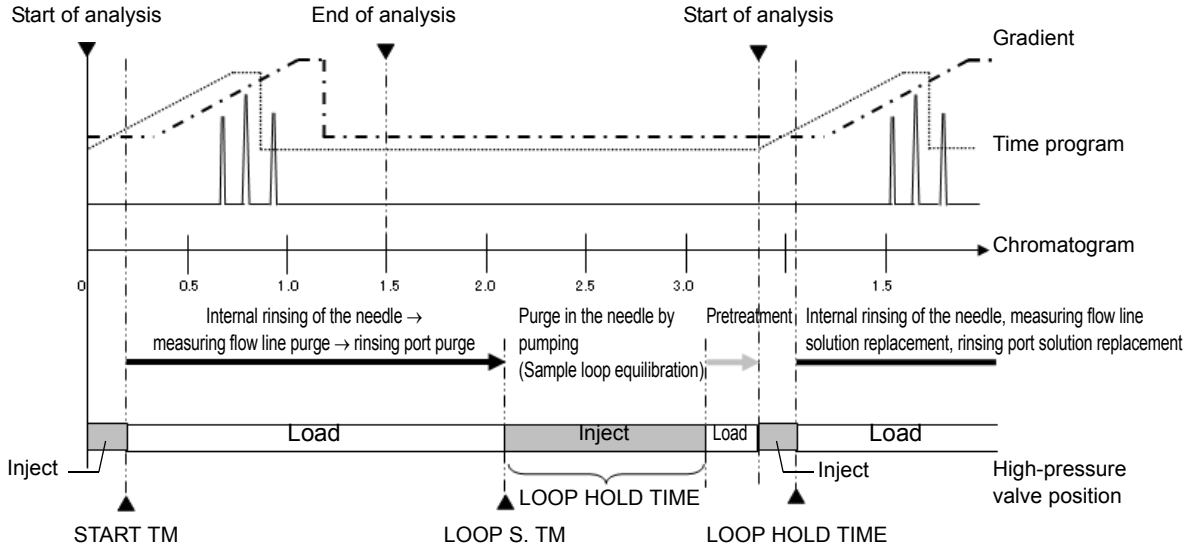


Fig. 5.17

5.9 Pretreatment Function

5.9.1 About Pretreatment Function

The pretreatment function enables you to perform a variety of sample injection operations that are different from the standard injection operation. Each operation can be executed by creating a simple program. Basic operations, such as sample dilution or addition of a reagent, can be programmed by simply inputting values for the relevant parameters on the screen.

NOTE

The pretreatment program can be created under the system configuration shown below and can only be set from the workstation, LabSolutions (version 5.2 and later).
Note that the program cannot be created on the SIL-30AC or CBM-20A/20Alite.

System configuration for using the pretreatment function

Workstation	CBM-20A/lite	SIL-30AC	[OP MODE] Setting
LabSolutions Ver. 5.2 and later	Ver. 2.00 and later	Ver. 2.00 and later	[0 : 30A]

NOTE

• Performance expected when the pretreatment function is used

<Total injection>

Item	Performance Expected	Remark
Dilution accuracy	± 10 % max.	
Area reproducibility between vials	RSD ≤ 1 % (When 5 μL is injected)	This assumes the case where the diluent is drawn from rinse solution R0 and is used for dilution at a dilution factor less than 10 and the total volume of diluted solution is 100 μL.

<Loop injection> using a 5 μL loop

Item	Performance Expected	Remark
Dilution accuracy	± 10 % max.	
Area reproducibility between vials	RSD ≤ 2 % (5 μL loop: when 2 μL is injected) (20 μL loop: when 5 μL is injected)	This assumes the case where the diluent is drawn from rinse solution R0 and is used for dilution at a dilution factor less than 10 and the total volume of diluted solution is 100 μL.

NOTE

- Sample vials recommended when the pretreatment function is used

To improve the mixing performance, use the sample vials given below.

Part Name	Capacity	Material	Part No.	Application	Remark
1 mL sample vial	1 mL	Polypropylene Cap: Polyethylene	S228-31600-91	For general purpose / small capacity Disposable Needle stroke must be 50 mm or less.	200 vials (with cap)

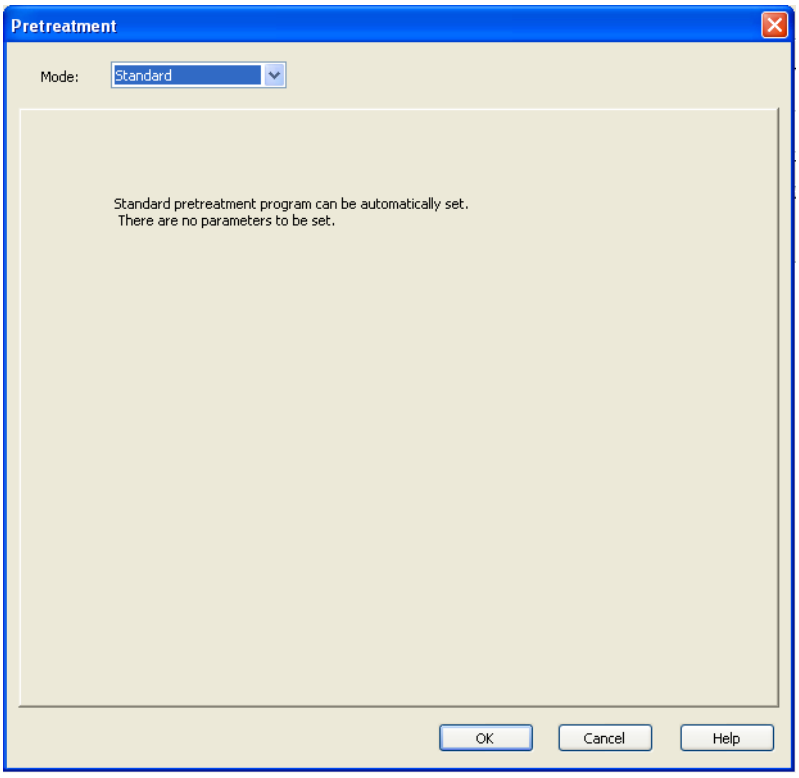
5.9.2 Pretreatment Program Creation Modes

The [Pretreatment] screen has four modes: Standard, Pretreatment Program, Dilution, and Reagent.

■ Standard Mode

In the standard mode, the standard pretreatment program is automatically set and no further settings are required.

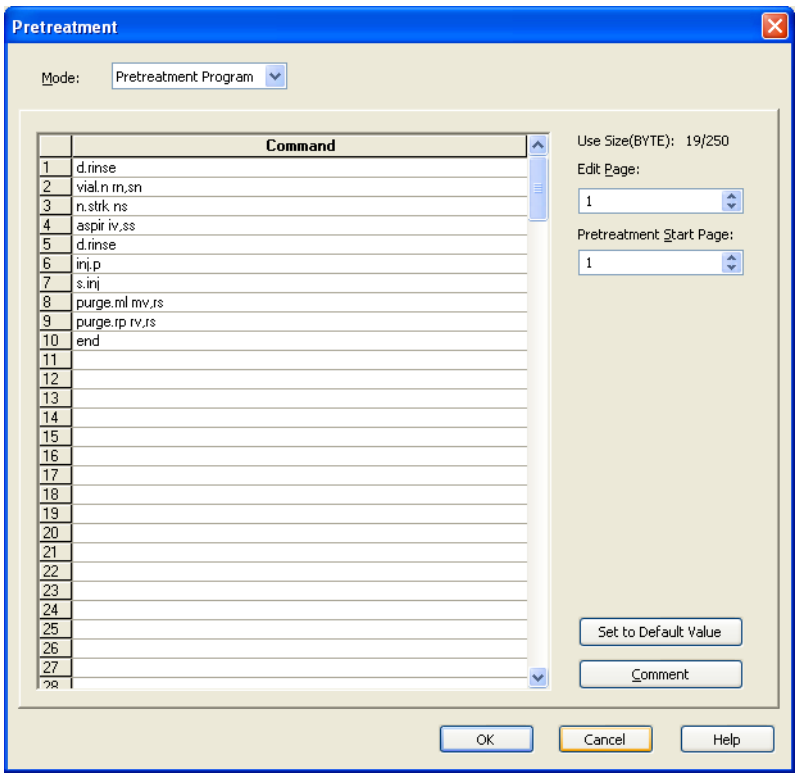
- Standard mode screen



■ Pretreatment Program Mode

The pretreatment program mode enables you to combine multiple commands to perform a variety of sample injection operations that are different from the standard injection operation. The pretreatment program mode screen as well as the functions and setting ranges for the commands is explained.

- Pretreatment program mode screen



Item	Description	Set Range (Units)	Default Value
Edit Page	The pretreatment program has a limitation of 250 bytes per page. The number of bytes used for each command is from 1 byte to 10 bytes depending on the command. If the program becomes too large, an error will be displayed. In such a case, divide the program. NOTE Use page 0 to set the pretreatment program for injection. Set any pretreatment programs other than that for injection on page 1 and later.	1 to 19	1
Set to Default Value	Click to set the program that is configured in the standard mode on the page currently displayed.	-	-
Pretreatment Start Page	Use this page and onwards for pretreatment programs.	1 to 19	1

Item	Description	Set Range (Units)	Default Value
Command	A computer program language similar to BASIC is used. The pretreatment process (rinsing the needle, sample dilution, addition of reagent, agitation, wait for reaction, etc.) that is necessary for injection using an autosampler can be programmed. Each program consists of multiple command (instruction to the computer in the program language) lines.		
Comment	Set a comment for the pretreatment program.	Within 7 lines, 1024 one-byte characters per line	Blank

• List of Commands

The available commands and their meaning are shown below.

Command	Description	Set Range (Units)
inj.p	Move the needle to the injection port and move it down to the bottom of the injection port.	-
home	Return the needle and valve to the home position.	-
v.inj	Switch the high-pressure valve to the injection side (the flow line to which the sample loop is connected to the pump and the column).	-
v.load	Switch the high-pressure valve to the load side (the flow line to which the sample loop is not connected to the pump and the column).	-
s.inj	Set the high-pressure valve to the injection side and inject the sample into the column. At the same time, the start signal is output from the autosampler to the data processor.	-
call (file number)	Call another file (FILE0 to FILE19). Only one file can be called.	f0 - f19
event (relay number)	Turn ON the relay(s) (1 - 3) of external devices connected to the CBM-20A/20Alite. Set the relay number(s). When turning ON relay 1 and relay 2, set [EVENT12].	0 - 123 (0, 1, 2, 3, 12, 13, 23, 123)
n.strk (distance)	Set the distance of moving the needle down. If the needle stroke setting is not appropriate for analysis, an error is given from the autosampler.	Variable:ns (Needle stroke value set in the "Method" section) a0 - a7 (user variable) Value: 0 - 100 (integer not smaller than 0)
wait (time)	Set the wait time in minutes.	0.1 - 999.0 (min)
wait.sec (time)	Set the wait time in seconds.	0.5 - 9.9, 10 - 99 (sec)

Command	Description	Set Range (Units)
wait.rt (time)	Wait until the set time arrives. When "0" is set, the system waits until the end of analysis before starting the next process.	0.01 - 9.99 (min) 10.0 - 99.9 (min) 100 - 9999 (min)
vial.n (rack number), (sample vial number)	Move to the sample vial of the specified number. *1	Rack number: 0 - 12 Sample vial number: The setting range varies depending on the rack type set in the "Method" section.
go to (line number or file number)	Jump to the specified line number or file number.	Line number: 0 - 255 File number: f0 - f19
purge.ml (volume), (speed)	Purge the measuring flow line.	Volume: 1 - 2000 (μL) Speed: 1 - 35 (μL/sec)
purge.rp (volume), (speed)	When the internal/external rinsing of the needle is selected for [Rinse Type]: Deliver the specified volume of the solution selected for dip-rinsing in the rinsing port at the specified speed. When the internal/external rinsing of the needle is not selected for [Rinse Type]: Deliver the specified volume of rinse solution R0 to the rinsing port at the specified speed.	Volume: 1 - 2000 (μL) Speed: 1 - 35 (μL/sec)
i.rinse	Rinse the internal surface of the needle under the conditions set in the needle internal rinsing setting dialog.	-
d.rinse	Dip the needle for the duration of the set needle dipping time. (No rinse solution is delivered.)	-
f.rinse	Rinse the needle for the duration of the set rinsing time for the rinsing pump.	-
start	Output the digital start signal and event terminal from the high-pressure valve without sending it to the injection.	-
n.drain	Return the needle to the home position, move it to the drain position, and lower it a short distance.	-
aspir (volume), (speed)	Aspirate the specified volume of solution at the specified speed. In the offline condition, this defaults to the needle loop volume of the autosampler connected last time.	Volume: 0.1 - 9.9, 10 - needle loop capacity (μL) Speed: 0.1 - 15.0 (μL/sec)
disp (volume), (speed)	Dispense the specified volume of solution at the specified speed. "disp 0" denotes a pre-pushing.	Volume: • Direct injection 0.1 - 9.9, 10 - 2000 (μL) • Loop injection 0 (pre-pushing) 0.1 - 9.9, 10 - 2000 (μL) Speed: 0.1 - 35.0 (μL/sec)
air.a (volume), (speed)	Aspirate the specified volume of air at the specified speed.	Volume: 0.1 - 9.9, 10 - needle loop capacity (μL) Speed: 0.1 - 15.0 (μL/sec)

Command	Description	Set Range (Units)
apport (rack number 1), (sample vial number 1), (rack number 2), (sample vial number 2), (volume), (speed)	The specified volume (μL) of sample in vial No. 1 at rack No. 1 is taken and supplied to vial No. 2 at rack No. 2. *1	Rack number 1: 0 - 12 Sample vial number 1: Depends on the rack. Rack number 2: 0 - 12 Sample vial number 2: Depends on the rack. Volume: 0.1 - 9.9, 10 - 2000 (μL) Speed: 0.1 - 15.0 (μL/sec)
dil (rack number), (vial number), (volume), (speed), (solution)	Rinse solution type "0" denotes R0, "1" denotes R1, and "2" denotes R2. ① Move the needle to the drain position and purge the flow line up to the needle tip with the selected solution. ② Move the needle to the specified sample vial number and move it down. ③ Dispense the specified volume of sample from the needle tip. ④ Move the needle to the drain position and fill the sample loop and the flow line up to the needle tip with mobile phase (R0). When R0 is selected for the solution, this operation is not performed.	Rack number: 0 - 12 Sample vial number: Depends on the rack. Volume: 1 - 2000 (μL) Speed: 1 - 35 (μL/sec) Solution: 0, 1, 2
mix (times), (upper air volume), (sample aspiration volume), (aspiration speed), (dispensing speed)	Mixing is performed for the specified volume at the specified aspiration speed and dispensing speed for the specified number of times.	Times: 1 - 10 Upper air volume: 0 - 5 (μL) Sample aspiration volume: 1 - "needle loop capacity - 5" (μL) Aspiration speed: 1 - 35 (μL/sec) Dispensing speed: 1 - 35 (μL/sec)
if (variable), (code), (constant)	IF statement; if the condition is true, execute the next line. *2	Variable: A0 to A7 (decimals available; any setting range from ① to ③ must be selected.) Code: <, >, = Constant: ①Integer setting: 1 - 65535 ②One decimal place setting: 0.1 - 6553.5 ③Two decimal places setting: 0.01 - 655.35
else if		
else	-	-
end if	This is the end of IF statement.	-
for (variable), (code), (start value), (end value)	Perform the operation repeatedly. *3	Variable: A0 - A7 Start: 0 - 65535 End: 0 - 65535
next (variable)	Repeat from the start value to the end value between FOR and NEXT.	Variable: A0 - A7

Command	Description	Set Range (Units)
a0=	User variables	any setting range from ① to ③ must be selected. ①Integer setting: 1 - 65535 ②One decimal place setting: 0.1 - 6553.5 ③Two decimal places setting: 0.01 - 655.35
a1=		
a2=		
a3=		
a4=		
a5=		
a6=		
a7=		
rn	Rack number	MTP, DWP: 0, 1, 2 Rack Changer: 0 - 12 Other: 0, 1
av	Air volume	0, 0.1 - 5.0 (μL)
ds (Valid for loop injection method only)	Dispensing speed	0.1 - 35.0 (μL)
ff (Valid for loop injection method only)	Loop filter factor	1.0 - 5.0 (μL)
mv	Purging measuring line volume (Measuring flow line purge volume)	0 - 2000 (μL)
sn	Sample vial number	-
iv	Sample injection volume	0.1 - 2000 (μL)
ns	Needle stroke	1-mL sample vial rack: 17- 54 (mm) 1.5-mL sample vial rack: 17 - 54 (mm) 1.5-mL sample vial cooling rack: 17 - 54 (mm) 4-mL sample vial rack: 17 - 54 (mm) Microtiter plate rack: 10 - 52 (mm) Deep-well plate rack: 10 - 52 (mm) Rack-changer rack: 10 - 52 (mm)
rv	Rinse solution volume	0 - 2000 (μL)
rs	Aspiration and dispensing speed of rinse solution	1 - 35 (μL/sec)
ss	Sample aspiration and dispensing speed	0.1 - 15.0 (μL/sec)

Command	Description	Set Range (Units)
ev (Valid for loop injection method only)	Excess volume of sample to be aspirated with respect to the sample injection volume	4 - 20 (μL)
end	Terminate the pretreatment program.	-

*1 Rack numbers have the following meanings:
0: Control vial rack
[In the case of sample vial racks]
1: Sample vial rack
[In the case of microtiter plates / deep-well plates]
1: Plate at the front of the rack
2: Plate at the rear of the rack
[In the case of rack-changer racks]
1 - 12: Corresponds to rack-changer rack 1 to 12

*2 Decimal values can be used for judgment.

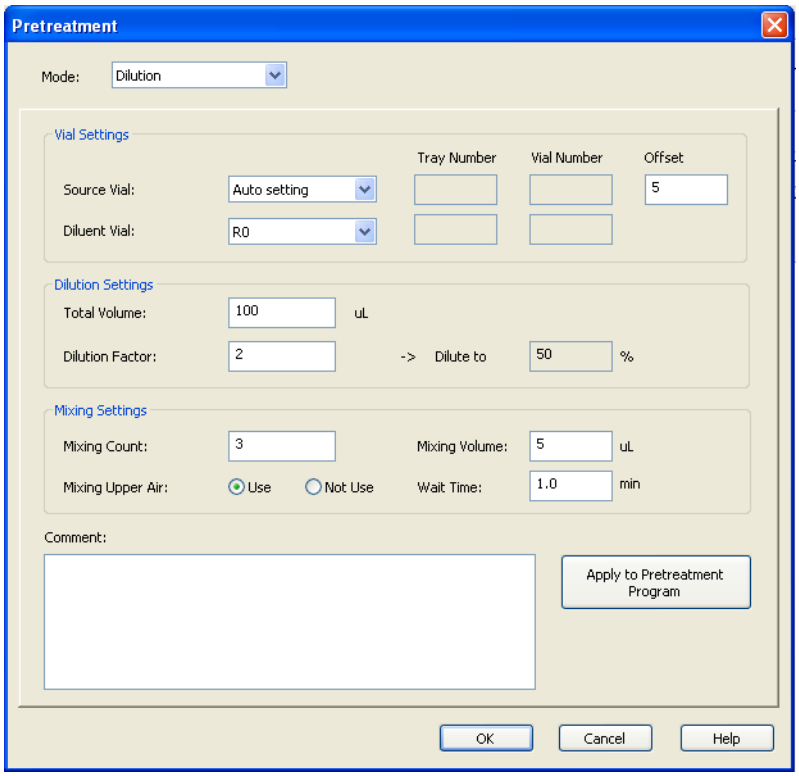
*3 A0 to A7 can be used.
Only integers can be used.
If a decimal value is set at A0 to A7, the fraction part will be discarded.
The step of "For" is incremented by one (+1).

*4 With 1.5-ml vial plates: 10 - 46 (mm)

■ Dilution Mode

Sample dilution included in the pretreatment function can be programmed simply with the parameter settings on the screen shown below.

- Dilution mode screen



- Vial Settings

Set the sample vial that contains a stock solution to be diluted at [Source Vial]. Set the sample vial number that contains a diluent or the rinse solution to be used for dilution at [Diluent Vial].

Item	Description	Set Range (Units)	Default Value
Source Vial	Select the method for specifying the sample vial that contains a stock solution to be diluted.	Auto setting Vial number setting	Auto setting
Diluent Vial	Set the sample vial number that contains a diluent or the rinse solution to be used for dilution.	[Rinse type = no rinsing, external rinsing, or no rinsing (fast)] R0 Vial number setting [Rinse type = internal/external rinsing of the needle] R0 R1 R2 Vial number setting	R0

Item		Description	Set Range (Units)	Default Value
Source Vial (Vial setting)	Tray Number	Specify the tray number of the sample vial that contains a stock solution to be diluted.	[MTP, DWP] 0, 1, 2 [Rack Changer] 0 to 12 [Other] 0, 1	1
	Vial Number	Specify the number of the sample vial that contains a stock solution to be diluted.	[Control vial rack] 1 to 10 [1.5 mL (105 vials) rack] 1 to 105 [1.5 mL (70 vials) rack] 1 to 70 [Cooling rack 1 mL] 1 to 175 [Cooling rack 4 mL] 1 to 50 [Cooling MTP 96] 1 to 96 [Cooling MTP 384] 1 to 384 [Cooling Deep Well 96] 1 to 96 [Cooling Deep Well 384] 1 to 384 [RackChanger MTP96] 1 to 96 [RackChanger DWP96] 1 to 96 [RackChanger MTP384] 1 to 384 [RackChanger DWP384] 1 to 384 [RackChanger 1.5 mL] 1 to 54	1
	Offset (Auto setting)	When auto setting is selected at [Source Vial], set the distance between the injection vial and the source vial.	[1.5 mL (105 vials) rack] 1 to 104 [1.5 mL (70 vials) rack] 1 to 69 [Cooling rack 1 mL] 1 to 174 [Cooling rack 4 mL] 1 to 49 [Cooling MTP 96] 1 to 95 [Cooling MTP 384] 1 to 383 [Cooling Deep Well 96] 1 to 95 [Cooling Deep Well 384] 1 to 383 [RackChanger] 1 to 383	60 35 88 25 48 192 48 192 5
Diluent Vial (Vial setting)	Tray Number	Specify the tray number of the sample vial that contains a diluent.	[MTP, DWP] 0, 1, 2 [Rack Changer] 0 to 12 [Other] 0, 1	1
	Vial Number	Specify the number of the sample vial that contains a diluent.	[Control vial rack] 1 to 10 [1.5 mL (105 vials) rack] 1 to 105 [1.5 mL (70 vials) rack] 1 to 70 [Cooling rack 1 mL] 1 to 175 [Cooling rack 4 mL] 1 to 50 [Cooling MTP 96] 1 to 96 [Cooling MTP 384] 1 to 384 [Cooling Deep Well 96] 1 to 96 [Cooling Deep Well 384] 1 to 384 [RackChanger MTP96] 1 to 96 [RackChanger DWP96] 1 to 96 [RackChanger MTP384] 1 to 384 [RackChanger DWP384] 1 to 384 [RackChanger 1.5 mL] 1 to 54	1

• Dilution Settings

Item	Description	Set Range (Units)	Default Value
Total Volume	Set the volume of the mixed solution after dilution.	100 to 1000 (μL)	100
Dilution Factor	Sets (mixed solution volume) / (stock solution volume).	2 to 100	2

NOTE

If the total volume after dilution is not divisible by the dilution factor, the dilution factor takes precedence so that the total volume will be less than the specified volume.
Example: When "3" is set at [Dilution Factor] and 100 mL is set at [Total Volume]:
Stock solution 33 mL and diluent 66 mL are mixed and the total volume after dilution becomes 99 mL.

• Mixing Settings

Item	Description	Set Range (Units)	Default Value
Mixing Count	The instrument performs mixing while aspirating and dispensing the mixed solution through the needle during dilution. Set the number of mixing times here.	1 to 10	3
Mixing Volume	The instrument performs mixing while aspirating and dispensing the mixed solution through the needle during dilution. Set the volume to be aspirated and dispensed at one time.	1 to (needle loop capacity - 5) (μL)	45
Mixing Upper Air	Select whether or not to perform bubbling by discharging air through the needle during dilution.	Use	Use
		Not Use	
Wait Time	Set the wait time before sample injection after completion of pretreatment.	0.1 to 999.0 (min)	1.0

• Comment
Set a comment for dilution.

NOTE

A comment can be described within 7 lines, 1024 one-byte characters per line.

• Apply to Pretreatment Program
Click this button to create a pretreatment program with the dilution settings you made.

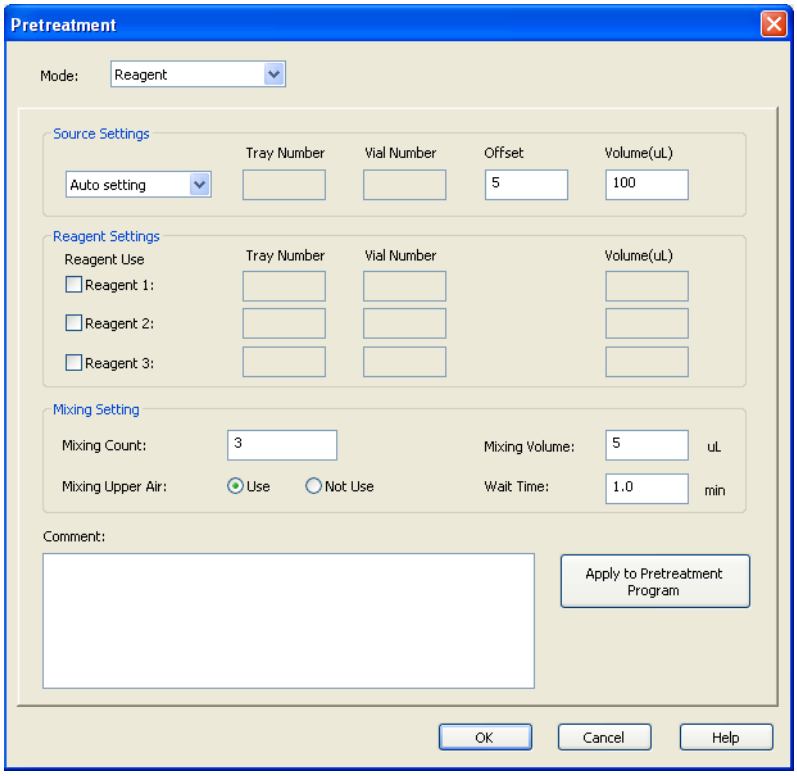
NOTE

If you press the [OK] button without pressing the [Apply to Pretreatment Program] button, the settings are saved but they are not reflected to the pretreatment program.

■ Reagent Mode

Reagent addition included in the pretreatment function can be programmed simply with parameter settings on the screen shown below.

- Reagent mode screen



- Source Settings

Item	Description	Set Range (Units)	Default Value
Source Vial	Select the method for specifying the sample vial that contains a stock solution to which a reagent is to be added.	Auto setting Vial number setting	Auto setting

Item	Description	Set Range (Units)	Default Value
Tray Number (For vial number setting)	Specify the tray number of the sample vial that contains a stock solution to which a reagent is to be added.	[MTP, DWP] 0, 1, 2 [Rack Changer] 0 to 12 [Other] 0, 1	1

Item	Description	Set Range (Units)	Default Value
Vial Number (Vial setting)	Specify the number of the sample vial that contains a stock solution to which a reagent is to be added.	[Control vial rack] 1 to 10 [1.5 mL (105 vials) rack] 1 to 105 [1.5 mL (70 vials) rack] 1 to 70 [Cooling rack 1 mL] 1 to 175 [Cooling rack 4 mL] 1 to 50 [Cooling MTP 96] 1 to 96 [Cooling MTP 384] 1 to 384 [Cooling Deep Well 96] 1 to 96 [Cooling Deep Well 384] 1 to 384 [RackChanger MTP96] 1 to 96 [RackChanger DWP96] 1 to 96 [RackChanger MTP384] 1 to 384 [RackChanger DWP384] 1 to 384 [RackChanger 1.5 mL] 1 to 54	1
Offset (Auto setting)	When auto setting is selected at [Source Vial], set the distance between the injection vial and the source vial.	[1.5 mL (104 vials) rack] 1 to 105 [1.5 mL (70 vials) rack] 1 to 69 [Cooling rack 1 mL] 1 to 174 [Cooling rack 4 mL] 1 to 49 [Cooling MTP 96] 1 to 95 [Cooling MTP 384] 1 to 383 [Cooling Deep Well 96] 1 to 95 [Cooling Deep Well 384] 1 to 383 [RackChanger] 1 to 383	60 35 88 25 48 192 48 192 5
Volume	Set the volume of stock solution to which a reagent is added.	1 to 1000 (μL)	100

• Reagent Settings

Item	Description	Set Range (Units)	Default Value
Reagent Use Reagent 1 to 3	Select whether or not to use reagent 1 to 3.	ON, OFF	OFF
Tray Number	Specify the tray number of the sample vial that contains reagent 1 - 3 to be added.	[MTP, DWP] 0, 1, 2 [Rack Changer] 0 to 12 [Other] 0, 1	1

Item	Description	Set Range (Units)	Default Value
Vial Number	Specify the number of the sample vial that contains reagent 1 - 3 to be added.	[Control vial rack] 1 to 10 [1.5 mL (105 vials) rack] 1 to 105 [1.5 mL (70 vials) rack] 1 to 70 [Cooling rack 1 mL] 1 to 175 [Cooling rack 4 mL] 1 to 50 [Cooling MTP 96] 1 to 96 [Cooling MTP 384] 1 to 384 [Cooling Deep Well 96] 1 to 96 [Cooling Deep Well 384] 1 to 384 [RackChanger MTP96] 1 to 96 [RackChanger DWP96] 1 to 96 [RackChanger MTP384] 1 to 384 [RackChanger DWP384] 1 to 384 [RackChanger 1.5 mL] 1 to 54	1
Volume	Set the volume of reagent 1 to 3 to be added.	1 to 1000 (μL)	100

• Mixing Settings

Item	Description	Set Range (Units)	Default Value
Mixing Count	The instrument performs mixing while aspirating and dispensing the mixed solution through the needle during reagent addition. Set the number of mixing times here.	1 to 10	3
Mixing Volume	The instrument performs mixing while aspirating and dispensing the mixed solution through the needle during reagent addition. Set the volume to be aspirated and dispensed at one time.	1 to (needle loop capacity - 5) (μL)	45
Mixing Upper Air	Select whether or not to perform bubbling by discharging air through the needle during reagent addition.	Use	Use
		Not Use	
Wait Time	Set the wait time before sample injection after completion of pretreatment.	0.1 to 999.0 (min)	1.0

• Comment

Set a comment for addition of a reagent.

NOTE

A comment can be described within 7 lines, 1024 one-byte characters per line.

• Apply to Pretreatment Program

Click this button to create a pretreatment program with the reagent settings you made.

NOTE

If you press the [OK] button without pressing the [Apply to Pretreatment Program] button, the settings are saved but they are not reflected to the pretreatment program.

5.9.3 Example Programs

The pretreatment file "f0" (standard injection operation is programmed as default) contains the programs shown below. Depending on the rinse type setting, the program contents may differ. The programs using default values of the SIL-30AC are shown below.

■ Total Injection Method

#	Program	Description
1	d.rinse	Moves to the rinsing port, dips the needle in the rinsing port, and rinses the external surface of the needle.
2	vial.n rn, sn	Moves to the sample vial of the specified rack number (rn) and vial number (sn).
3	n.strk ns	Moves the needle down by [ns] mm.
4	aspir iv, ss	Aspirates the sample of [iv] μL at [ss] μL/sec.
5	d.rinse	Moves to the rinsing port, dips the needle in the rinsing port, and rinses the external surface of the needle.
6	inj.p	Moves the needle to the injection port.
7	s.inj	Switches the high-pressure valve from load to the injection position and starts analysis.
8	purge.ml mv, rs	Purges the measuring flow line with [mv] μL at [rs] μL/sec.
9	purge.rp rv, rs	Purges the rinse solution in the rinsing port with [rv] μL at [rs] μL/sec.
10	end	End command

■ Loop Injection Method (Partial Loop Method)

#	Program	Description
1	d.rinse	Moves to the rinsing port, dips the needle in the rinsing port, and rinses the external surface of the needle.
2	vial.n rn sn	Moves to the sample vial of the specified rack number (rn) and vial number (sn).
3	air.a av ss	Aspirates air of [av] μL at [ss] μL/sec.
4	n.strk ns	Moves the needle down by [ns] mm.
5	a0 = iv + ev	Defines variable [a0] as [iv + ev1].
6	aspir a0, ss	Aspirates the sample of [a0] μL at [ss] μL/sec.
7	air.a av, ss	Aspirates air of [av] μL at [ss] μL/sec.
8	d.rinse	Moves to the rinsing port, dips the needle in the rinsing port, and rinses the external surface of the needle.
9	inj.p	Moves the needle to the injection port.

#	Program	Description
10	disp 0, ss	Performs pre-pushing.
11	v.load	Switches the high-pressure valve position to the load side.
12	disp iv, ds	Dispenses the sample of [iv] μL to the sample loop at [ds] μL/sec.
13	s.inj	Switches the high-pressure valve from load to the injection position and starts analysis.
14	purge.ml mv, rs	Purges the measuring flow line with [mv] μL at [rs] μL/sec.
15	purge.rp rv,rs	Purges the rinse solution in the rinsing port with [rv] μL at [rs] μL/sec.
16	end	End command (this is not the analysis end time)

■ Loop Injection Method (Full Loop Method)

#	Program	Description
1	d.rinse	Moves to the rinsing port, dips the needle in the rinsing port, and rinses the external surface of the needle.
2	vial.n rn sn	Moves to the sample vial of the specified rack number (rn) and vial number (sn).
3	air.a av ss	Aspirates air of [av] μL at [ss] μL/sec.
4	n.strk ns	Moves the needle down by [ns] mm.
5	a1 = iv * ff	Defines variable [a1] as [iv * ff].
6	a0 = a1 + ev	Defines variable [a0] as [a1 + ev].
7	aspir a0, ss	Aspirates the sample of [a0] μL at [ss] μL/sec.
8	air.a av, ss	Aspirates air of [av] μL at [ss] μL/sec.
9	d.rinse	Moves to the rinsing port, dips the needle in the rinsing port, and rinses the external surface of the needle.
10	inj.p	Moves the needle to the injection port.
11	disp 0, ss	Performs pre-pushing.
12	v.load	Switches the high-pressure valve position to the load side.
13	disp a1, ds	Dispenses the sample of [a1] μL to the sample loop at [ds] μL/sec.
14	s.inj	Switches the high-pressure valve from the load to the injection position and starts analysis.
15	purge.ml mv, rs	Purges the measuring flow line with [mv] μL at [rs] μL/sec.
16	purge.rp rv,rs	Purges the rinse solution in the rinsing port with [rv] μL at [rs] μL/sec.
17	end	End command (this is not the analysis end time)

rn: Rack number
sn: Vial number
ns: Needle stroke
iv: Injection volume
ss: Sample aspiration speed
mv: Measuring flow line purge volume
rs: Rinse solution aspiration speed
rv: Rinse solution volume
av: Air gap
ev: Excess sample aspiration volume
ds: Sample dispensing speed

5.10 Loop Injection Method (Option)

5.10.1 Overview

When the loop injection base kit and a sample loop for loop injection are attached to the SIL-30AC, the injection method can be changed from the standard total injection method to the loop injection method. The loop injection method is effective at reducing gradient delay volume in ultra fast HPLC analysis.

NOTE

The actual capacity of the sample loop for loop injection may differ according to the individual loop as shown below.

Sample Loop for Loop Injection	Volume Error	Remark
5 μL loop	±30 % max.	Internal capacity of the high-pressure valve excluded
20 μL loop	±20 % max.	Internal capacity of the high-pressure valve excluded

5.10.2 Sample Injection Modes

The loop injection method has two kinds of methods: the full loop method of filling the entire sample loop with the sample and injecting it to the HPLC column, and the partial loop method of injecting the set volume of the sample to the HPLC column.

■ Full Loop Method

The full loop method is used to inject a volume of the sample that is equal to the sample loop capacity to the HPLC column. The sample loop must be filled (purged) with the sample. When injecting the sample into the sample loop, the sample volume obtained by multiplying the sample loop capacity by the preload factor will be aspirated. An additional volume of 10 μL (default value) will be required to prevent diffusion during sample aspiration or dispensing. Be sure to check the sample volume that is required in total. The sample volume required for one analysis session with the full loop method is given below. The preload factor may need adjustment according to the viscosity of the sample.

☞ "5.2.2 Parameter Settings Group" P. 80, "5.10.5 Parameters" P. 205

Sample consumption with full loop method using respective sample loops (default setting)

Loop Capacity (μL)	Preload Factor	Required Sample Volume (μL)	Sample Volume Injected to the HPLC Column (μL)
5	3	25	5
20	3	70	20

■ Partial Loop Method

With the partial loop method, the maximum volume of the sample that can be injected into the HPLC column is smaller than the sample loop capacity. The recommended injection volume for each sample loop type is given below. When using a sample loop having a capacity not listed below, consider 70 % of the loop capacity as the upper limit. In addition, depending on the viscosity of the sample, good linearity may not be obtained even if the recommended volume is injected. With the partial loop method, as in the case of the full loop method, an additional volume of 10 μL (default value) will be required to prevent diffusion during sample aspiration or dispensing. Be sure to check the required sample volume.

Recommended injection volume range for each sample loop type

Loop Capacity (μL)	Recommended Injection Volume
5	0.1 - 3.0
20	0.1 - 15.0

■ Difference Between Full Loop Method and Partial Loop Method

There are some differences in sample consumption and reproducibility between these two methods. If you have a sufficient volume of sample and there is no problem in the volume injected into the HPLC column, you are recommended to use the full loop method. The advantages and disadvantages of two methods are listed below.

Loop Injection Type	Reproducibility of Injection Volume		Sample Consumption		Injection Volume	
Full loop method	◎	A sufficient volume of sample is injected into the sample loop so that there is almost no error in injection volume between injections.	△	A volume of sample equivalent to three times (default value) the sample loop capacity plus an additional volume is required. Example: For the 5 μL loop, 25 μL is required for one analysis session.	×	This depends on the sample loop capacity. To change the injection volume, it is necessary to replace the sample loop.
Partial loop method	○	Errors in injection volume between injections may occur compared with the full loop method.	◎	Since only an additional volume is required, consumption can be reduced to a minimum. Example: When injecting 5 μL, 15 μL is required for one analysis session.	◎	The injection volume can be adjustable up to 70 % of the sample loop capacity.

5.10.3 Sample Injection

Sample injection will be made according to the sequence as described below. Note that this section describes the standard sequence and that if one uses the instrument in the multi-rinse mode the sequence may not be the same.

1. Standby (READY)

The mobile phase delivered by the pump flows through the high-pressure valve → sample loop → high-pressure valve to the column.

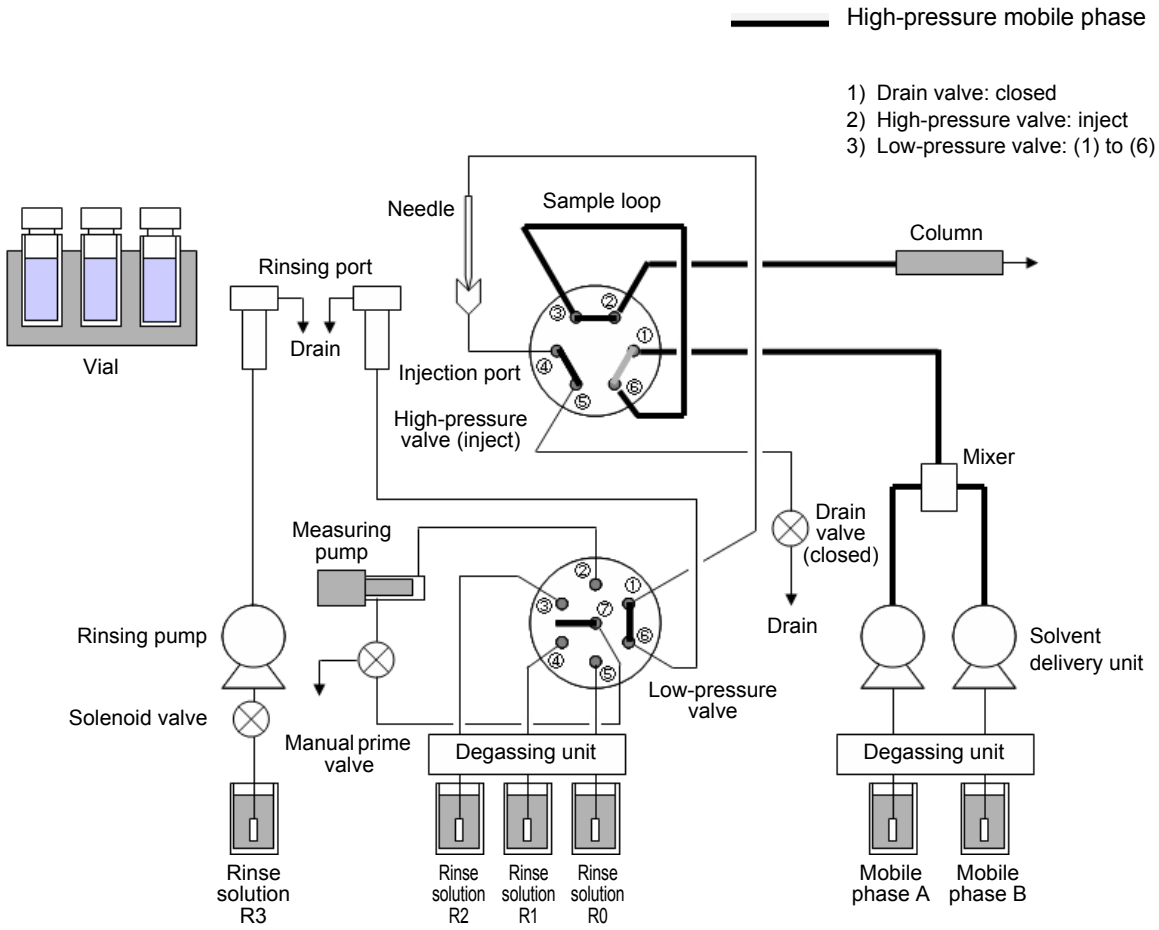


Fig. 5.18

2. Needle movement

The low-pressure valve rotates to the measuring position (210° in the counter-clockwise direction) and the needle moves up.

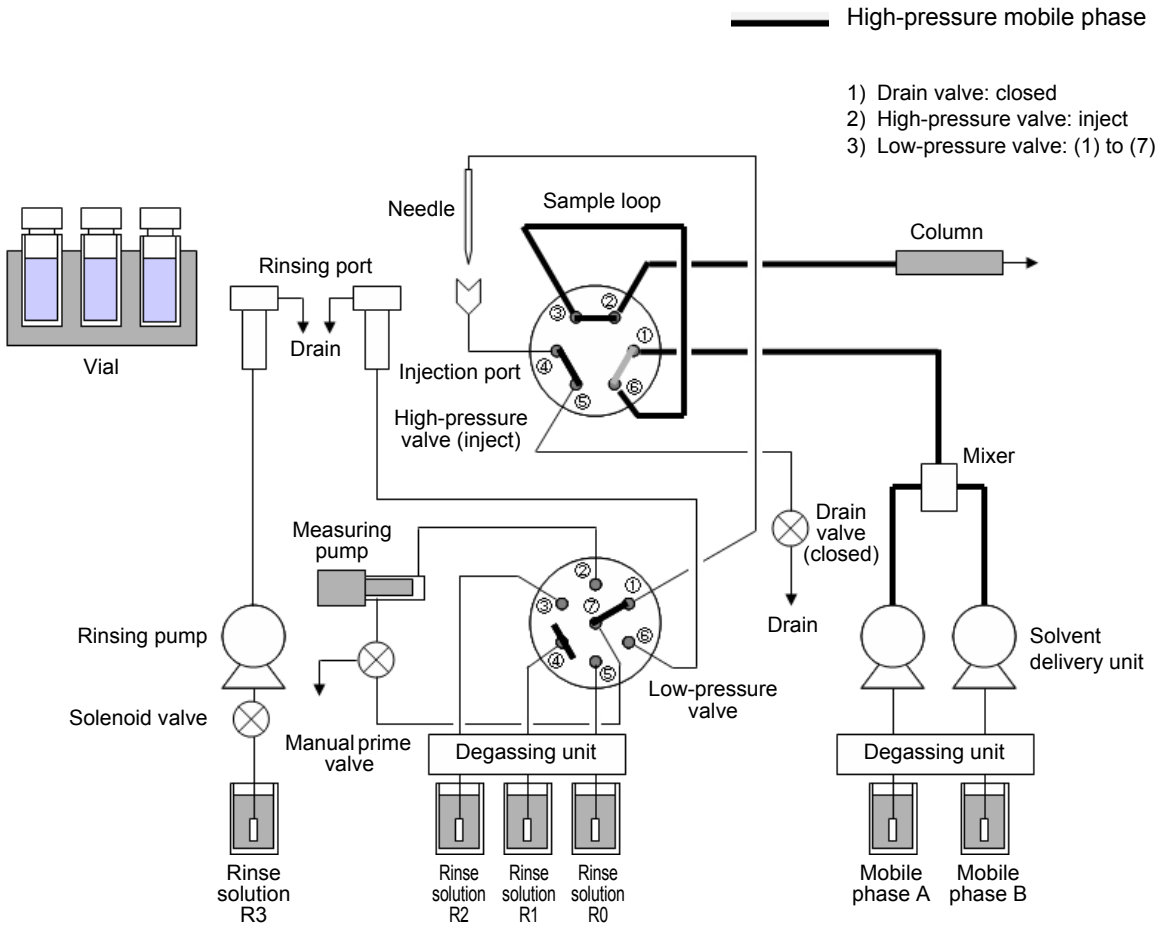


Fig. 5.19

3. External rinsing of needle before sample aspiration

The needle is inserted into the rinsing port and the external surface of the needle is rinsed with the rinse solution in the rinsing port.
It is possible to set to skip external rinsing of the needle before sample aspiration.
Rinsing can be performed with two kinds of rinse solution when a rinsing pump is used.

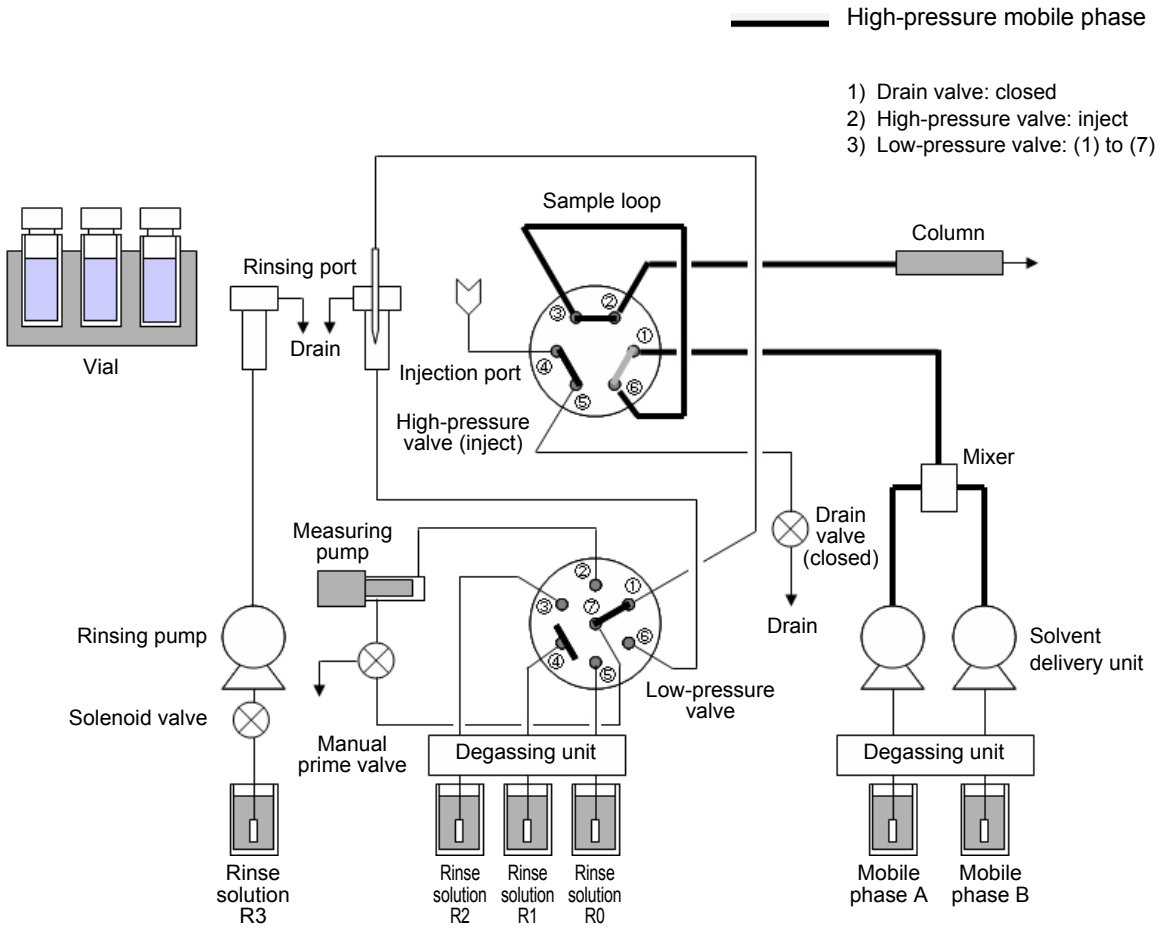


Fig. 5.20

4. Aspiration of air gaps

The needle moves to a position above the sample vial, and aspirates a volume of air set for "air gap" immediately before aspirating the sample.Air gaps are useful to prevent the sample from diffusing in the rinse solution inside the needle.

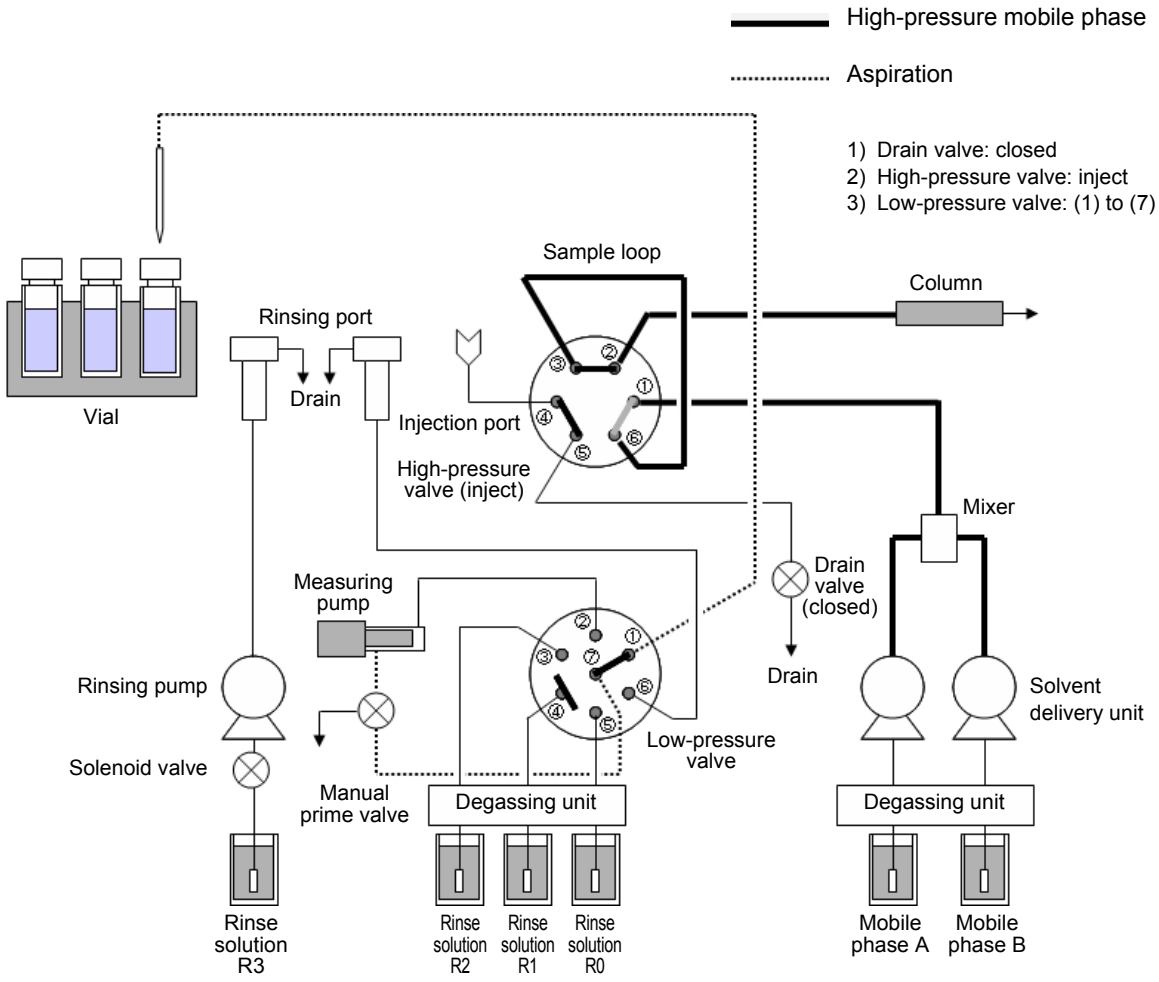


Fig. 5.21

5. Sample aspiration

The needle is inserted in the sample vial. Then the measuring pump plunger starts and aspirates the sample into the needle and the sample loop.

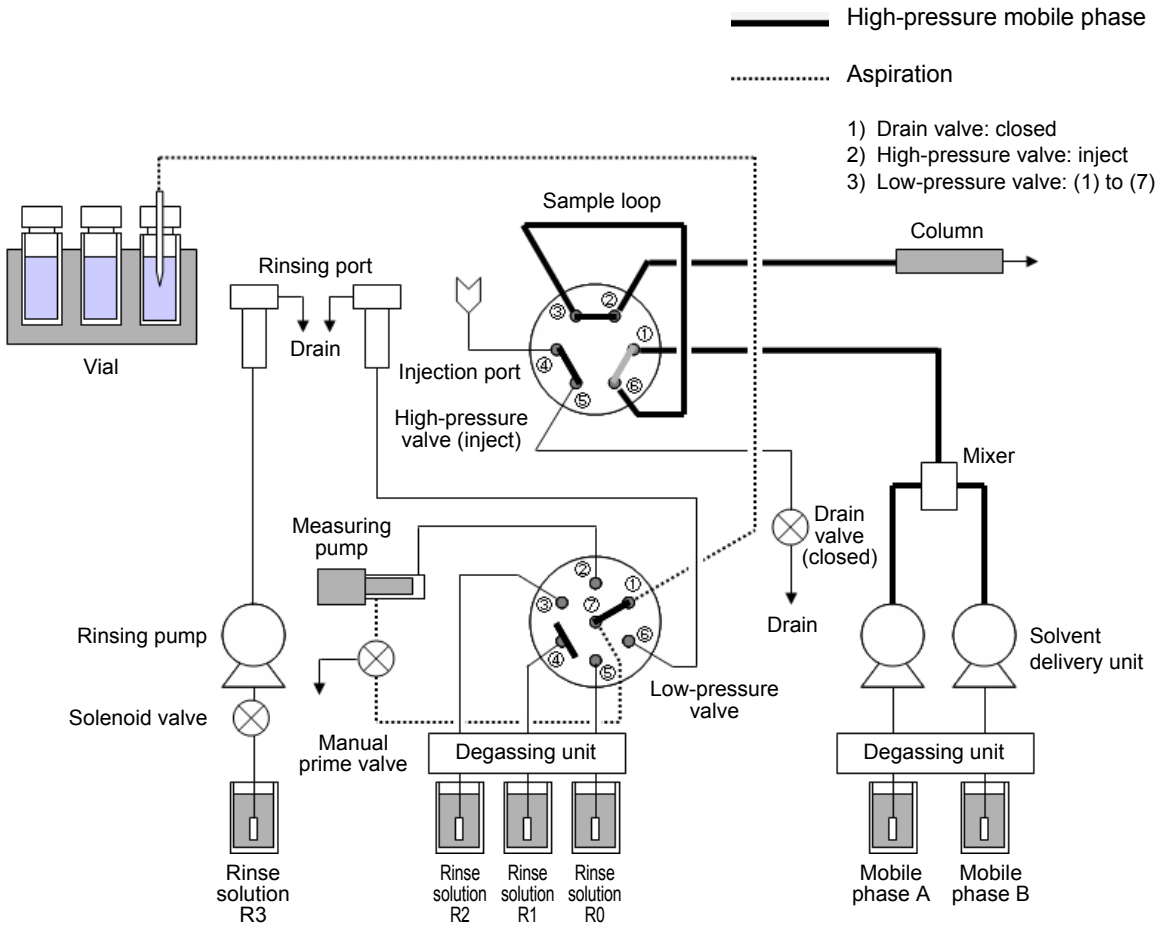


Fig. 5.22

6. Aspiration of air gaps

After sample aspiration, the needle moves up and aspirates air again. The aspirated air is used to prevent the sample from mixing and diffusing in the rinse solution.

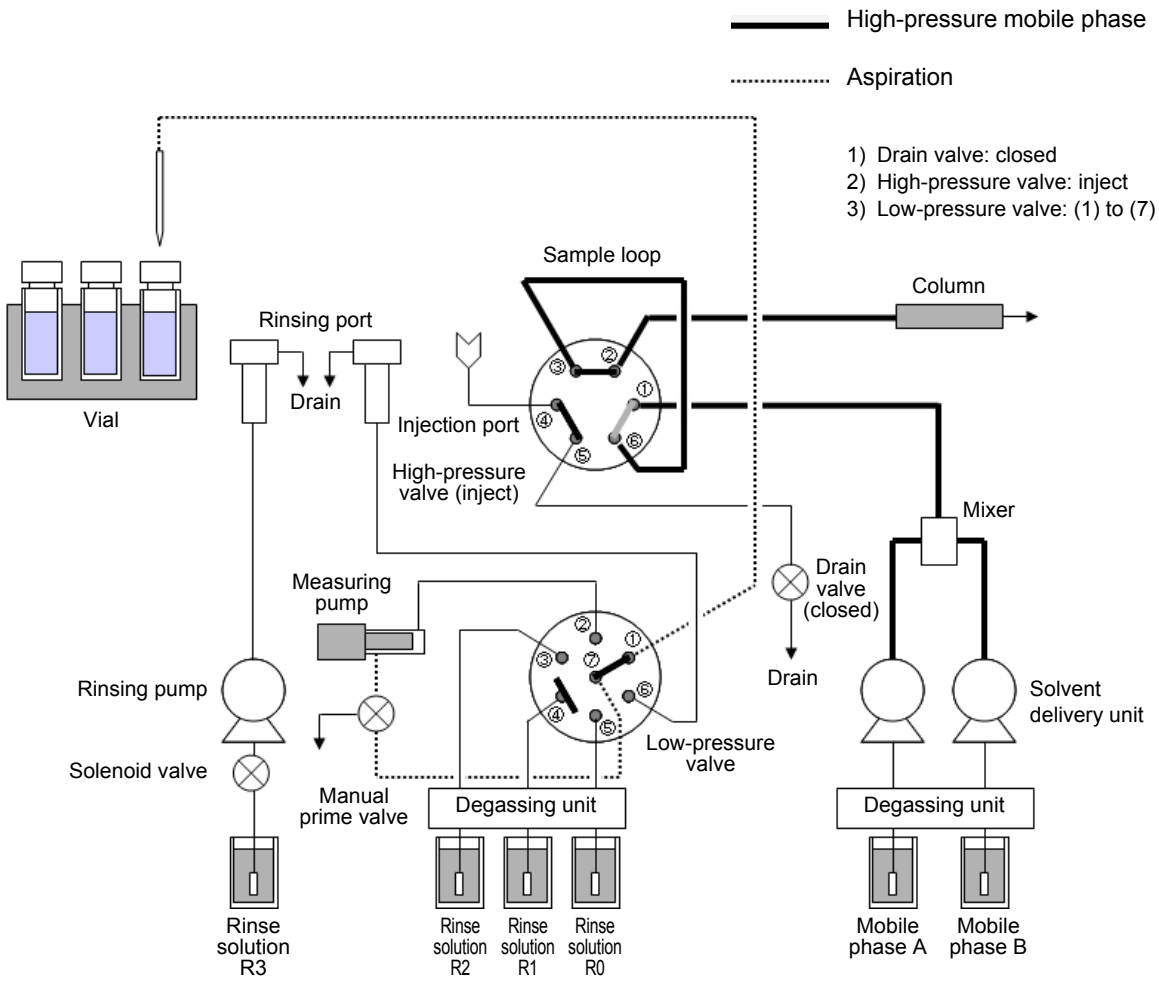


Fig. 5.23

7. External rinsing of needle after sample aspiration

The needle is inserted into the rinsing port and the external surface of the needle is rinsed with the rinse solution in the rinsing port.
It is possible to skip external rinsing of the needle after sample aspiration.
Rinsing can be performed using two kinds of rinse solution when a rinsing pump is used.

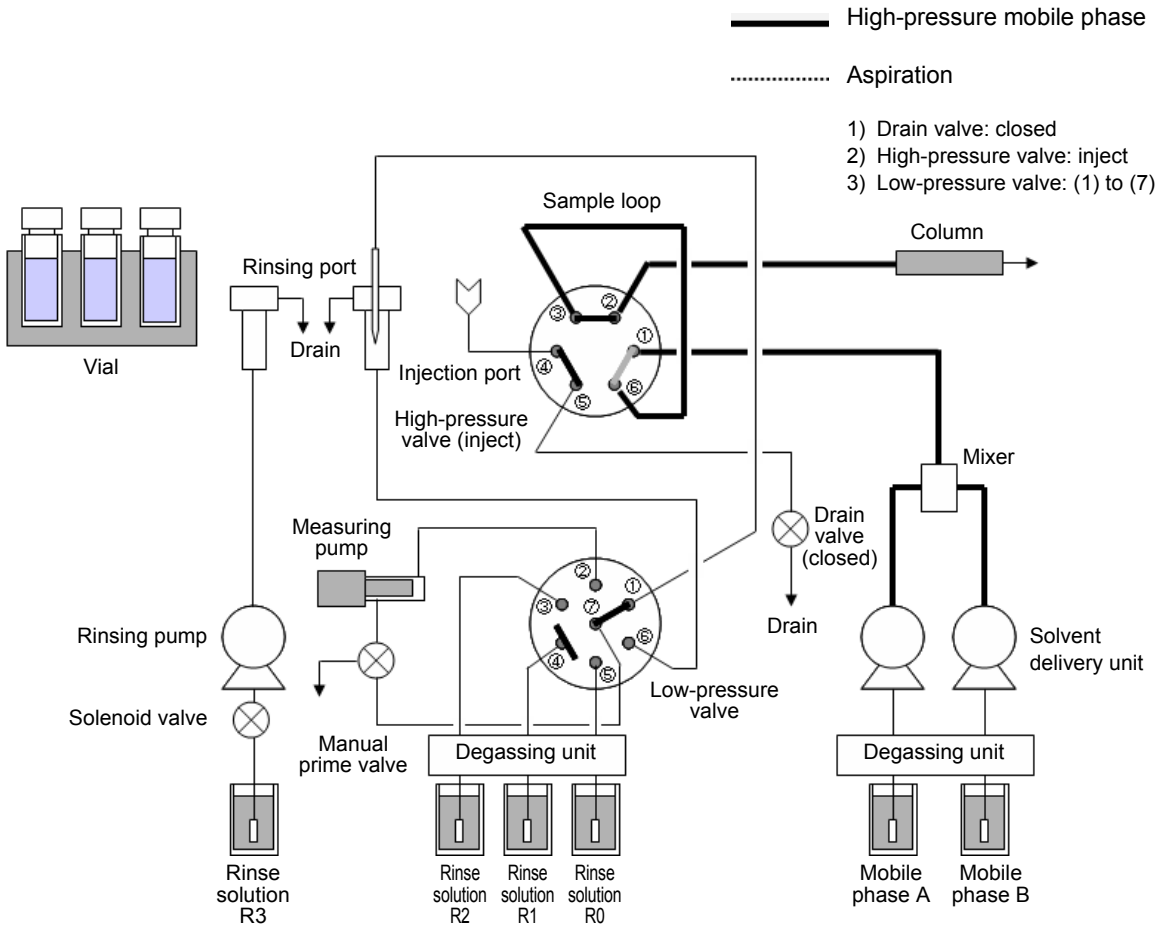


Fig. 5.24

8. Pre-pushing

Pre-pushing is the motion to drain the sample that is subject to diffusion influences from the needle tip. The drain valve is opened to drain the volume of a half the excess volume plus air gap volume using the measuring pump. With the default values, the excess volume is 10 μL and air gap volume is 0.1 μL so that the volume of 5.1 μL is drained.

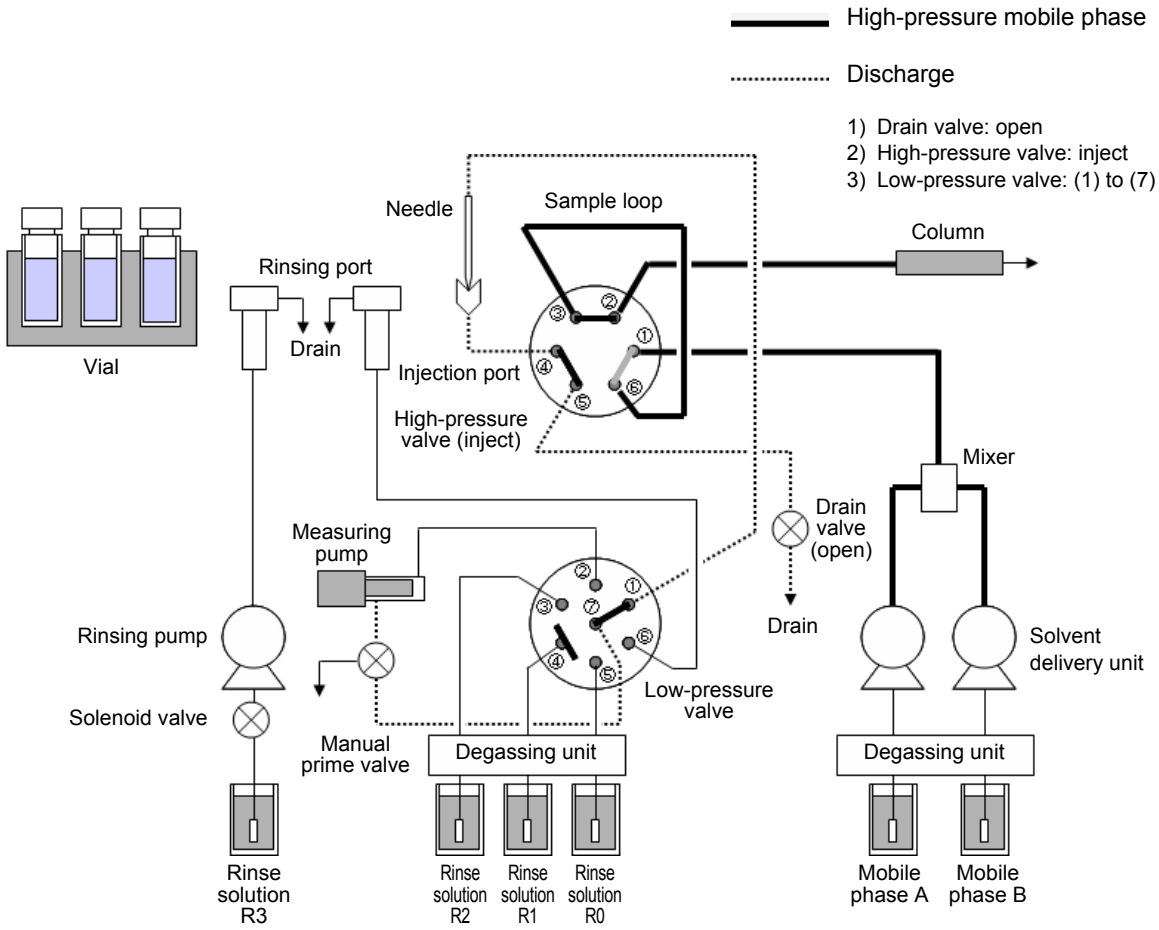


Fig. 5.25

9. Release of pressure from the flow line

The high-pressure valve rotates to the load position (60° in the clockwise direction), and the high-pressure sample loop mobile phase remaining in the sample loop flows through the sample loop → high-pressure valve → drain valve, releasing the pressure from the sample loop.

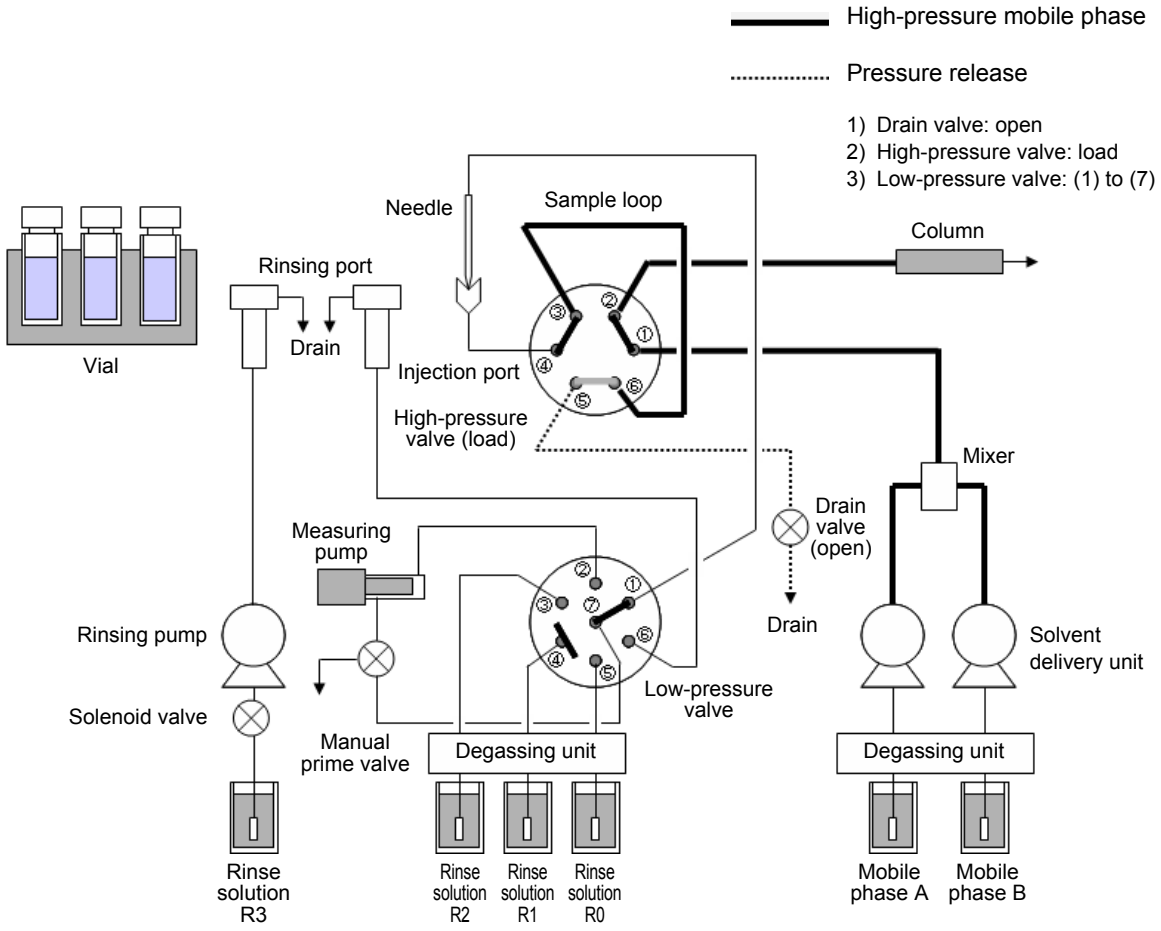


Fig. 5.26

10. Injecting the sample into the sample loop

The measuring pump dispenses sample into the sample loop. The dispensing speed is set to the speed (1.0 μL/sec) slower than the aspiration speed in case that a small I.D. sample loop may be used. Depending on the viscosity of the sample, you may need to decrease the dispensing speed further.

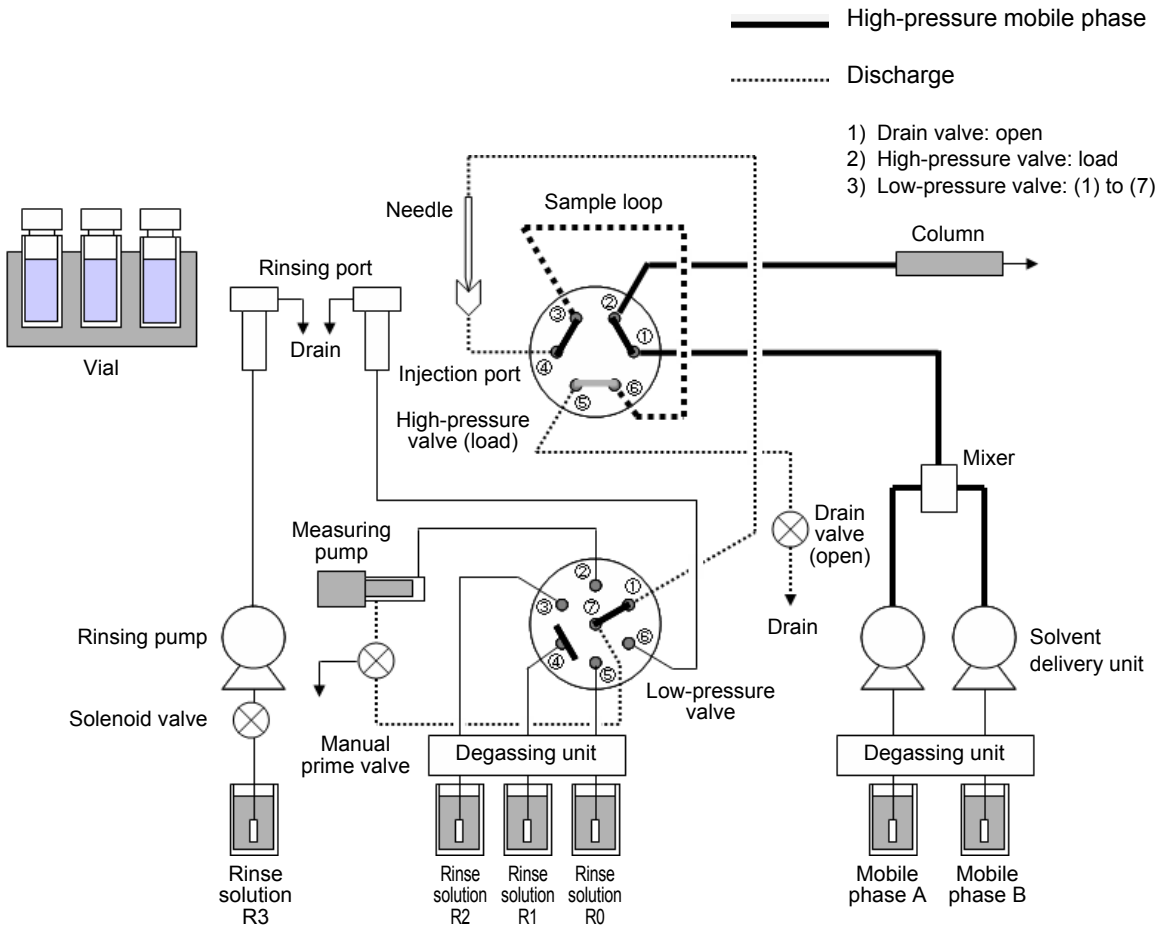


Fig. 5.27

11. Start of analysis (sample injection)

The high-pressure valve rotates to the injection position (60° in the counter-clockwise direction).
The sample is injected into the sample loop and, along with the mobile phase, flows through the high-pressure valve and into the column, where analysis begins.

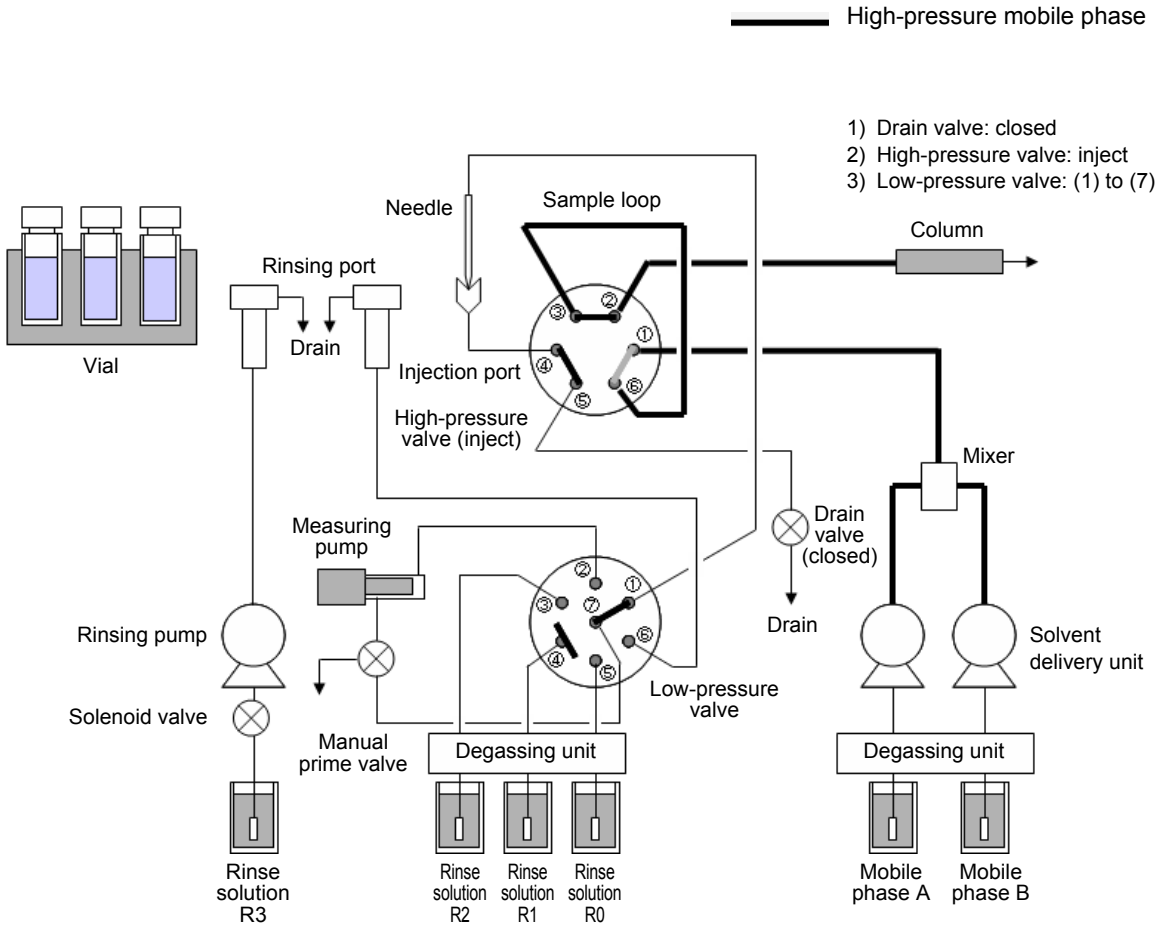


Fig. 5.28

12. Measuring pump home position setting

The drain valve is opened, and the measuring pump dispenses the sample and sets the home position.

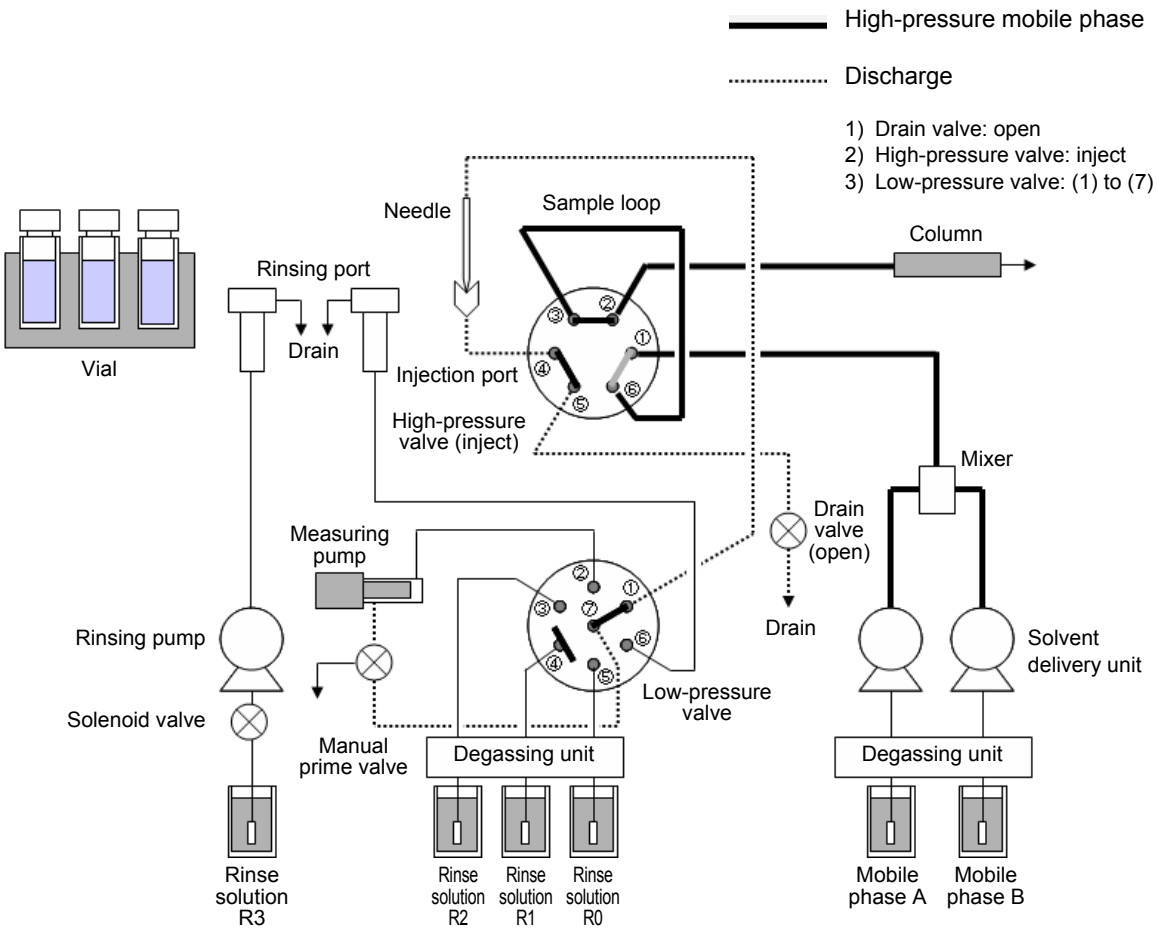


Fig. 5.29

13. Aspiration of rinse solution (R0)

The low-pressure valve rotates to the position (120° in the clockwise direction) where ports 5 and 7 are connected, and the plunger operates for aspiration of rinse solution (R0).

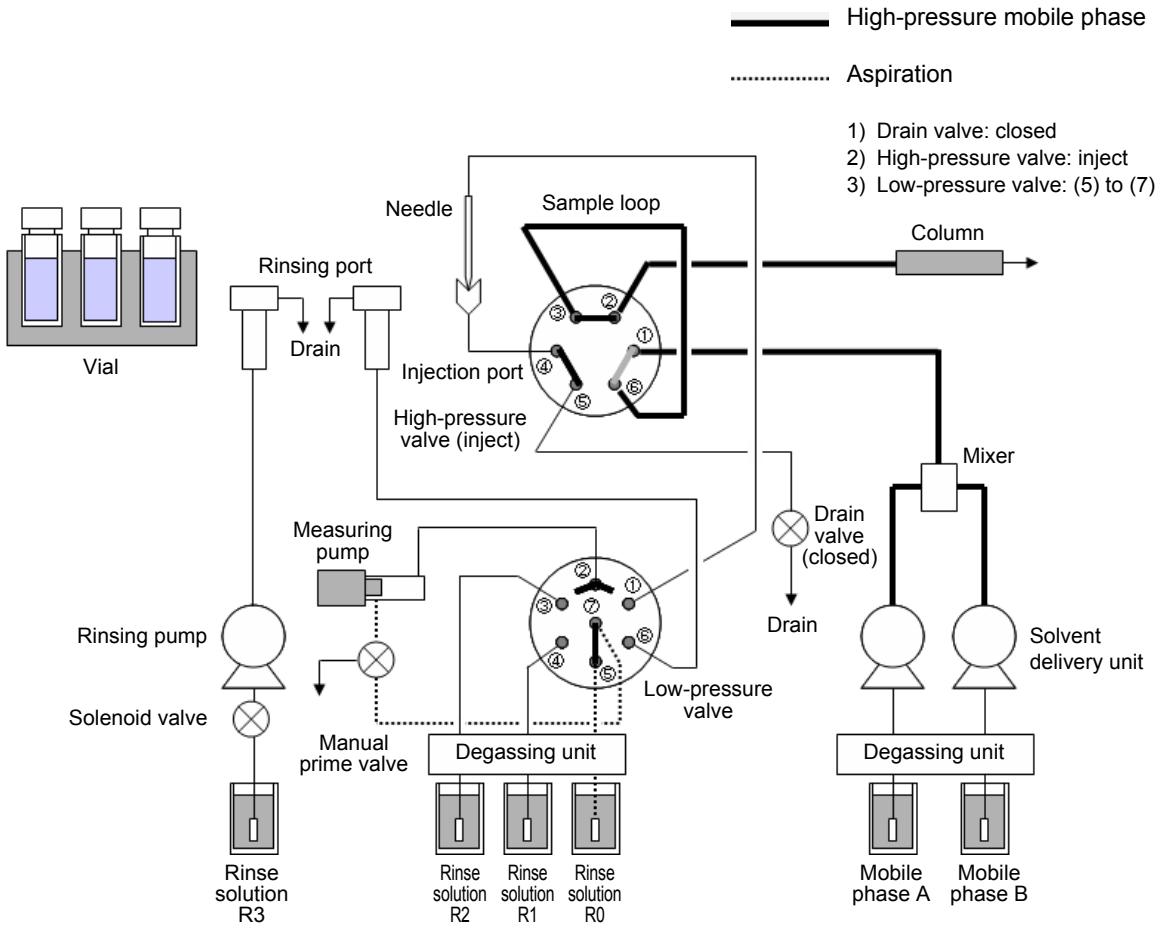


Fig. 5.30

14. Dispensing rinse solution (R0) to the measuring flow line

The low-pressure valve rotates 30° in the clockwise direction, and the plunger of the measuring pump operates for dispensing rinse solution (R0) to the drain valve to purge the measuring flow line.

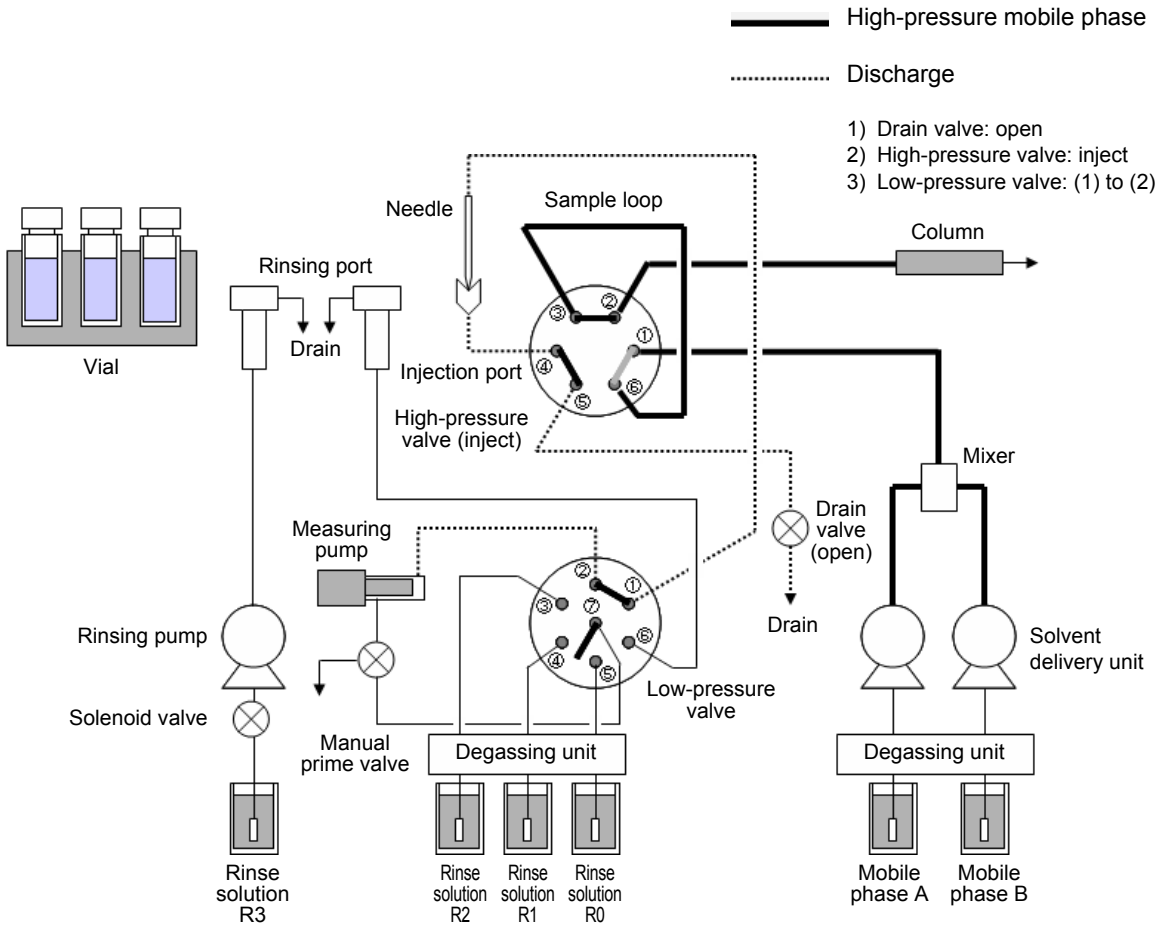


Fig. 5.31

15. Dispensing rinse solution (R0) to the rinsing port

The low-pressure valve rotates 30° in the counter-clockwise direction, and the plunger operates for aspiration of rinse solution (R0). Then the low-pressure valve rotates 60° in the counter-clockwise direction, and the plunger of the measuring pump operates for dispensing rinse solution (R0) to the rinsing port.

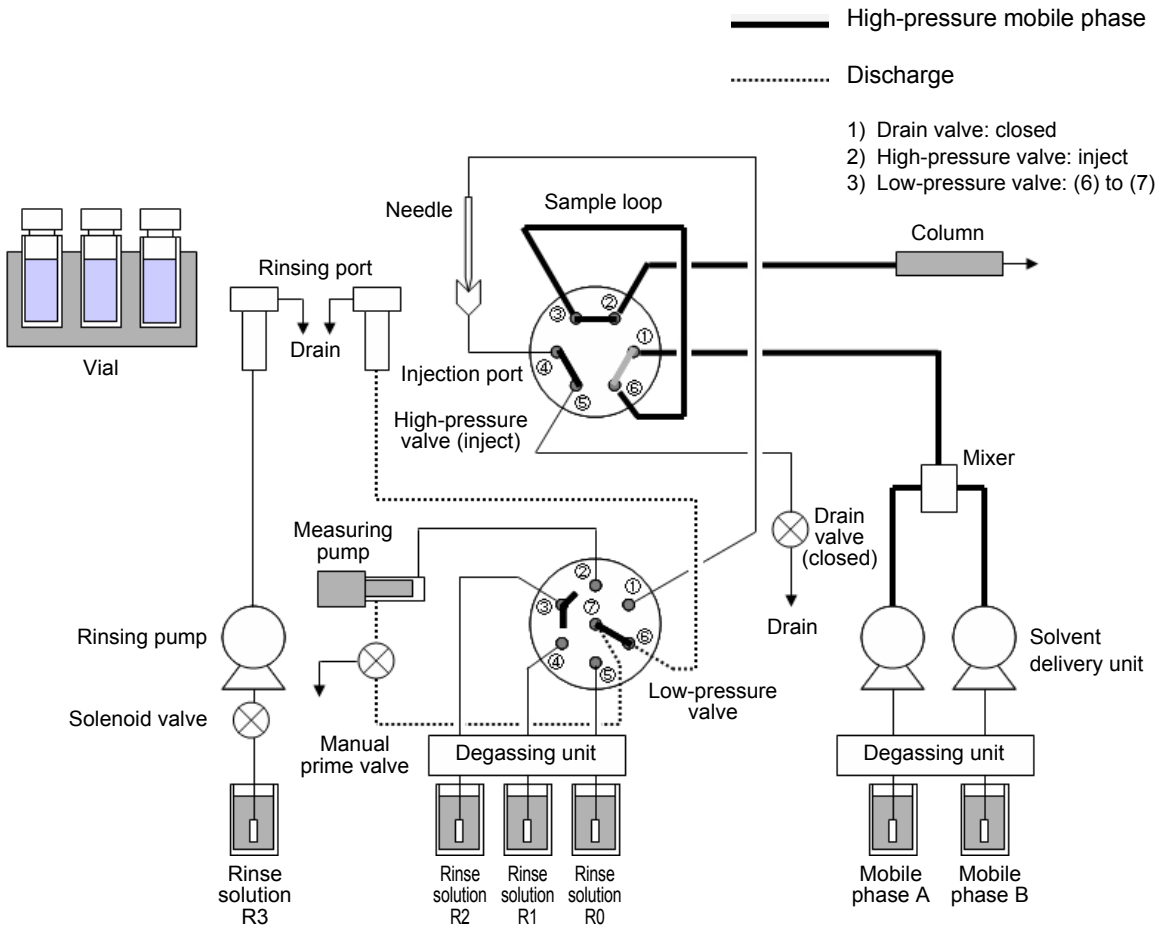


Fig. 5.32

5.10.4 Component Parts

The loop injection base kit (S228-45421-91) comprises the parts listed below. Check the parts against the list after unpacking.

No.	Part Name	Part No.	Q'ty
1	Installation Manual	S228-90782	1
2	SUS tubing, LOOP-LV#1	S228-52513-41	1
3	SUS tubing HP OUT (0.1 × 600 mm)	S228-53184-94	1

NOTE

No sample loop is included in the loop injection base kit. Prepare a sample loop suitable for the injection volume from optional parts shown below.

■ Sample Loops for Loop Injection

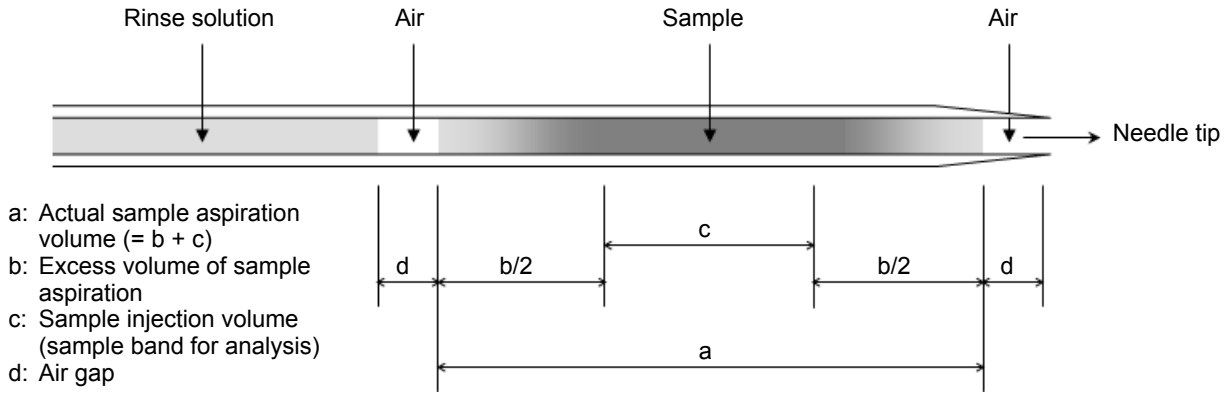
Part Name	Part No.
Sample loop 5 μL	S228-52612-42
Sample loop 20 μL	S228-52612-43

5.10.5 Parameters

With the loop injection method, some parameters differ from those used for the total injection method, which is the standard method of the SIL-30AC. This section explains the parameters of which settings are required when using only the loop injection method. For details about the parameters that are used for the total injection method, refer to "5.2.2 Parameter Settings Group" P. 80 and "5.2.6 System Settings Group" P. 106.

■ Parameters Related to Sample Aspiration

When aspirating the sample, an excess volume of sample as well as air is aspirated before and after the sample to be injected into the sample loop. The following figure illustrates the state of the aspirated sample band.



NOTE

Role of "air gap" and "excess volume of sample aspiration"
If the sample is aspirated into the sample needle filled with rinse solution, it will be diffused into the rinse solution. To prevent the sample from diffusing, air (air gap, d) is aspirated before and after sample aspiration; however, due to the influence of rinse solution remaining on the internal wall of the needle it is not possible to completely eliminate diffusion.
As a measure, the sample is aspirated in excess of the volume to be used for analysis (a), and the diffused portion is discarded and is not injected to the sample loop so that the pure sample band (c) can be injected. The sample volume in excess to be aspirated is called "excess volume of sample aspiration (EXCESS VOL, b).

■ Air Gap (AIR GAP)

Air is trapped (called "air gap") at both ends of the sample band to prevent the aspirated sample from diffusing into the rinse solution. With the default setting of 0.1 μL, an air gap of about 1 mm is inserted before and after the sample inside the needle. If the value is not appropriate, the peak area may become smaller.
☞ "[AIR GAP]" P. 92

■ Excess Volume of Sample Aspiration (EXCESS VOL)

Set the excess volume for sample aspiration that is not injected to the sample loop. For example, if 10 μL is set for the excess volume of sample aspiration, about 5 μL before and after the sample band, that is equal to a half the excess volume, will be discarded. Note that if you have changed any parameter including the sample dispensing speed (DISP SPEED), you may need to adjust this volume as well.
☞ "[EXCESS VOLUME]" P. 92

■ Sample Dispensing Speed (DISP SPEED)

Set the speed for dispensing the sample from the needle to the sample loop. This defaults to 1.0 μL/sec. When using a small I.D. sample loop, the speed must be decreased. Likewise, when using a highly viscous sample, decrease the dispensing speed.
☞ "[DISP SPEED]" P. 85

■ Preload Factor (LOOP FILL FACTOR) (Full Loop Method Only)

With the full loop method, the volume obtained by multiplying the sample loop capacity by the specified factor is aspirated from the sample vial. For example, when "3" is set for the preload factor with a 20 μL sample loop installed, the volume of "60 μL (= 20 μL × 3) + air gap × 2 + excess volume of sample aspiration" is aspirated at one time. The total aspiration volume can be set up to the value of [NEEDLE LOOP VOL]. For example, If you change the preload factor, 100 μL (default value) is set for [NEEDLE LOOP VOL], Note that the total aspiration volume must be less than 100 μL.
☞ "[LOOP FILL FACTOR]" P. 93

■ Loop Capacity (VALVE LOOP VOL)

Set the capacity of sample loop. The sample aspiration volume with the full loop method is determined based on the value set here and the preload factor (LOOP FILL FACTOR). This can be set for the partial loop method; however, it will not affect the analysis.

☞ "5.2.6 System Settings Group" P. 106

6 Troubleshooting

6.1 Troubleshooting and Corrective Action

This section describes the probable causes of problems that can arise, and the corrective actions to be taken to eliminate the causes. For more detailed procedures, refer to the indicated page.
If the problem cannot be resolved even after taking the indicated measures, or if there are problems not included in the following tables, contact your Shimadzu representative.

Symptom	Probable Cause	Corrective Action	Page
Power does not turn ON even after switching ON power.	Power plug disconnected.	• Connect plug correctly.	∗1
	Power cord internal wires are cut.	• Replace with a new cord of the same type.	∗1
	Power supply does not meet specifications for this instrument.	• Use power supply that meets specifications for this instrument.	∗1
	Fuse blown.	• Replace the fuse.	P. 264
No peaks.	Mobile phase is not flowing.	• Check whether pump is functioning normally. Take corrective action as necessary.	∗1 ∗2
	Insufficient amount of sample in vials.	• Add more sample to vials.	
	Injection program incorrect.	• If program is user-written, check and correct the contents.	P. 52
	Sample path injection flow lines clogged.	• Inspect flow lines for clogging. Replace plumbing if clogs are found.	P. 213
	Column performance has deteriorated.	• Check column performance under known analysis conditions. If performance has deteriorated, replace column.	
	Detector is not functioning normally or is not connected.	• Check whether detector is functioning normally. Take corrective action as necessary.	∗1 ∗2

∗1 Refer to the Nexera X2 SYSTEM GUIDE.
∗2 Refer to the instruction manuals for respective instruments.

Symptom	Probable Cause	Corrective Action	Page
Peak retention time fluctuates.	Pump flow rates unstable.	• Check whether pump is functioning normally. Take corrective action as necessary.	∗1 ∗2
	Column temperature is fluctuating.	• Use a column oven.	∗2
	Column performance has deteriorated.	• Check whether column oven is functioning normally. Take corrective action as necessary. • Check column performance under known analysis conditions. If performance has deteriorated, replace column.	
	Composition of mobile phase varies.	• Replace mobile phase, and check composition of new mobile phase.	
	Room temperature fluctuating.	• Install the instrument in a room with minimal temperature variations.	∗1
	There is clogging in the needle or the plumbing.	• Clean the high-pressure valve by a reversing the flow direction. • Clean by reversing the flow direction. If the problem persists, replace the needle or the plumbing.	P. 269 P. 262 P. 271
	Rinse solution used for internal rinsing of the needle remains in the high-pressure flow lines such as the needle or sample loop.	• Set 600 μL or more at [ML PURGE VOL] for purging the measuring flow line, and sufficiently purge the flow line with mobile phase (R0) using the measuring pump after internal rinsing of the needle. • Enable [LOOP S. TM] for sample loop equilibration, and purge the flow line with mobile phase using high-pressure rinsing with the pump after internal rinsing of the needle.	P. 83 P. 98
Peak shapes are abnormal (peaks are broad, or tailing, etc.).	Column performance has deteriorated.	• Check column performance under known analysis conditions. If performance has deteriorated, replace column.	
	Plumbing connections between pump and column were reversed.	• Reconfigure the plumbing.	P. 3
	Dead volume exists between flow line connections.	• Check connections for dead volume. Reconfigure connections to eliminate dead volume.	∗1
	Flow lines leaking.	• See "In Case of Leaking" in this section.	P. 212

∗1 Refer to the Nexera X2 SYSTEM GUIDE.
∗2 Refer to the instruction manuals for respective instruments.

Symptom	Probable Cause	Corrective Action	Page
Peak shapes are abnormal (peaks are broad, or tailing, etc.).	There is clogging in the needle or the plumbing.	• Clean the flow lines by reversing the flow direction. • Clean the interior of the needle with a mobile phase <NDLE FLUSH>. • Clean by reversing the flow direction. If the problem persists, replace the needle or the plumbing.	P. 269 P. 268 P. 262 P. 271
	Rinse solution used for internal rinsing of the needle remains in the high-pressure flow lines such as the needle or sample loop.	• Set 600 μL or more at [ML PURGE VOL] for purging the measuring flow line, and sufficiently purge the flow line with mobile phase (R0) using the measuring pump after internal rinsing of the needle. • Enable [LOOP S. TM] for sample loop equilibration, and purge the flow line with mobile phase using high-pressure rinsing with the pump after internal rinsing of the needle.	P. 83 P. 98
Ghost peaks appear.	No rinse solution.	• Check whether rinse solution is present.	
	The rinsing port is soiled.	• Set 450 μL or more at [DIP-R VOL] for the purge volume in the rinsing port.	P. 94
	Previous mobile phase remains in mobile phase flow lines.	• Clean the flow lines.	P. 63
	Previous rinse solution remains in rinse flow lines.	• Clean the flow lines.	P. 64
	Rinse solution used for internal rinsing of the needle remains in the high-pressure flow lines such as the needle or sample loop.	• Set 600 μL or more at [ML PURGE VOL] for purging the measuring flow line, and sufficiently purge the flow line with mobile phase (R0) using the measuring pump after internal rinsing of the needle. • Enable [LOOP S. TM] for sample loop equilibration, and purge the flow line with mobile phase using high-pressure rinsing with the pump after internal rinsing of the needle.	P. 83 P. 98

∗1 Refer to the Nexera X2 SYSTEM GUIDE.
∗2 Refer to the instruction manuals for respective instruments.

Symptom	Probable Cause	Corrective Action	Page
Poor reproducibility.	Flow lines are not being rinsed sufficiently, or there is no rinse solution.	• [PURGE] or [RINSE] the flow lines. • Add rinse solution. • Operate [PUMP HEAD FLUSH].	P. 63
	Composition or flow rate of mobile phase varies	• Check pump and mobile phase.	∗1 ∗2
	Needle seal is worn.	• Replace the needle seal.	P. 229
	Flow lines leaking.	• See "In Case of Leaking" in this section.	P. 212
	Room temperature is fluctuating.	• Install the instrument in a room with minimal temperature fluctuations.	∗1
	Column performance has deteriorated.	• Check column performance under known analysis conditions. If performance has deteriorated, replace column.	
Baseline drifting.	Flow lines dirty.	• Thoroughly clean the instrument and detector flow lines.	P. 63
	Faulty detector.	• Check whether detector is functioning normally. Take corrective action as necessary.	∗1 ∗2
	Room temperature is fluctuating.	• Install the instrument in a room with minimal temperature fluctuations.	∗1
	Flow rates fluctuate.	• Check whether pump is functioning normally. Take corrective action as necessary.	∗1 ∗2
Large pressure fluctuation when high-pressure valve is switched.	High-pressure valve is clogged.	• Disassemble and clean high-pressure valve. • If disassembly and cleaning does not unclog the valve, replace the rotor and stator seal.	P. 247
	High-pressure valve does not rotate to the correct positions.	• The message shown below is displayed. Contact your Shimadzu representative.	P. 222
	Flow lines clogged.	• Rinse the flow lines with reverse flow. • Check the flow lines and replace the tubing if any clogging is found.	P. 269 P. 271
Column inlet pressure is too high.	Column is clogged.	• Check column pressure. If column is clogged, replace it.	
	Flow lines clogged.	• Rinse the flow lines with reverse flow. • Inspect the flow line. Clean or Replace any clogged plumbing.	P. 269 P. 271

∗1 Refer to the Nexera X2 SYSTEM GUIDE.
∗2 Refer to the instruction manuals for respective instruments.

■ In Case of Leaking

Symptom	Probable Cause	Corrective Action	Page
High-pressure valve leaking.	Rotor and stator sealing ability has deteriorated.	• Replace the rotor seal, inspect the stator and replace it if necessary. ∗3	P. 247
Low-pressure valve leaking.	Rotor and stator sealing ability has deteriorated.	• Inspect the rotor and stator and replace them if necessary. ∗3	P. 242
Flow line connections leaking.	Male nuts are loose or stripped.	• Tighten male nuts. • If tightening does not stop the leak, replace the male nuts and ferrules.	∗1

∗1 Refer to the Nexera X2 SYSTEM GUIDE.

∗3 The high-pressure valve stator is made of specially hard coated material, and the low-pressure valve stator is made of ceramic. If there are no visible scratches, they do not need to be replaced.

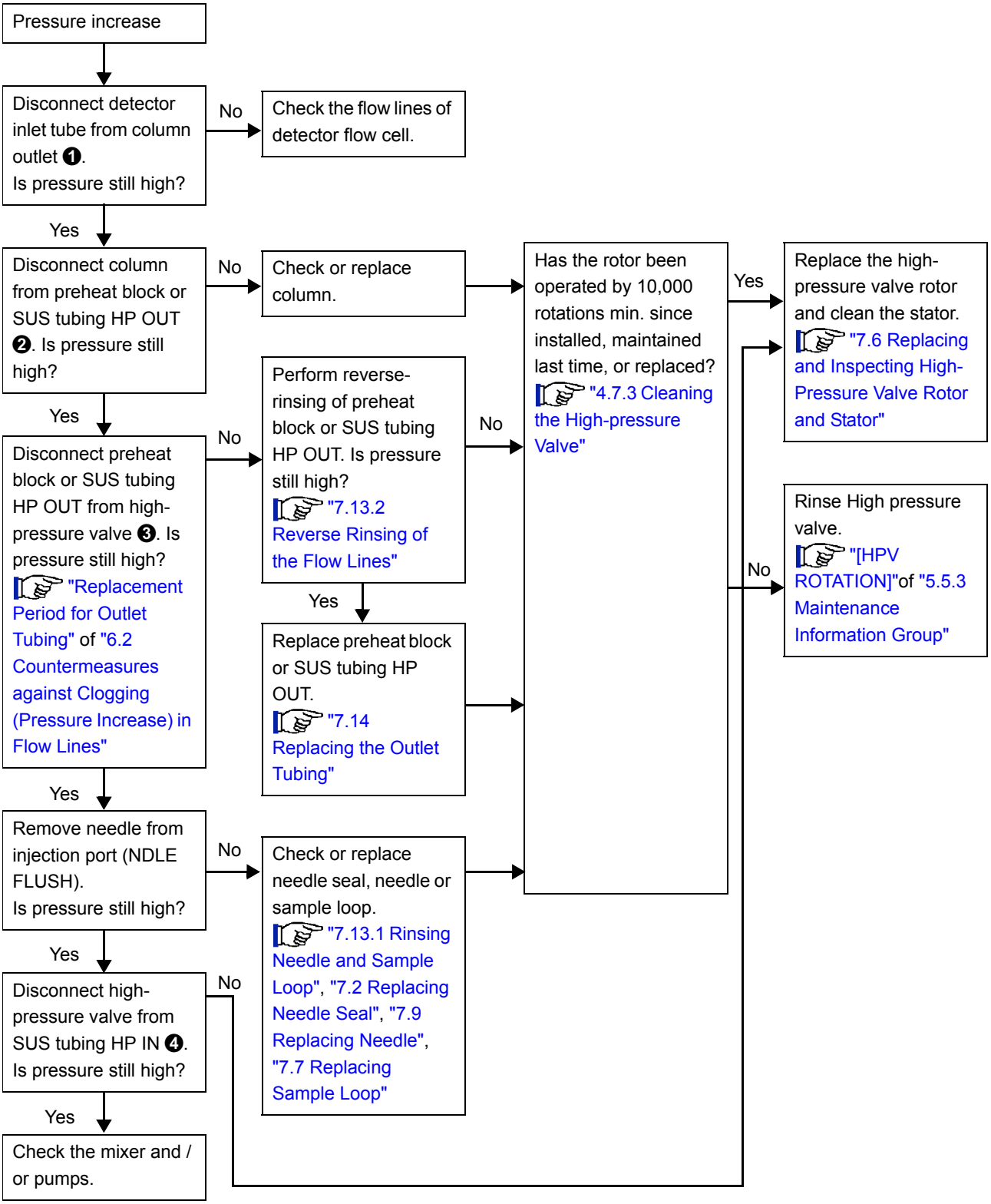
6.2 Countermeasures against Clogging (Pressure Increase) in Flow Lines

If the pump pressure has noticeably increased, it may be because some portions of the flow lines have become clogged. In such cases, disconnect tubing one by one from downstream as shown in the diagram below. Evaluate the pressure changes to find the clogged portion.

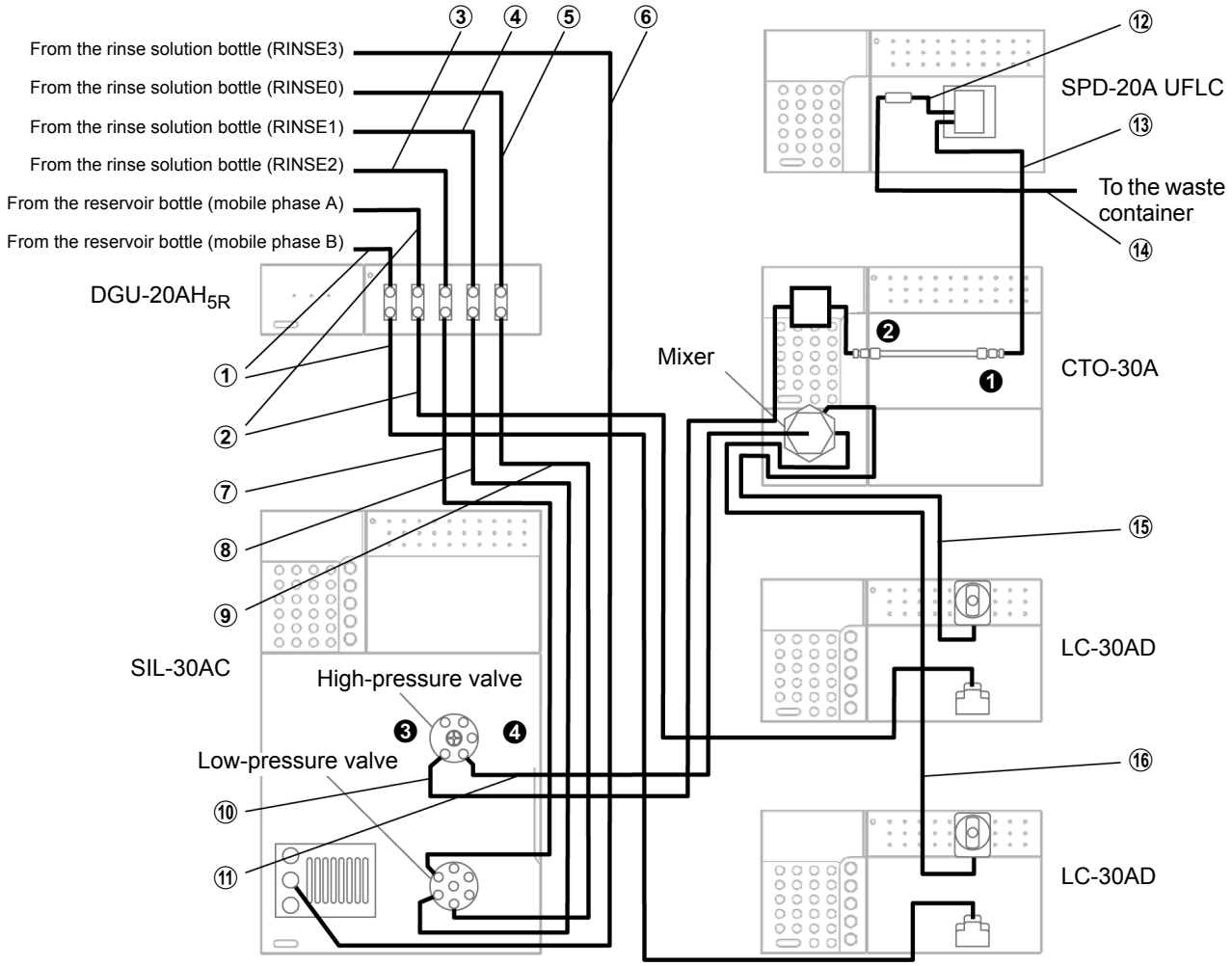
⚠ CAUTION

Before removing the SUS tubing HP IN, SUS tubing HP OUT, sample loops, needles or needle seals from the high-pressure flow line, be sure to stop pumping at the solvent delivery unit. Before removing high-pressure flow line parts, ensure that the pump pressure of the mobile phase displayed on the screen of the solvent delivery unit is reduced to zero.

Always wear protective gloves and goggles when removing high-pressure flow line parts. If solvent gets into the eyes, blindness could result. Should solvent get into the eyes, flush immediately with large amounts of water and get medical attention.


Pressure increase
Disconnect detector inlet tube from column outlet ❶. Is pressure still high?
No
Check the flow lines of detector flow cell.
Yes
Disconnect column from preheat block or SUS tubing HP OUT ❷. Is pressure still high?
No
Check or replace column.
Yes
Disconnect preheat block or SUS tubing HP OUT from high-pressure valve ❸. Is pressure still high?
"Replacement Period for Outlet Tubing" of "6.2 Countermeasures against Clogging (Pressure Increase) in Flow Lines"
No
Perform reverse-rinsing of preheat block or SUS tubing HP OUT. Is pressure still high?
"7.13.2 Reverse Rinsing of the Flow Lines"
Yes
Replace preheat block or SUS tubing HP OUT.
"7.14 Replacing the Outlet Tubing"
Yes
Remove needle from injection port (NDLE FLUSH). Is pressure still high?
No
Check or replace needle seal, needle or sample loop.
"7.13.1 Rinsing Needle and Sample Loop", "7.2 Replacing Needle Seal", "7.9 Replacing Needle", "7.7 Replacing Sample Loop"
Yes
Disconnect high-pressure valve from SUS tubing HP IN ❹. Is pressure still high?
No
Yes
Check the mixer and / or pumps.
Has the rotor been operated by 10,000 rotations min. since installed, maintained last time, or replaced?
"4.7.3 Cleaning the High-pressure Valve"
No
Yes
Replace the high-pressure valve rotor and clean the stator.
"7.6 Replacing and Inspecting High-Pressure Valve Rotor and Stator"
No
Rinse High pressure valve.
"[HPV ROTATION]"of "5.5.3 Maintenance Information Group"

■ Two-liquid High-pressure Gradient System (with CTO-30A)



No.	Part Name	Part No.	Package
①	Suction filter ASSY	S228-45708-91	LC-30AD
②	Suction filter ASSY	S228-45708-91	LC-30AD
③	Suction filter ASSY	228-45708-93	SIL-30AC
④	Suction filter ASSY	228-45708-93	SIL-30AC
⑤	Suction filter ASSY	228-45708-93	SIL-30AC
⑥	Suction filter ASSY	228-45708-93	SIL-30AC
⑦	SUS tubing, DGU-LV#3	S228-52448-42	SIL-30AC
⑧	SUS tubing, DGU-LV#4	S228-52449-42	SIL-30AC
⑨	SUS tubing, DGU-LV#5	S228-52450-42	SIL-30AC
⑩	Preheat block	S228-52597-43	CTO-30A
⑪	SUS tubing HP IN (0.3 × 300 mm)	S228-53184-92	SIL-30AC
⑫	Cell outlet tube	–	SPD-20A
⑬	Cell inlet tube	–	SPD-20A

No.	Part Name	Part No.	Package
⑭	Tubing for plumbing	S228-18495-06	SPD-20A
⑮	SUS tubing HP (0.3 × 700 mm)	S228-53184-93	LC-30AD
⑯	SUS tubing HP (0.3 × 700 mm)	S228-53184-93	LC-30AD

NOTE

Tubing ⑥, ⑦, ⑧, ⑨, ⑩, ⑪, ⑫, ⑬ is provided to the instrument when it is packaged.

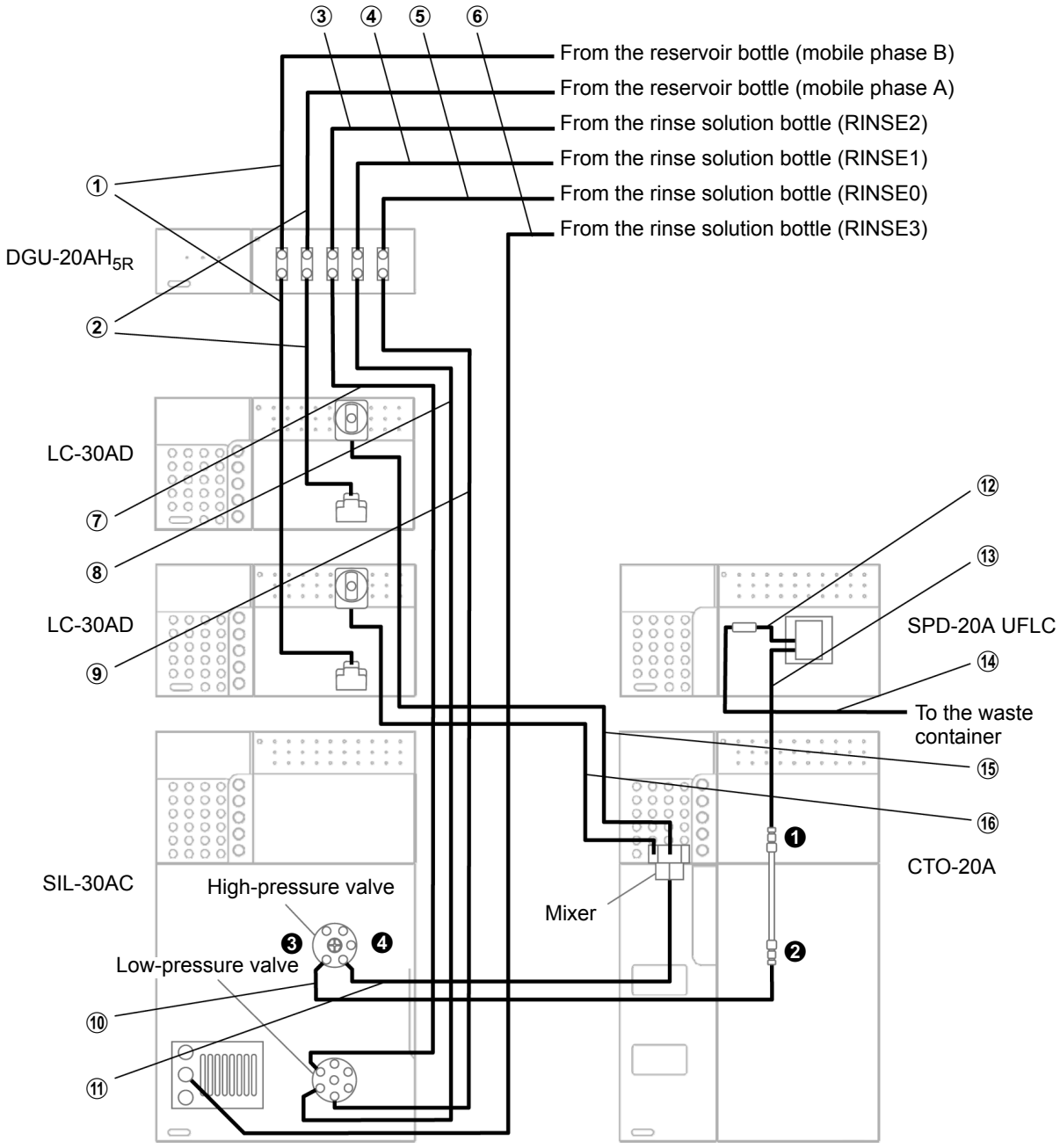
Pass ① and ② through the tubing cover on the right side of the SIL-30AC.

Pass ⑮ and ⑯ in almost the center of the front of the LC-30AD.

Pass ⑩, ⑪, ⑮ and ⑯ through the hole in the bottom on the left side of the CTO-30A.

Pass ⑬ through the hole in the top of the CTO-30A and under the SPD-20A UFLC.

■ Two-liquid High-pressure Gradient System (with CTO-20A)



No.	Part Name	Part No.	Package
①	Suction filter ASSY	S228-45708-91	LC-30AD
②	Suction filter ASSY	S228-45708-91	LC-30AD
③	Suction filter ASSY	228-45708-93	SIL-30AC
④	Suction filter ASSY	228-45708-93	SIL-30AC
⑤	Suction filter ASSY	228-45708-93	SIL-30AC
⑥	Suction filter ASSY	228-45708-93	SIL-30AC

No.	Part Name	Part No.	Package
⑦	SUS tubing, DGU-LV#3	S228-52448-42	SIL-30AC
⑧	SUS tubing, DGU-LV#4	S228-52449-42	SIL-30AC
⑨	SUS tubing, DGU-LV#5	S228-52450-42	SIL-30AC
⑩	SUS tubing HP OUT (0.1 × 600 mm)	S228-53184-91	SIL-30AC
⑪	SUS tubing HP IN (0.3 × 300 mm)	S228-53184-92	SIL-30AC
⑫	Cell outlet tube	–	SPD-20A
⑬	Cell inlet tube	–	SPD-20A
⑭	Tubing for plumbing	S228-18495-06	SPD-20A
⑮	SUS tubing HP (0.3 × 700 mm)	S228-53184-93	LC-30AD
⑯	SUS tubing HP (0.3 × 700 mm)	S228-53184-93	LC-30AD

NOTE

Tubing ⑥, ⑦, ⑧, ⑨, ⑩, ⑪, ⑫, ⑬ is provided to the instrument when it is packaged.

6.2.1 Causes and Countermeasures against Clogging

Possible causes and countermeasures against clogging that is likely to occur with LC are given below. The following is the summary of the countermeasures for each item. When clogging is found, check those points and take necessary measures.

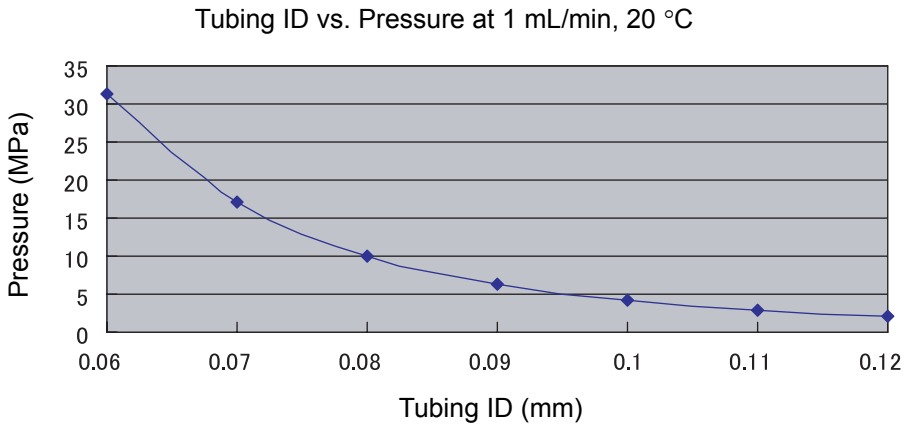
Cause of Clogging	Countermeasures	Necessary Apparatus
Insoluble matters in the mobile phase	Mobile phases should be filtered prior to use especially when buffer solutions are used, because insoluble matters in the salts can cause clogging. It is recommended that customers filter buffer solutions using a commercially available filtering apparatus and 0.22 μm membrane filters or less. This is also important for the column protection.	• Filtering apparatus with a filter holder • Membrane filter (pore size 0.22 μm or less)
Insoluble matters in the sample	Similarly, insoluble matter in the sample can cause clogging in the system plumbing or column. If the sample is cloudy or if particulates are visible, filter the sample solution using a commercially available disposable filter before analysis. Such filters are available also from Shimadzu.	• Disposable filter (Refer to p.114 - 115 of Shimadzu Column Catalogue for details.)
Suspended particles or dust in the environment	Dust in the environment can enter the flow line from the injection port and cause clogging. Normally this is less likely to occur because the instrument is operated with the front door closed. Avoid dusty areas when working on the instrument with the front door open for the purpose of maintenance, etc.	
Flakes of the needle seal in the injection port	If a deviation of the needle position occurs by any reason, the needle can scrape the needle seal and resulting flakes may cause clogging. The needle position can be checked through upward and downward motion by pressing [TEST INJ PORT] ("[TEST INJ PORT]" P. 105). In case a deviation of the position is found, perform teaching of the needle on the injection port.	
Flakes of vial septa	Flakes of vial septa can cause clogged plumbing. Various kinds of septum are commercially available and they differ in material and coating. Shimadzu's genuine septa have passed consecutive injection tests and organic solvent resistance tests. However, due to price, some customers use septa from different vendors. Although septa are usually laminated with a film (PTFE for example) resistant of organic solvents to prevent flaking, some flakes are still possible when the needle pierces the septum. These flakes can eventually clause clogging. If using septa from sources other than Shimadzu and clogging is a problem, it is recommended that only Shimadzu's genuine septa and vials be used.	

Cause of Clogging	Countermeasures	Necessary Apparatus
Particles scraped off the rotor seal of the high-pressure valve	As the rotation of the high-pressure valve is repeated, particles are produced from the initial wear of the PEEK rotor seal. These particles are so fine that they seldom causes clogging in the plumbing, but they can result in clogging at the column inlet. When replacing the PEEK rotor seal with a new one, refer to "7.6 Replacing and Inspecting High-Pressure Valve Rotor and Stator". After completing the replacement, rotate the high-pressure valve to remove any particles caused by initial wear. Although wide-bore tubing can be used for plumbing to avoid clogging, the particles may cause clogging at the column inlet. Consequently this is not an optimal solution, and it is therefore not a desirable countermeasure. For this reason, it is advised that wide bore tubing not be used to correct this problem. These precautions are also the case when this instrument is connected to another vendor's HPLC. Note, similar attention should be paid to other vendor's pumps and plumbing, and necessary countermeasures taken. Clean the high-pressure valve after approximately 10,000 injections.	• Methanol or 2-propanol
Particles from the pump system	The following are possible causes of particles being introduced from the pump system. (1) Particles on the flow line parts, such as the suction filter and line filter. (2) Particles from worn plunger seals. To remove particles (1), especially when these parts are replaced, rinse the pump with methanol or isopropanol for 15 minutes at 5 mL/min prior to making the connection with the SIL. The particles (2) are trapped in the line filter of the pump; replace the line filter at a regular intervals.	• Methanol or 2-propanol
Particles inside of tubing	If a high pressure of 66 MPa or above is applied to the tubing, be sure to use the tubing parts dedicated to the Nexera X2. Do not cut the Nexera X2 tubing parts when using them. Regardless of the pressure level, do not use any stainless steel tube that is cut to length, for example, using a file (for cutting stainless steel tubes). Metal powders produced during cutting will enter the flow line and could lead to pressure buildup or clogging of the tubing. When plumbing tubing has been replaced, rinse the tubing thoroughly before making connections. Rinse with methanol or isopropanol for 15 minutes at 5 mL/min.	• Methanol or 2-propanol

■ Replacement Period for Outlet Tubing

The chart below shows the relationship between the tubing ID and the pressure when pure water is pumped at 1 mL/min through the tubing (600 mm long). The pressure can be as high as 6 MPa without connecting the column to the SUS tubing HP OUT (0.1 mm ID, 600 mm long). If the pressure exceeds 10 MPa, the tubing becomes clogged. In such cases, reverse-rinse the tubing to remove clogs. If the clogs cannot be removed by reverse-rinsing, replace the outlet tubing with a new one.

☞ "7.13.2 Reverse Rinsing of the Flow Lines" P. 269, "7.14 Replacing the Outlet Tubing" P. 271



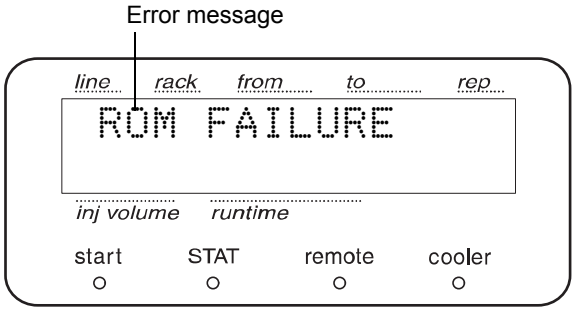
6.3 Error Messages

The instrument has several diagnostic functions. Upon detection of a problem, an alarm sounds and an error message appears on the display panel.
The following list describes the error messages along with the causes and corrective actions.

NOTE

Each message is classified into the following three types.
The type is indicated under the type column.

Fatal: The instrument stops operation.
The error message is not cleared by pressing CE.
Alarm: The instrument stops operation.
Press CE to clear the error message.
Warning: The instrument does not stop operation.
Press CE to clear the error message.



Error Message	Type	Cause and Action
ROM FAILURE (ROM error)	Fatal	Cause: ROM error (electronic failure). Action: Turn power OFF and contact your Shimadzu representative.
RAM FAILURE (RAM error)	Fatal	Cause: RAM error (electronic failure). Action: Turn power OFF and contact your Shimadzu representative.
ERR NDLE HOME X (Needle X motor slip error)	Fatal	Cause: Needle's X-axis (sideways) movement is incorrect. Action: Turn the power OFF and contact your Shimadzu representative.
ERR NDLE HOME Y (Needle Y motor slip error)	Fatal	Cause: Needle's Y-axis (forward/backward) movement incorrect. Action: Turn the power OFF and contact your Shimadzu representative.
ERR NDLE HOME Z (Needle Z motor slip error)	Fatal	Cause: Needle's Z-axis (up/down) movement incorrect. Action: Turn the power OFF and contact your Shimadzu representative.
ERR HPV HOME (HPV motor slip error)	Fatal	Cause: High-pressure valve does not operate correctly. Action: Turn the power OFF and contact your Shimadzu representative.

Error Message	Type	Cause and Action
ERR LPV HOME (LPV motor slip error)	Fatal	Cause: Low-pressure valve does not operate correctly. Action: Turn the power OFF and contact your Shimadzu representative.
ERR PUMP HOME (Pump motor slip error)	Fatal	Cause: Measuring pump does not operate correctly. Action: Turn the power OFF and contact your Shimadzu representative.
ERR P.FILE (P.FILE error)	Alarm	Cause: Sample injection was performed incorrectly. Action: Perform re-analysis after correcting the error(s) of the rack type and/or vial No. of the sample to be injected.
NO VIAL DETECTED (Vial not detected error)	Alarm	Cause: No sample vial was placed in the rack position. Action: During an analysis sequence, the missing sample vial will be ignored and analysis will proceed using the next vial specified.
ERR LEAK DETECT (Leak detection error)	Alarm	Concentration of organic mobile phase vapor inside the instrument has exceeded the leak sensor actuation level. Cause: Leak sensor was detected leakage. Action: Inspect the leakage from the flow line. Wipe out spilled liquid after fixing the leakage. Cause: Sensor is too sensitive. Action: Adjust the leak sensor actuation level. "[LEAK THR]" P. 145
NDLE PROTECTED (Foreign substance detection error)	Fatal	Cause: A foreign substance was detected at the tip of the needle. Action: Check for foreign substances inside the autosampler.
NO PUMP ADJUSTED (Pump motor fine adjustment error)	Fatal	Cause: Measuring pump does not operate correctly. Action: Turn the power OFF and contact your Shimadzu representative.
ERR SLIP X (Needle X slip error)	Fatal	Cause: Needle's X-axis (sideways) movement is incorrect. Action: Turn the power OFF and contact your Shimadzu representative.
ERR SLIP Y (Needle Y slip error)	Fatal	Cause: Needle's Y-axis (forward/backward) movement is incorrect. Action: Turn the power OFF and contact your Shimadzu representative.
ERR COOLER (Cooler error)	Fatal	Cause: There is an error in the sample cooler's cooling unit. Action: Turn the power OFF and contact your Shimadzu representative.

Error Message	Type	Cause and Action
ERR HEATER (Heater error)	Fatal	Cause: There is an error in the sample cooler's heating unit. Action: Turn the power OFF and contact your Shimadzu representative.
SYSTEM ERROR XXX (System error) XXX: Sub error code indicating the exact cause	Fatal	Cause: Refer to the following list. Sub error code / Cause: RC0 to 9 — There is an error in the rack changer. Other — There is a hardware failure in electric parts (PCB board etc.) of the autosampler. Action: Turn the power OFF and contact your Shimadzu representative.
DOOR IS OPEN (Door open/close display)	Warning	Cause: The front door is open or panel F has been removed. Action: Close the front door or attach panel F.
ERR INJ VOL or CBM-20A ERROR (injection volume error)	Alarm	Cause: The injection volume exceeds the set value for the maximum volume of injection. Action: Reduce the injection volume to a value less than the set value of the maximum volume of injection. "[MAX INJ. VOLUME]" P. 108, "[VALVE LOOP VOL]" P. 108
ERR TEMP SENSOR (Sample cooler temperature sensor error)	Fatal	Cause: Sample cooler temperature sensor error Action: Turn OFF the power and contact your Shimadzu representative.
ERR TEMP SENS DH (Peltier dehumidifier temperature sensor error)	Fatal	Cause: Peltier dehumidifier temperature sensor error Action: Turn OFF the power and contact your Shimadzu representative.
ERR TEMP SENS RM (Room temperature sensor error)	Fatal	Cause: The room temperature sensor detects an abnormally high or low temperature. Action: If the room temperature is higher than 50 °C, reduce the room temperature to 40 °C or less. If the room temperature is lower than minus 10 °C, raise the room temperature to 4 °C or higher. Cause: Room temperature sensor error Action: Turn OFF the power and contact your Shimadzu representative.

Error Message	Type	Cause and Action
PFILE NOT EXIST (P.FILE missing error)	Fatal	Cause: No description of the specified pretreatment program is found. Action: Check if the specified pretreatment program exists, and if it exists, specify the edit page number on the pretreatment screen for the autosampler method at the workstation LabSolutions, and check the contents of the pretreatment program.
ERR LINK TIMEOUT (Link timeout error)	Alarm	Cause: Remote connection between the instrument and the system controller CBM-20A/20Alite is cut off while analysis is being performed. Action: Check that the optical cable behind the instrument and the CBM-20A/20Alite is connected correctly and is not damaged.

7 Maintenance

7.1 Periodic Inspection and Maintenance

It is necessary to perform periodic inspections of this instrument to ensure its safe use.
It is possible to have these periodic inspections performed by Shimadzu service representatives on a contractual basis.
For information regarding the maintenance inspection contract, contact your Shimadzu representative.

⚠ WARNING

- Unless the instructions here specified, turn off the power always and unplug the instrument prior to performing inspection and maintenance. Otherwise, fire, electric shock or malfunction may occur.

⚠ CAUTION

- When replacing parts, use only the parts listed in "1.5 Component Parts" and "8.2 Maintenance Parts". If any other parts are used, injury or malfunction may occur.
- Never remove the main cover. Otherwise, injury or malfunction may occur.
  Contact your Shimadzu representative to remove the main cover.

7.1.1 Prior to Inspection and Maintenance

- Replace the mobile phase in the flow lines with water.
- Wipe away any dirt from the front panel and the main cover.
- Wipe away any dirt from the keypad with tissue paper or a soft cloth moistened with water.
- Be sure to stop the pump before any inspection or maintenance work.
- When the work is finished, reinstall the panel F before starting the pump.

7.1.2 List of Periodic Inspection and Maintenance

NOTE

The replacement and maintenance periods listed in this table are not guarantee periods, but are presented only as guidelines. These will vary depending on usage conditions.

Maintenance Items Based on Usage Frequency

Inspection/Maintenance Item	Maintenance Timing	Remark	Page
Replacement of needle seal	Replace after approximately 40,000 injections.	*1	P. 229
Replacement of low-pressure valve rotor	Replace after approximately 1,000,000 injections.	*2	P. 242
Replacement of low-pressure valve stator	Replace after approximately 1,000,000 injections.	*2	P. 242
Replacement of high-pressure valve rotor	Replace after approximately 10,000 times.	*1, *2	P. 247
Replacement of high-pressure valve stator	Replace after approximately 20,000 times.	*1, *2	P. 247
Cleaning the high-pressure valve	Carry after approximately 10,000 times.		P. 247
Replacement of sample loop	Replace after approximately 40,000 injections.		P. 256
Needle replacement	Replace after approximately 40,000 injections.		P. 262
Replacement of rinsing port cap	Replace after approximately 10,000 injections.	*3	P. 266
Replacement of vial detection spring	Replace after approximately 40,000 injections.		Contact your Shimadzu representative
Rinsing pump (optional)	Replace after approximately 700,000 seconds.		Contact your Shimadzu representative

Maintenance Items Based on Years Used

Inspection/Maintenance Item	Maintenance Interval				Remark	Page
	1 year	2 years	3 years	6 years		
Replacement of measuring pump plunger seal	×				*4	P. 233
Inspection/replacement of measuring pump plunger	×				*4	P. 238
Replacement of suction filter	×					P. 260
Replacement of preheat block or SUS tubing HP OUT	×				Replace when the clogging does not remove after reverse cleaning.	P. 271
Replacement of panel F			×		Replace if there is excessive condensation.	P. 265
Cleaning and oiling of the needle drive section (Z mount)	×					Contact your Shimadzu representative
Cleaning and oiling of other drive sections			×			Contact your Shimadzu representative
Fuse replacement			×			Contact your Shimadzu representative

*1 This is given for reference on the assumption that a mixed solution of water and organic solvent as described in the system validation section in the Nexera X2 SYSTEM GUIDE is used.
Depending on the type of solvent or pumping pressure, the service life of each part may be longer or shorter.
If a buffer solution that crystallizes or precipitates when dried is used as mobile phase and the instrument is kept unused for a long time, the service life of each part may be shortened significantly.
If purified water exceeding 80 % is used as mobile phase and is continuously pumped at a high pressure, the service life of each part may be shortened.

*2 Rinse the flow line sufficiently with distilled water or deionized water.

*3 If you have a severe cross contamination, replace the rinsing port cap with a new one.

*4 These intervals are given for reference on the assumption that the pretreatment function or internal rinsing of the needle is used.
If these functions are not used, the interval should be about three years as a guide.

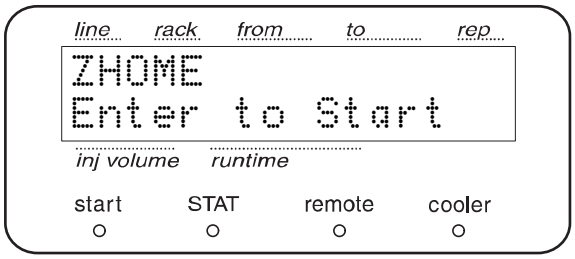
7.2 Replacing Needle Seal

Necessary parts

Part Name	Part Type	Part No.	Remark
Needle seal 30A (standard)	Consumable part	S228-52253	Applicable pH: 1 to 9 Maximum withstand pressure: 130 MPa
PEEK needle seal 30A (option)	Consumable part	S228-53178-91	Applicable pH: 1 to 14 Maximum withstand pressure: 66 MPa
Housing needle seal 30A	Maintenance part	S228-52228	
Cap needle seal 30A	Maintenance part	S228-51904	
Needle seal 30A kit	Maintenance part / consumable part	S228-52401-92	3 needle seals 30A (standard) and 1 housing, needle seal 30A in a set
Tool for needle seal XR	Service tool	S228-50570	

7.2.1 Removing the Needle Seal

1 From the initial screen, press (↑) to display the [Z HOME] screen.



2 Press **enter**.
The needle rises to the highest position and then moves to the center of the autosampler.

3 Turn OFF the instrument.

4 Open the door, and remove the sample racks.

5 Loosen the screws (5 points) and then slide the Panel F a little to the right before pulling it forward to remove it.

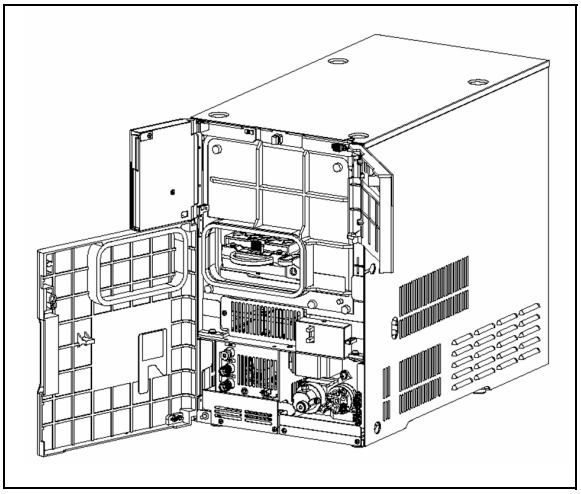


Fig. 7.1

6 Loosen the cap using the supplied needle seal XR installer / remover tool, and remove it.

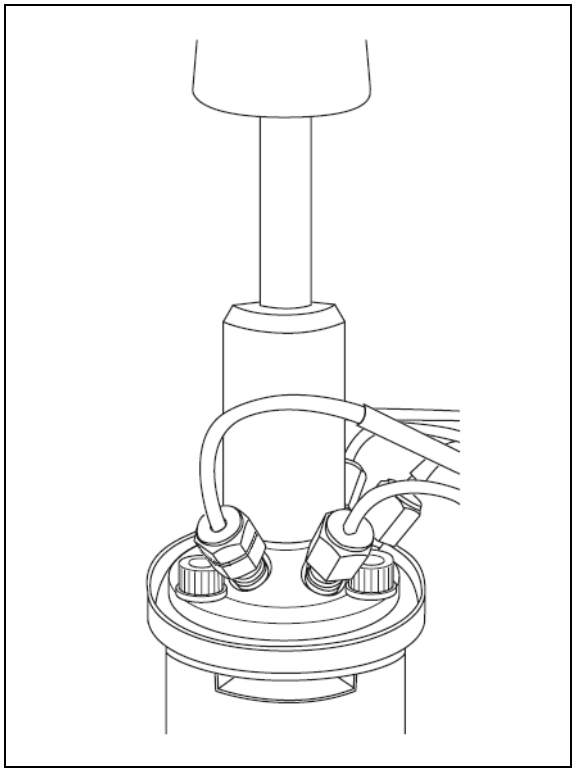


Fig. 7.2

7 Using tweezers, remove the housing and the needle seal from the stator.

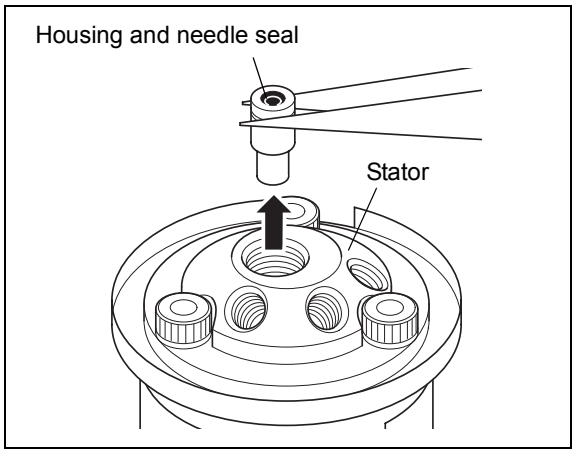


Fig. 7.3

8 Remove the needle seal from the housing. If it is hard to remove, press the needle seal from the top with the tweezers.

⚠ CAUTION

If the needle seal cannot be removed from the housing, do not attempt to pull it out by force.
Replace both the needle seal and the housing at the same time.
Attempting to pull it out by force may result in injury.

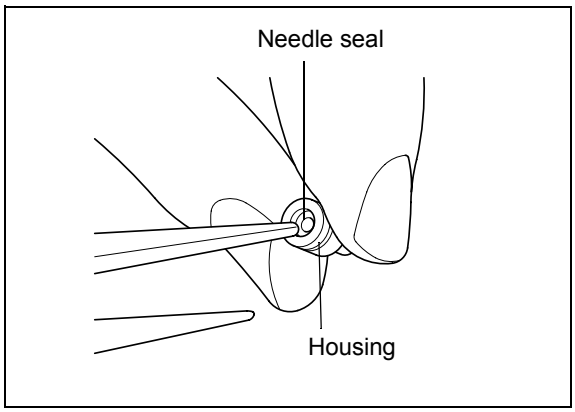


Fig. 7.4

7.2.2 Installing the Needle Seal

1 Insert the housing into the stator.

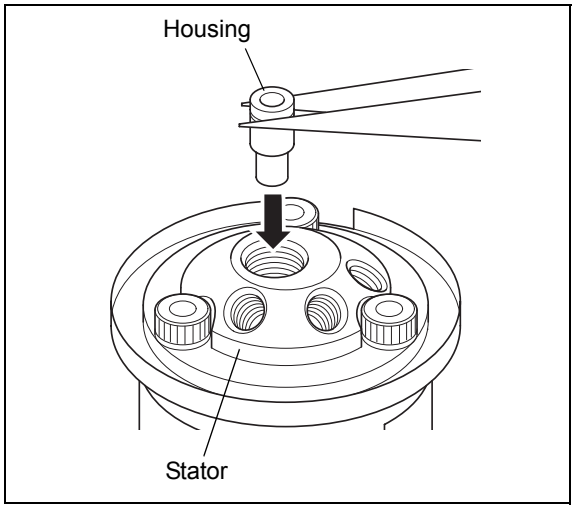


Fig. 7.5

2 Insert a new needle seal into the housing. Keep the tapered hole on the needle seal facing upward.

⚠ CAUTION

When inserting the needle seal into the housing, hold the needle seal using a piece of soft cloth or paper, being careful not to allow dust to enter the hole.
The entry of dust may create small gaps or voids that could lead to carryover or leaks.

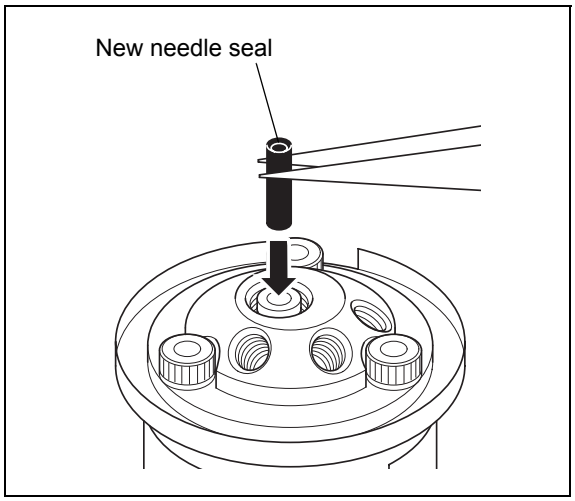


Fig. 7.6

3 Finger-tighten the cap on the stator, then tighten it further 180° using the needle seal XR installer / remover tool.

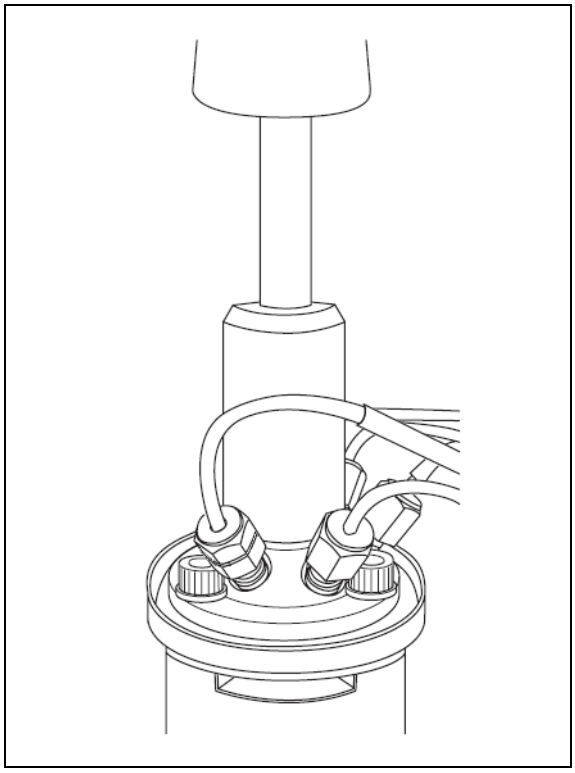


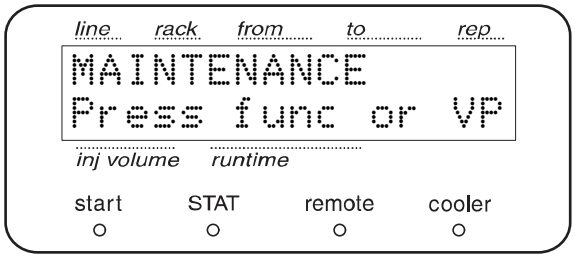
Fig. 7.7

4 Reattach the panel F so that the autosampler is hermetically sealed and close the door.

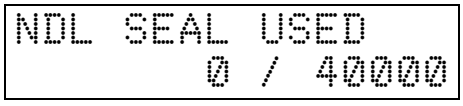
> **⚠ CAUTION**
>
> If the panel F has not been reattached properly, condensation may occur in the sample cooler.

5 Turn ON the instrument.

6 From the initial screen, press **VP** twice.
The maintenance information group screen is displayed.



7 Press **func** until [NDL SEAL USED] appears.
The needle seal's usage frequency and replacement alert value are displayed.



8 Press **0** and **enter**.
Reset the counter to [0].

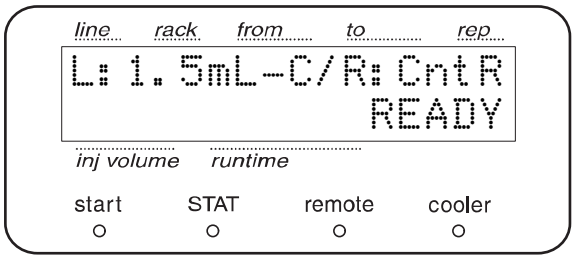
7.3 Replacing Plunger Seal

Necessary parts

Part	Type	Part No.
Plunger seal	Consumable part	S228-35145

7.3.1 Before Removing Pump Head

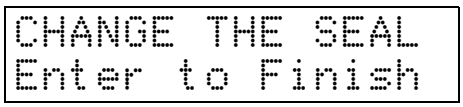
1 Press **CE** until the initial screen appears.



2 Press **VP** repeatedly until the Maintenance Information screen on the right is displayed.

3 Press **func** repeatedly until the [P-SET] screen on the right is displayed.

4 Press **enter**.
The plunger will be fully retracted.



7.3.2 Removing the Pump Head

1 Open the door.

2 Using the wrench (provided), loosen and remove the tubing from the top and bottom of the pump head as well as the male nuts from port No. 2 and the center port of the low-pressure valve. Then loosen the manual prime valve with your fingers and remove it.

3 Using the Allen wrench provided, loosen the two hex socket head bolt in the pump head, and remove the pump head.

4 Ease the pump head out gently, keeping it aligned with the plunger at all times.

⚠ CAUTION

If the pump head is pulled out diagonally, the plunger could be broken.

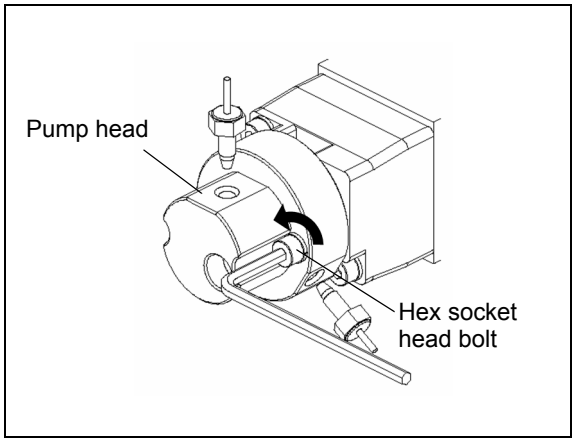


Fig. 7.8

NOTE

Remove the clamp that fastens the suction tube at the front of the pump head in advance. When pulling the pump head out, move the suction tube toward the bottom of the leak tray to facilitate pulling it out.

7.3.3 Replacing the Measuring Pump Seal

1 Remove the seal holder gently in the horizontal direction aligning it with the plunger to prevent damage to the plunger.

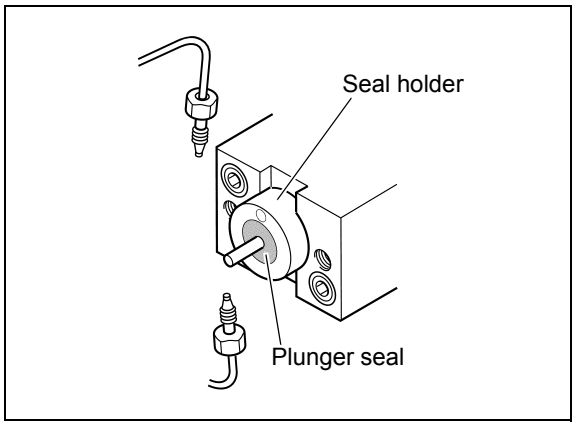


Fig. 7.9

2 Insert the flanged end of the seal installer/remover tool into the seal vertically.

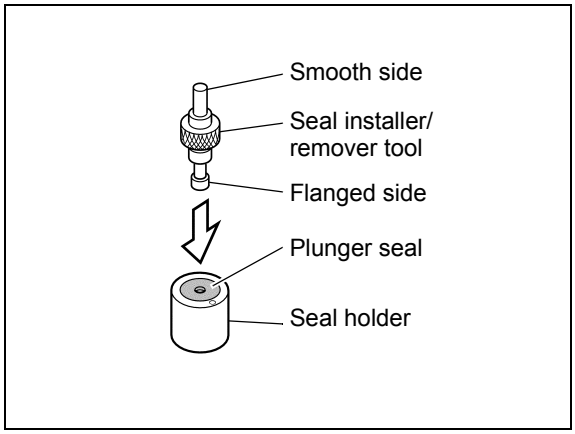


Fig. 7.10

3 Pull out the tool to remove the used seal vertically. The measuring pump seal is removed from the seal holder.

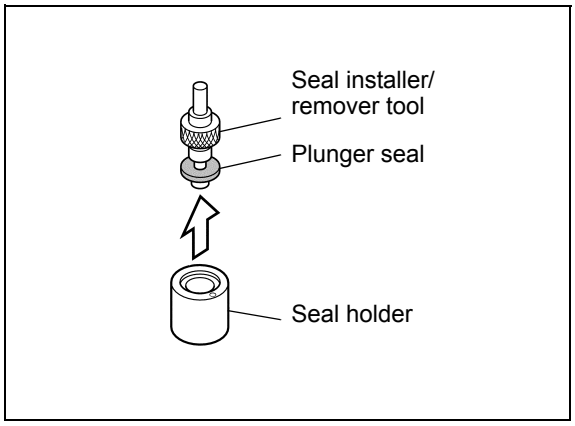


Fig. 7.11

4 Remove any contamination present on the pump head and seal holder.
Cleaning the measuring pump head wipe the inside of the pump head and seal holder using soft cloth or paper soaked with 2-propanol.

NOTE

If any material remains on these surfaces, the plunger cannot maintain a seal.

5 Place the new seal on the straight end of the seal installer/remover tool.
(Refer to Fig. 7.12 about seal direction.)

6 Lower the seal into the seal holder, then remove the seal installer/remover tool.

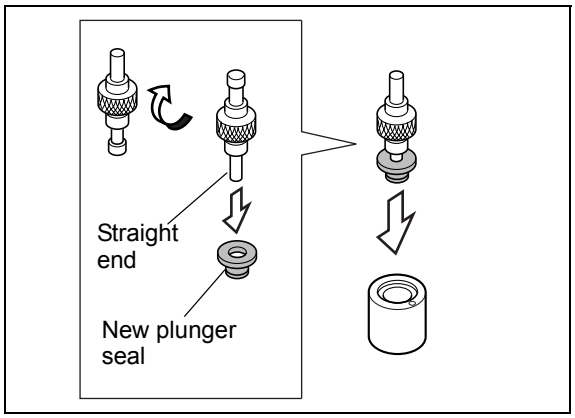


Fig. 7.12

7.3.4 Installing the Pump Head

1 Push the seal holder in slowly aligning it with the plunger, and then push the pump head in slowly and gently horizontally.

⚠CAUTION

If the pump head is pushed in forcefully, the plunger could be broken.

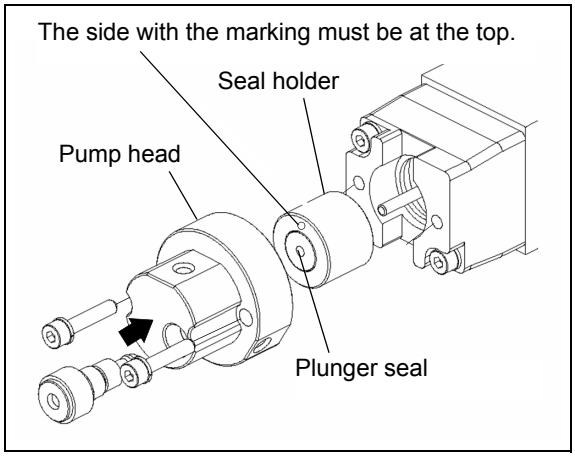


Fig. 7.13

NOTE

Press the seal surface using soft cloth or paper soaked with 2-propanol when installing the seal holder to prevent the dropping out the seal.

2 Place the two hex socket head bolts into the holes in the pump head, and screw them in alternately and uniformly with provided Allen wrench.

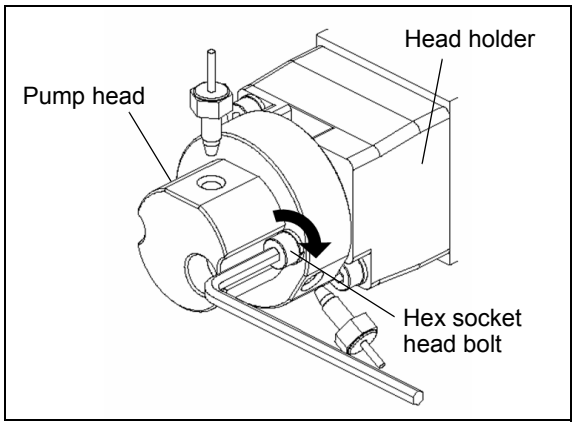


Fig. 7.14

NOTE

Screw the bolts in alternately 90°. At the end, tighten the bolts securely with the Allen wrench.

3 Attach the tubing to the top and bottom of the pump head as well as port No. 2 and the center port of the low-pressure valve. Then tighten the manual prime valve with your fingers and secure it in position.

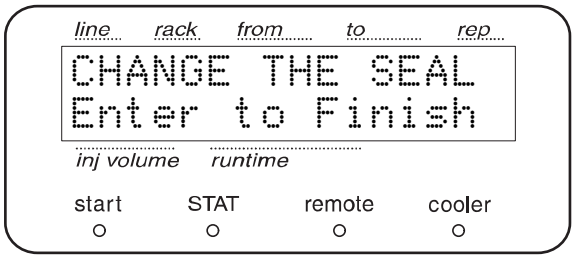
NOTE

If the clamp that fastens the suction tube has been removed from the front of the pump head, re-attach it.

4 Close the doors.

5 Press **enter**.
Return the plunger to the original position.

6 Press **purge**.
Start purging of the flow lines.



7.3.5 Check after Replacement

After replacing a plunger seal, check the following:

- There is no leakage from the gap between the pump head and the head holder.
- There is no leakage from the rinse flow line.

NOTE

When not even after replacement of the plunger seals, the surfaces of the plunger may be scratched or nicked, in which case the plunger must be replaced.

"7.4 Replacing and Inspecting Measuring Pump Plunger" P. 238

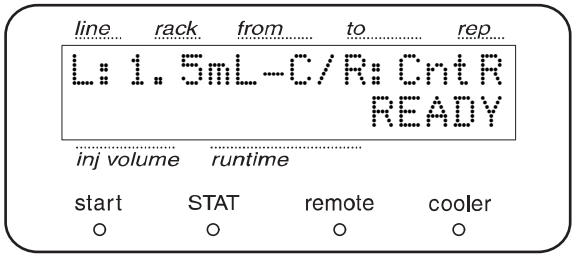
7.4 Replacing and Inspecting Measuring Pump Plunger

Necessary parts

Part	Type	Part No.
Measuring pump plunger	Replacement part	S228-35010-92

7.4.1 Before Removing the Measuring Pump Plunger

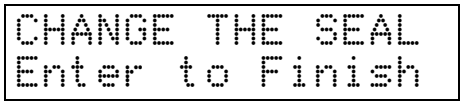
1 Press **CE** repeatedly to display the initial screen.



2 Press **VP** repeatedly to display the Maintenance Information screen on the right.

3 Press **func** repeatedly to display the [P-SET] screen on the right.

4 Press **enter**.
The plunger will be fully retracted.



7.4.2 Replacing the Measuring Pump Plunger

1 Open the door.

2 Using the wrench (provided), loosen and remove the tubing from the top and bottom of the pump head as well as the male nuts from port No. 2 and the center port of the low-pressure valve. Then loosen the manual prime valve with your fingers and remove it.

3 Using the Allen wrench provided, loosen the two hex socket head bolts in the pump head.

4 Pull the pump head out gently aligning it with the seal holder, and then pull the seal holder out gently and horizontally aligning it with the plunger.

⚠CAUTION

If the pump head is pulled out diagonally, the plunger could be broken.

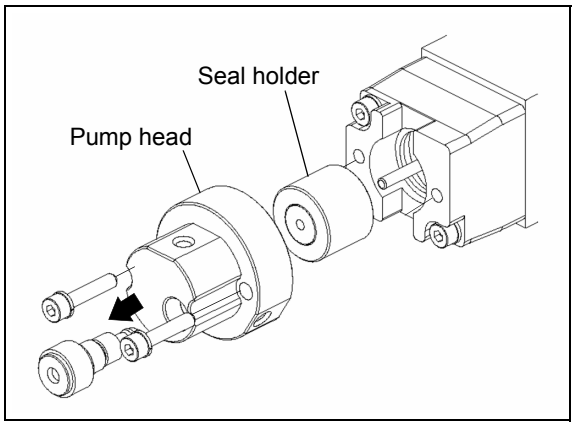


Fig. 7.15

NOTE

Remove the clamp that fastens the suction tube at the front of the pump head in advance. When pulling the pump head out, move the suction tube toward the bottom of the leak tray to facilitate pulling it out.

5 Using the plunger tool (Part No. S228-34672-02), rotate the plunger holder counterclockwise and remove the plunger ASSY.

⚠CAUTION

When sliding the plunger tool onto the plunger, be careful not to damage the plunger.

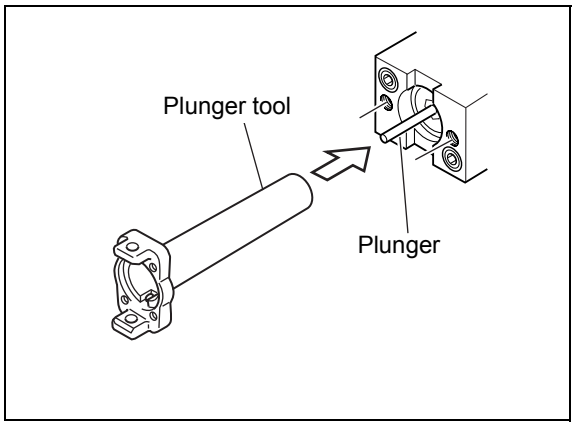


Fig. 7.16

6 Inspect the plunger for visible nicks or scratches.
- Replace the plunger ASSY if it is damaged.
- If it is not damaged but there is contamination present, remove the contamination. Wipe the inside of the pump head and seal holder using soft cloth or paper soaked with 2-propanol.

NOTE

The measurement accuracy may be reduced if there is any seal residue or other matter on the plunger.

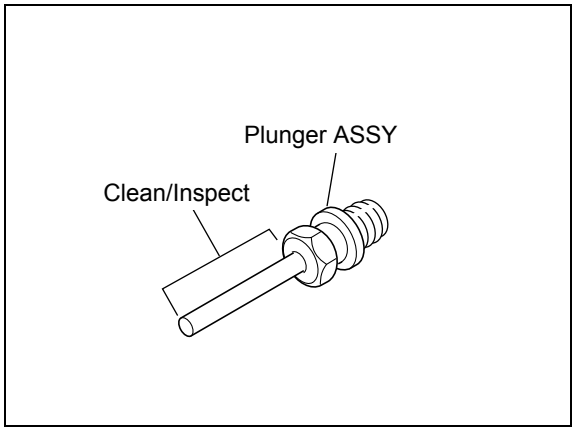


Fig. 7.17

7.4.3 Installing the Measuring Pump Plunger

1 Hold the plunger ASSY between the thumb and fingers, and hand-tighten it into the pump body.

2 Use the plunger tool to thoroughly tighten the plunger ASSY.

3 Push the seal holder in gently and horizontally aligning it with the plunger, and then push the pump head in gently and horizontally aligning it with the seal holder.

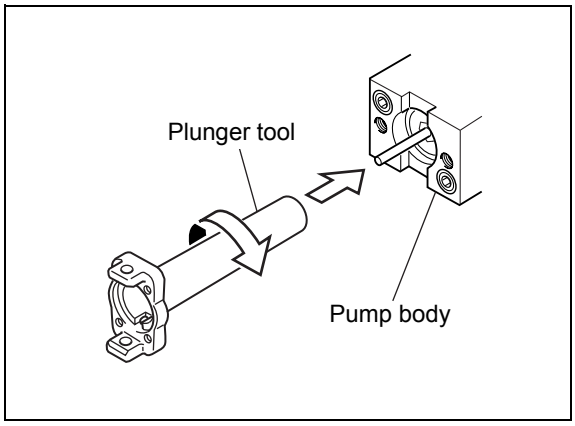


Fig. 7.18

⚠CAUTION

If the pump head is pushed in forcefully, the plunger could be broken.

NOTE

Press the seal surface using soft cloth or paper soaked with 2-propanol when installing the seal holder to prevent the dropping of the seal.

4 Place the two hex socket head bolts into the holes in the pump head, and screw them in alternately and uniformly with provided Allen wrench.

NOTE

Screw the bolts in alternately 90°. At the end, tighten the bolts securely with the Allen wrench.

5 Attach the tubing to the top and bottom of the pump head as well as port No. 2 and the center port of the low-pressure valve. Then tighten the manual prime valve with your fingers and secure it in position.

NOTE

If the clamp that fastens the suction tube has been removed from the front of the pump head, re-attach it.

6 Close the doors.

7 Return the plunger to the original position by pressing (enter) .

8 Press (purge) to start purging of the flow lines.

7.4.4 Check after Replacement

After replacing the measuring pump plunger seal, check the following:

- There is no leakage from the gap between the pump head and the head holder.
- There is no leakage from the piping connection.

7.5 Replacing and Inspecting Low-Pressure Valve Rotor and Stator

⚠ CAUTION

If liquid is leaked from the low-pressure valve, first check that the rotor is not damaged in the way described in steps 1 to 7 of "7.5.1 Removing the Low-Pressure Valve Rotor and Stator". If there is no damage, carefully wipe the rotating and sliding surfaces of the rotor and stator with using soft cloth or paper soaked with 2-propanol. Dirt or dust on the sliding surfaces may adversely affect the sealing and cause liquid to be leaked. If liquid leakage persists, replace the rotor and stator.

Necessary parts

Part	Type	Part No.
Low-pressure valve rotor 30A	Consumable part	S228-51922
Low-pressure valve stator 30A	Consumable part	S228-51663-01

7.5.1 Removing the Low-Pressure Valve Rotor and Stator

1. Turn the power switch OFF and open the door.
2. Lower the rinse solution container to a position lower than the low-pressure valve.
3. Remove the tubing cover from the right side of the instrument.
4. Using a wrench, loosen and remove all of the flow line male nuts from the housing.

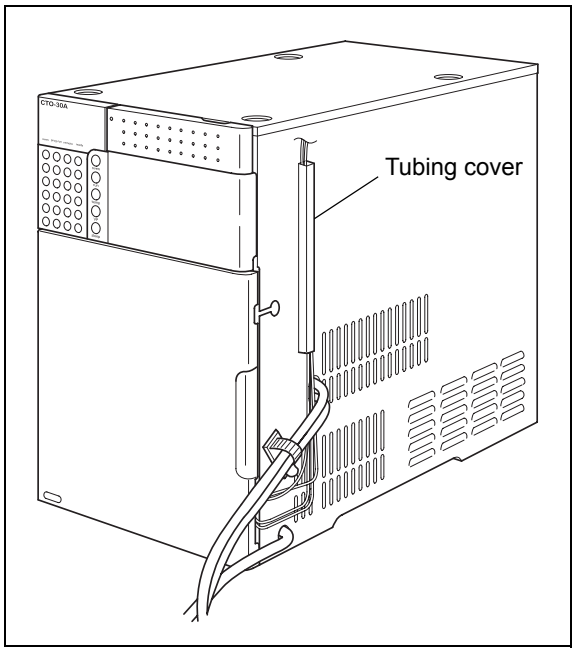


Fig. 7.19

5 Loosen and remove the three hex socket head bolts with the M4 Allen wrench.

NOTE

When removing the tubing, loosen the ports in order of No. 3 → No. 2 → No. 1 → center → No. 4 → No. 6 → No. 5 to facilitate your work.

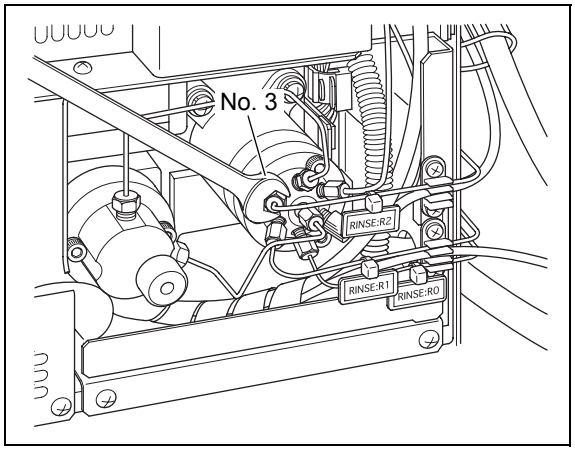


Fig. 7.20

6 The stator part consists of ring and stator ASSY. Be careful not to drop any of the pieces when lifting them from the instrument.

NOTE

Remove the ring and housing slowly to prevent the stator and packing from dropping out of the stator ASSY.

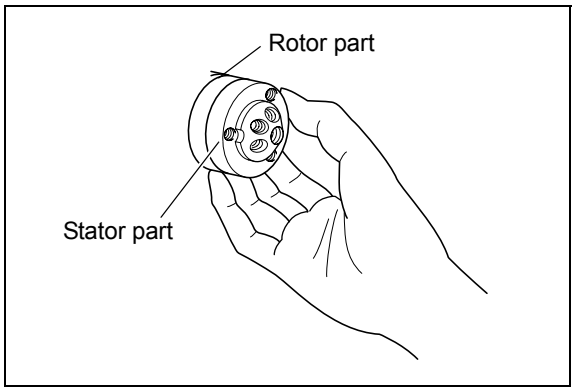


Fig. 7.21

7 Grip the rotor securely, and pull it out of the instrument.

NOTE

When handling the rotor, try to only touch the outer circumference.

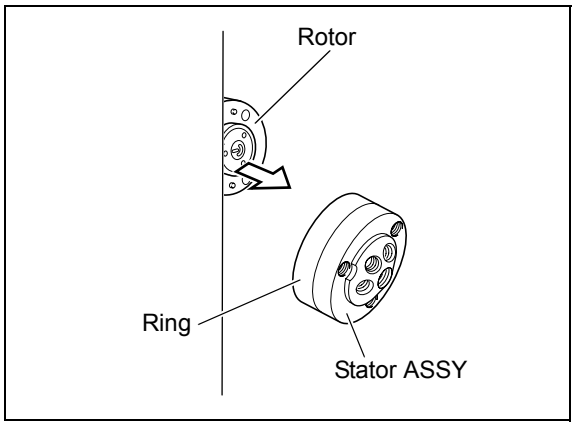


Fig. 7.22

8 Inspect the sliding surfaces of the rotor and the stator for visible nicks or scratches.
If the rotor or stator is damaged, replace the parts.
If the rotor or stator is not damaged, wipe the surface of rotor and stator using soft cloth or paper soaked with 2-propanol.

9 Place the rotor, stator, packing and housing in 2-propanol, and sonicate them for five minutes.

7.5.2 Installing the Low-Pressure Valve and Stator

1 Align the notch in the rotor with the mark on the shaft, and push the rotor all the way back onto the shaft.

NOTE

When handling the rotor, try to only touch the outer circumference. When returning the rotor to the instrument, wipe all sliding contact surfaces with 2-propanol.

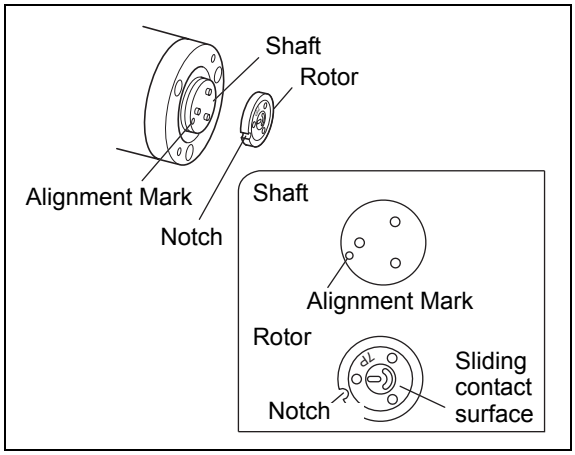


Fig. 7.23

2 Reassemble the housing in the order indicated in the figure to the right.

NOTE

Assembling the stator, packing and housing with the protrusion of the stator facing the ring, assemble the stator and packing to that the holes are aligned.

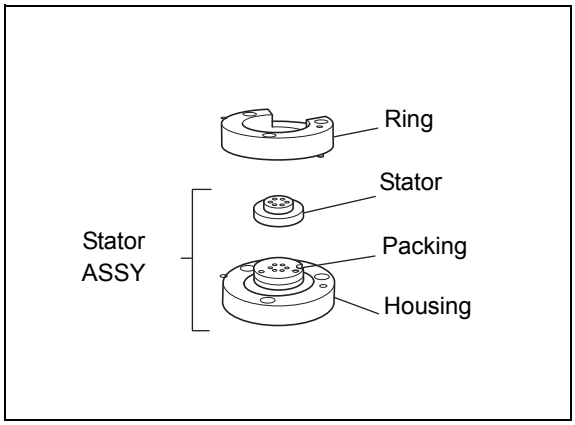


Fig. 7.24

3 With the stator facing out of the instrument, insert the ring and stator ASSY into the body. Be careful not to drop the stator.

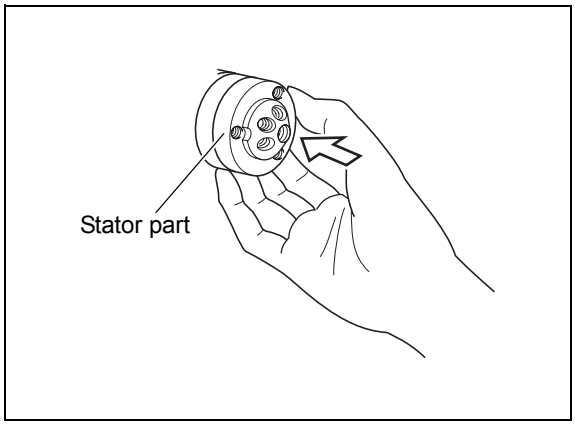


Fig. 7.25

4 Screw the three hex socket head bolts into the housing, tightening them gradually with the Allen wrench provided.

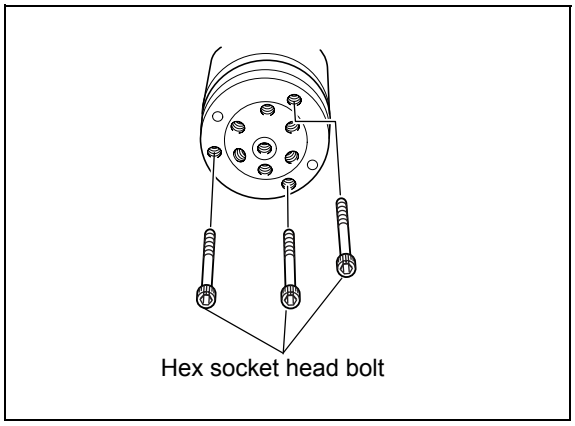


Fig. 7.26

NOTE

Alternately tighten three hex socket head bolts 90° at a time. Ensure that the hex socket head bolts are secured firmly. Use the long end of the Allen wrench as a handle to tighten them securely. Do not tighten any one of these socket head bolts, the stator will be inclined.

5 Reconnect the flow line tubing that was disconnected in step 4 of "7.5.1 Removing the Low-Pressure Valve Rotor and Stator".

NOTE

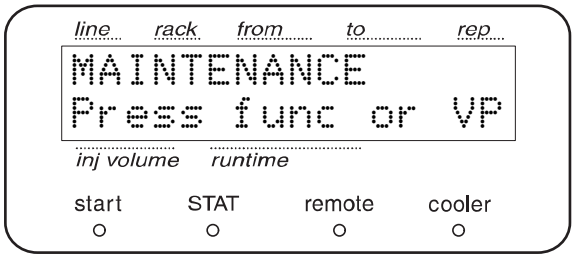
When installing the tubing, tighten the ports in order of No. 5 → No. 6 → No. 4 → center → No. 1 → No. 2 → No. 3 to facilitate your work.

6 Close the door and attach the tubing cover to the right side of the instrument.

7.5.3 Resetting the Usage Frequency

1 Reset the low-pressure valve rotor counter.

2 Turn the power ON and press **VP** repeatedly until the Maintenance Information screen on the right is displayed in the initial screen.



3 Press **func** repeatedly until the [LPV SEAL USED] screen on the right is displayed.
The usage frequency and replacement alert value of the low-pressure valve's rotor seal are displayed on the screen.

4 Press **0** and **enter**.
Reset the counter to [0].

5 Press (CE) until screen return to the initial screen.

6 Press (purge) to start purging of the flow lines.

7.6 Replacing and Inspecting High-Pressure Valve Rotor and Stator

⚠CAUTION

If liquid has leaked from the high-pressure valve, first check that the rotor and stator are not damaged as described in steps 1 to 5 of "7.6.2 Removing the High-Pressure Valve Rotor and Stator".
If you disassemble the stator, the rotor cannot be reused and a new rotor must be used. Inspect in detail the surface of the stator by using a loupe. If there are no scratches on the stator surface, the stator can be reused.
Dirt or dust on the sliding surfaces may adversely affect the sealing and cause liquid to leak. If liquid leakage persists, replace the rotor and stator at once.

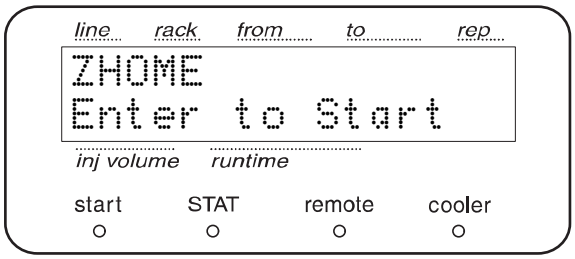
Necessary Parts

Part	Type	Part No.
High-pressure valve rotor 30A	Consumable part	S228-52139
High-pressure valve stator 30A	Consumable part	S228-48858-95
Jig rotor	Service tool	S228-48899-91

7.6.1 Before Removing the High-Pressure Valve Rotor and Stator

1 From the initial screen, press (↑) to display the [Z HOME] screen.

2 Press **enter**.
The needle rises to the highest position and then moves to the center of the autosampler.



3 Turn the power switch OFF, and unplug the instrument.

4 Open the front door and the panels on the right and left, and remove panel F.

7.6.2 Removing the High-Pressure Valve Rotor and Stator

1 Loosen all the male nuts at tubing connections and remove the tubing from the stator. Remove the needle seal.
When replacing only the rotor seal of the high-pressure valve, it is not necessary to remove the needle seal.
For details about how to remove the needle seal, see
"7.2.1 Removing the Needle Seal" P. 229
For details on the procedure for removing or tightening UHPLC fittings at the outlet tubing of the instrument, refer to the Nexera X2 SYSTEM GUIDE.
Nexera X2 SYSTEM GUIDE, "2.3.4 Prior to Plumbing the High Pressure Flow Line"

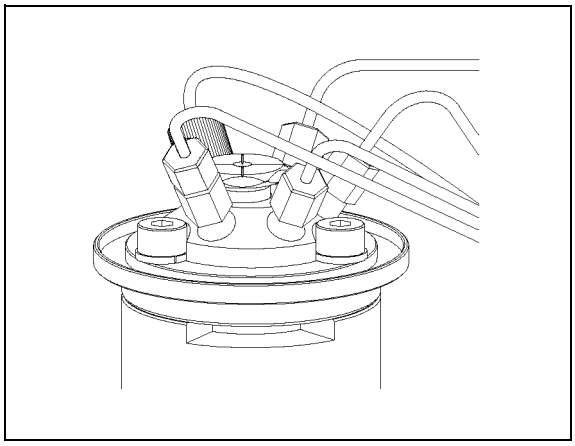


Fig. 7.27

2 Using the M4 Allen wrench, loosen the three hex socket head bolts 90° each time in order of A, B, and C.

NOTE

If you attempt to loosen only one of these socket head bolts, the stator will be inclined, followed by seizing of the bolt or inability to remove the stator.

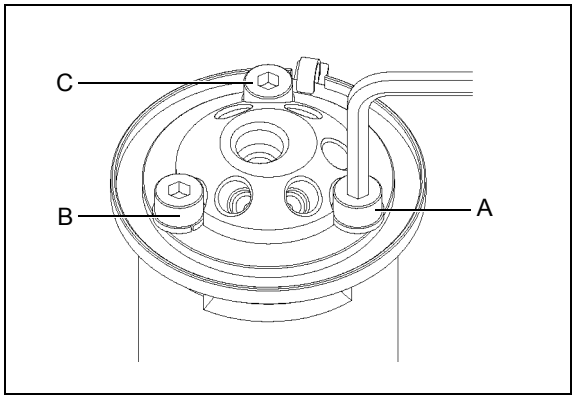


Fig. 7.28

3 Grip the rotor securely, and pull it out of the instrument.

NOTE

When handling the rotor, try to touch only the outer edge.

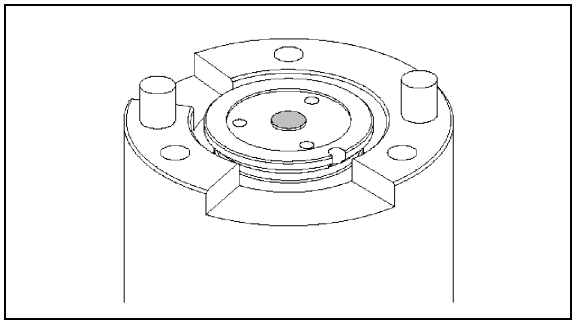


Fig. 7.29

4 Inspect in detail on the surface of the stator by using a loupe.If there are no scratches on the stator surface, the stator can be reused.

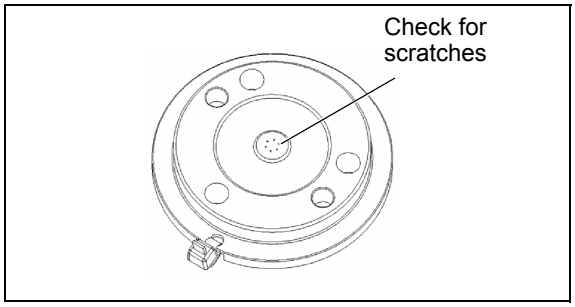


Fig. 7.30

NOTE

- Check the scratch around the six ports.
- If you disassemble the stator, the rotor cannot be reused and a new rotor must be used.

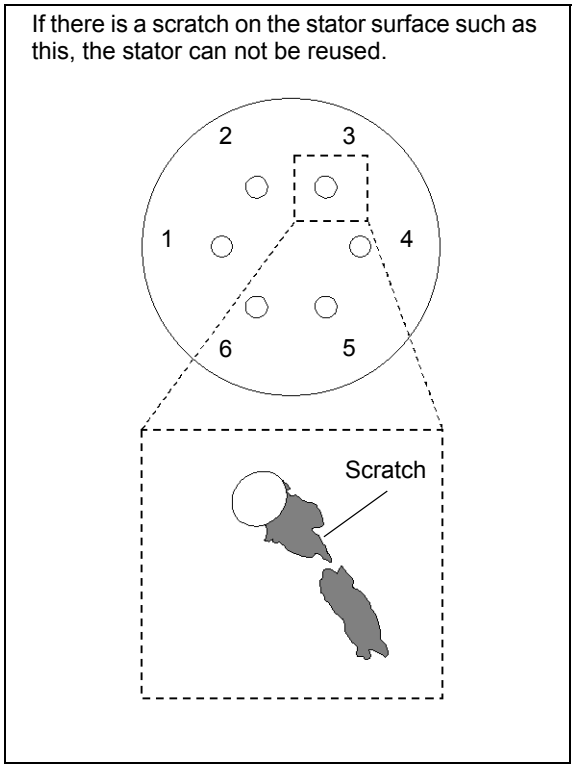


Fig. 7.31

5 If the stator can be reused, wipe the dirt on the surface of the stator using a paper soaked with 2-propanol.
If the dirt is hard to remove, immerse the stator in 2-propanol, and sonicate for 5 minutes.

7.6.3 Installing the High-Pressure Valve Rotor and the Stator ASSY

1 Align the notch in the rotor with the mark on the shaft, and push the rotor all the way back onto the shaft.

NOTE

- When handling the rotor, try to touch only the outer edge. After installing, wipe the sliding surface with the stator using soft cloth or paper soaked with 2-propanol.
- Keep the "XR IIA" marked face of the rotor toward the stator.

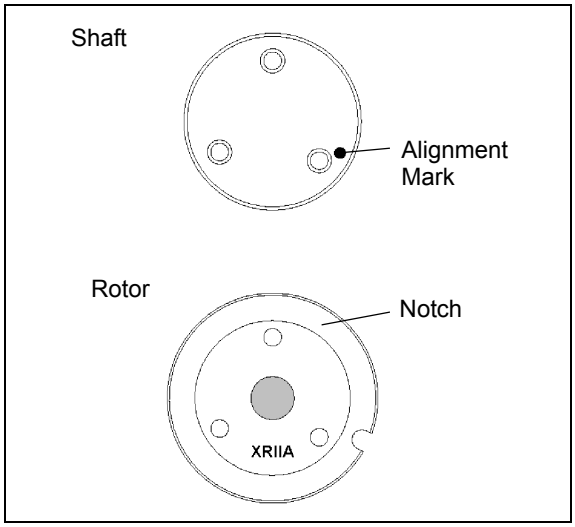


Fig. 7.32

2 Apply two or three drops (approx. 5 uL) of 2-propanol from a clean pipette or syringe onto the sliding surface in the center of the rotor.

NOTE

Do not apply 2-propanol in large amounts. This could cause it to permeate into the mechanism of the high-pressure valve body and hinder correct operations of the valve.

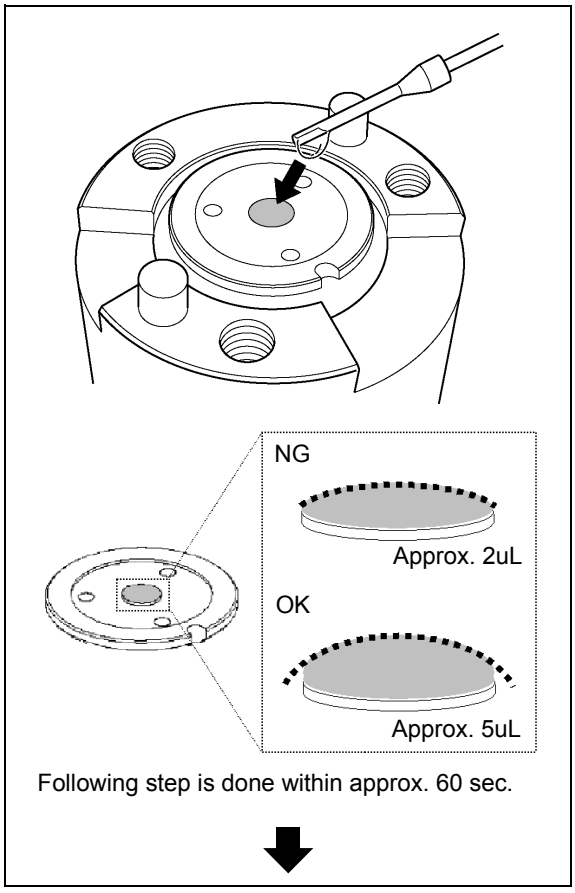


Fig. 7.33

3 Place the stator on the high-pressure valve body within approx. 60 sec from drop of 2-propanol, and tighten three hex head bolts gently with the longer side of an Allen wrench until the bolt heads touch the stator.

NOTE

Install the stator before the 2-propanol drops on the sliding surface in the center of the rotor have dried up.

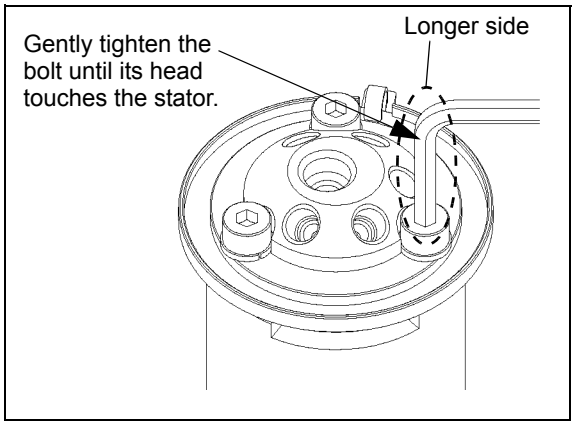


Fig. 7.34

4 Fully insert the high-pressure rotor replacement jig between the valve body and the stator. The jig is stored inside the front door.

⚠ CAUTION

Be sure to use the provided jig. Otherwise the stator may not uniformly contact the rotor surface, which may result in leaks or rapid wear of the rotor.

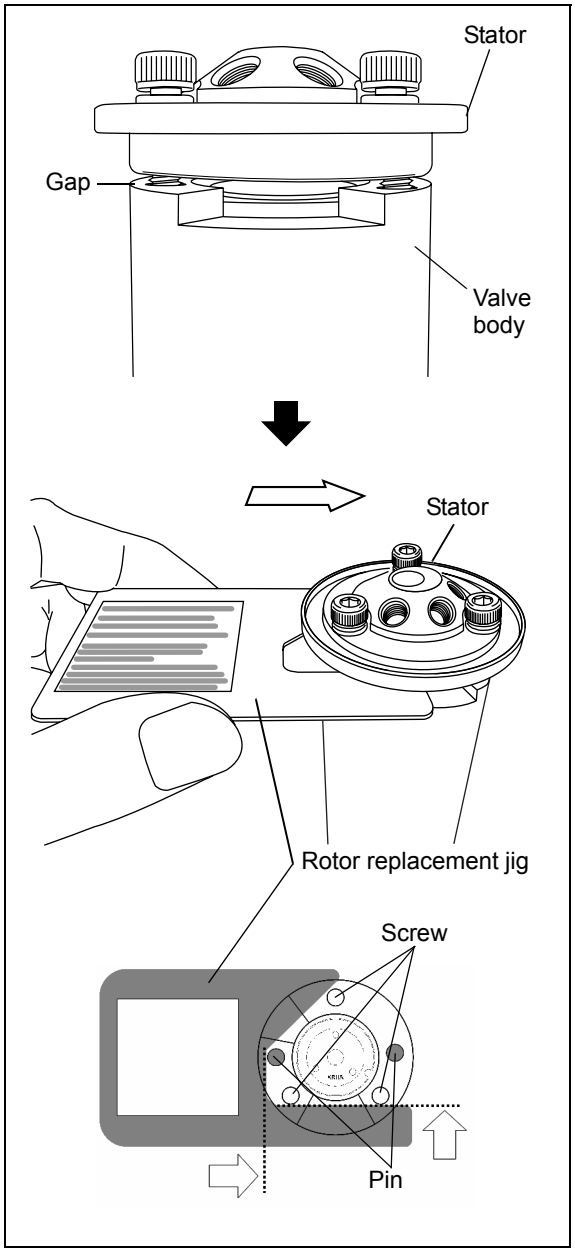


Fig. 7.35

5 Screw the three hex socket head bolts 45° at a time using the long leg of an Allen wrench as a handle and tighten them securely.

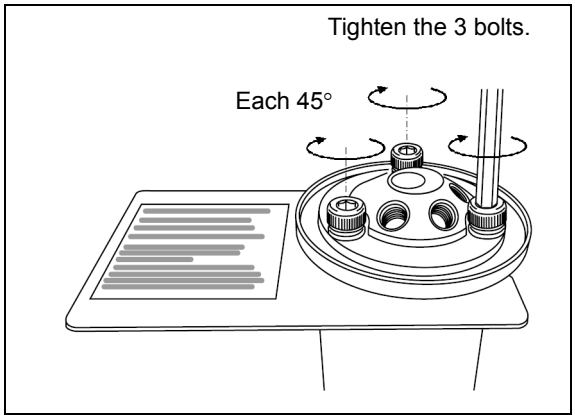


Fig. 7.36

6 Loosen the three hex socket head bolts to the angle of about 45 degrees and remove the jig.

NOTE

Loosen the three hex socket head bolts an additional 45 degrees at a time until the jig can be removed smoothy.

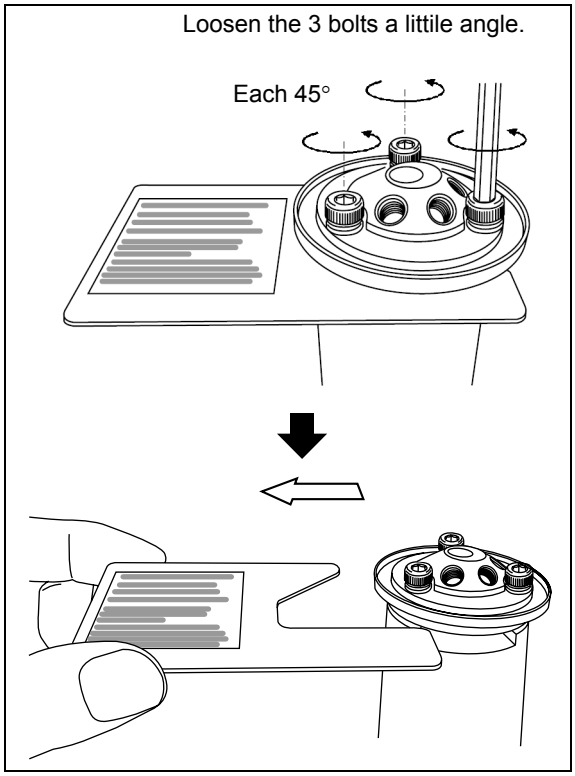


Fig. 7.37

7 Tighten the three hex socket head bolts 45° at a time in the sequence of A, B, C using an Allen wrench.

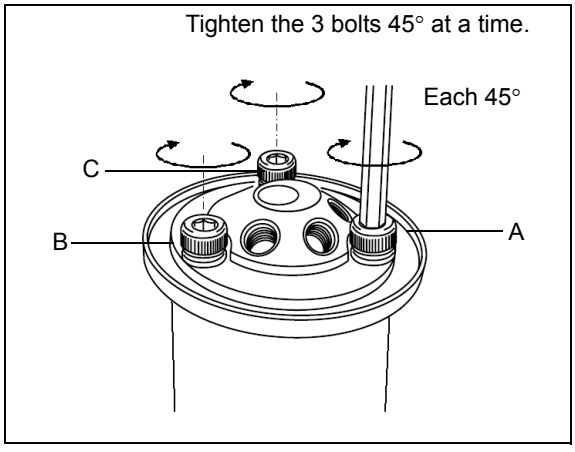


Fig. 7.38

8 After the hex socket head bolts are completely tightened, loosen the three hex socket head bolts to 45 degrees four times (180 degrees in total) in order of C, B, and A.Then tighten them completely by turning them 45 degrees each time in order of A, B, and C.

NOTE

Use the long side of the Allen wrench as a handle to tighten them securely.

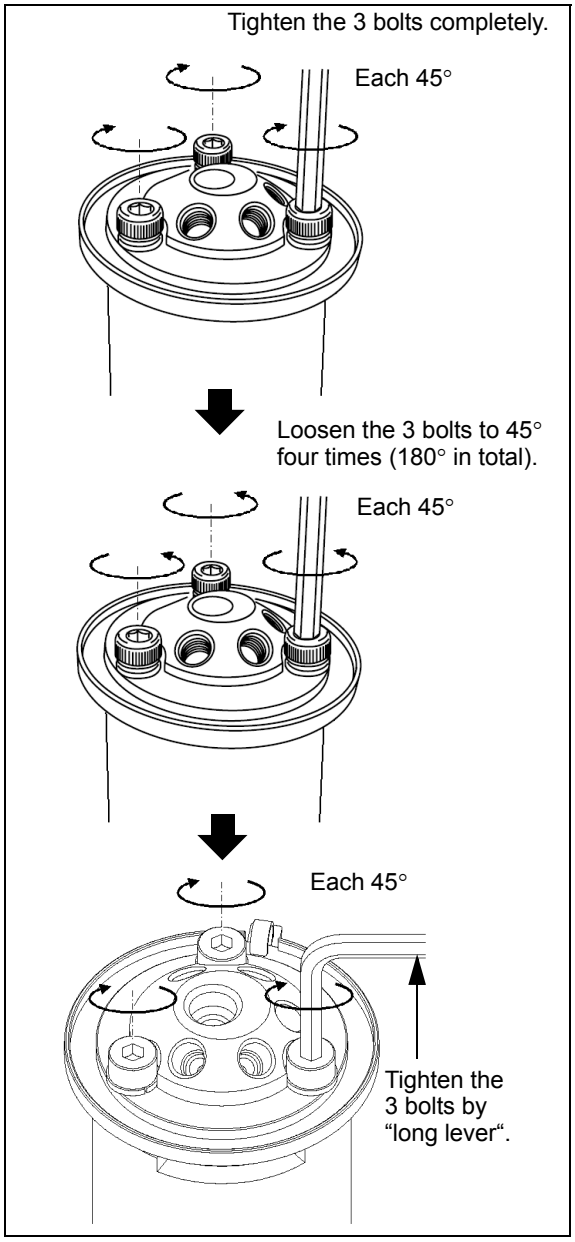


Fig. 7.39

9 Reinstall the tubing at each port of the high-pressure valve and reattach panel F in the original position. For details on the procedure for removing or tightening UHPLC fittings at the outlet tubing of the instrument, refer to the Nexera X2 SYSTEM GUIDE.

☞ Nexera X2 SYSTEM GUIDE, "2.3.4 Prior to Plumbing the High Pressure Flow Line"

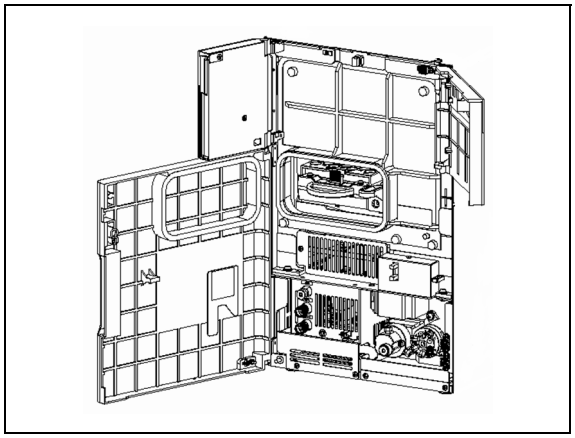


Fig. 7.40

⚠CAUTION

If the stator is not leveled, stator and rotor surface may be damaged and it may cause the leakage.

NOTE

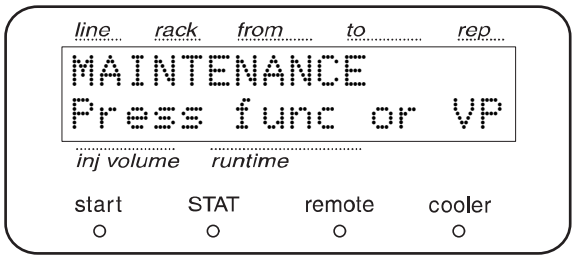
Go through the following steps to reset the usage frequency and the HPV rotation immediately.

10 To store the high-pressure valve rotor replacement jig, insert it inside the front door.

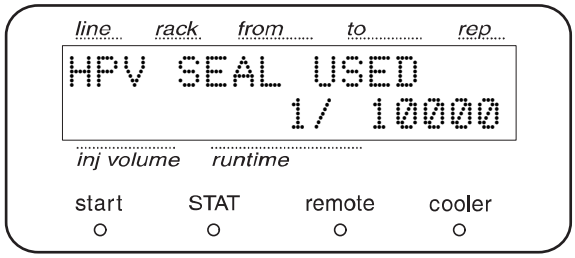
7.6.4 Resetting the Usage Frequency

1 Check that there is a gap of approximately 3 mm between the bottom side of the Z mount and the tubing connected to the high-pressure valve.

2 Turn the power ON and press **VP** until the Maintenance Information screen appears.



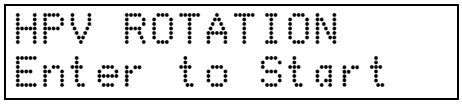
3 Press **func** repeatedly until the [HPV SEAL USED] screen on the right is displayed.
The usage frequency and replacement alert value of the high-pressure valve's rotor seal are displayed on the screen.



4 Press **0** and **enter**.
Reset the counter to [0].
When only the rotor seal has been replaced, move to step 7.

(When the stator has been replaced)

5 When the stator has been replaced, press **func** once to display the [HPV STATOR USED] screen.
The usage frequency and replacement alert value for the high-pressure valve stator are displayed on the screen.



6 Press (0) and (enter).
Reset the counter to [0].

7 Press (func) repeatedly until the [HPV ROTATION] screen is displayed.
Pump organic solvent such as 2-propanol, methanol, acetonitrile 100% at a rate of approximately 2 mL/min so that a pumping pressure of approximately 80 MPa will be obtained.

8 Press (enter).
The high-pressure valve switches over automatically once every few seconds. The number of rotations remaining is displayed on the second line.

NOTE

- When using a new rotor and stator, abrasion powder may be generated initially. For this reason, perform steps 7 and 8.
- Once operation has started it cannot be stopped until all 50 rotations have been completed. Abrasion powder may be generated initially and so be sure to remove the column before the operation.

9 Press (CE) to return to the initial screen.

10 Close the door, and press (purge) to start purging of the flow lines.

7.6.5 Installation and daily maintenance

■ Before using SIL-30AC

Pump the mobile phase using high pressure, while stopping the operation of the SIL-30AC for about 15 minutes.

- Use organic solvent such as IPA, methanol, acetonitrile 100%, 80MPa or more, which is ideal.
- Do not operate the SIL-30AC, while the mobile phase has not filled the flow lines (especially the HPV).

■ After using SIL-30AC

If unused for long periods, wash the flow lines by distilled water or organic solvent.

- When a buffer solution has been used as a mobile phase or rinse solution, the flow lines must be cleaned with distilled or de-ionized water. Otherwise, any remaining buffer solution evaporates and crystallizes over time. This residue could damage the instrument or clog the flow lines (especially the HPV).

7.7 Replacing Sample Loop

⚠CAUTION

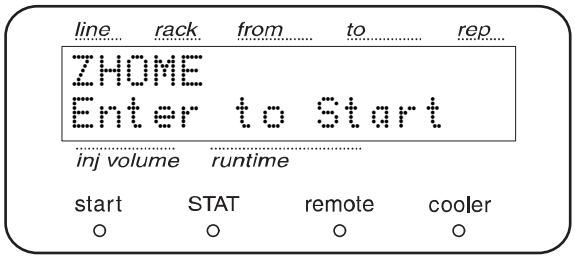
Carry out the replacement, process, and attachment of the sample loop correctly. Or, instrument capability could be worsened or the life of sample loop could be shortened.

Necessary parts

Part	Type	Part No.	Remark
Sample loop ASSY 50 µL	Consumable part	S228-45402-96	0.1 - 50 µL

7.7.1 Before Removing the Sample Loop

1 From the initial screen, press (↑). The [Z HOME] screen is displayed.



2 Press **enter**. The needle rises to the highest position and then moves to the center of the autosampler.

3 Turn the power switch OFF.

4 Unplug the instrument.

7.7.2 Removing the Sample Loop

1 Open the front door and remove panel F from the autosampler.

2 Loosen the male nut at port 1 of the high-pressure valve, which secures the sample loop and remove it.

3 Unscrew and remove the three screws from the Z mount cover, and remove the cover.

4 Using a wrench, unscrew and remove the male nut on the other end of the sample loop (opposite the needle).

5 Remove the sample loop from the hooks (2 places) on the back of the front upper section of the plastic cover inside the instrument.

6 Remove the sample loop from the hooks on the left side of the autosampler's interior.

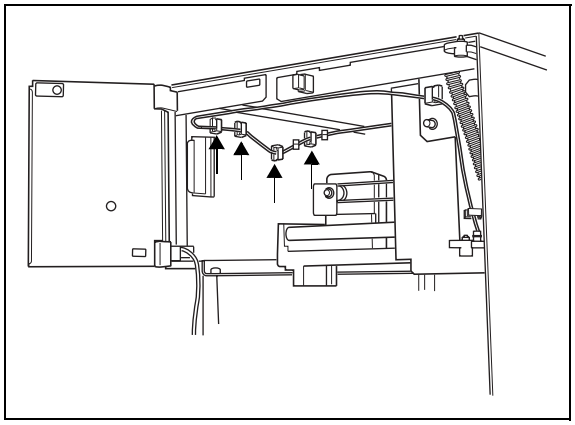


Fig. 7.41

7 Remove the sample loop from the hook of the square hole at the back of the Z mount, and then take the sample loop out of the autosampler.

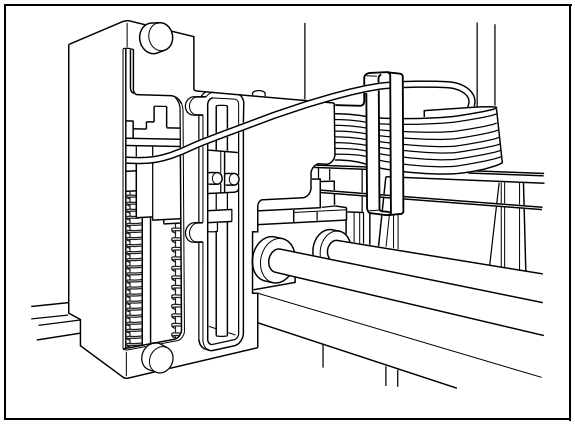


Fig. 7.42

7.7.3 Installing the Sample Loop

1 Pass the new sample loop through the square hole at the back of the Z mount, and insert it through the positioning hook on the right side of the Z mount.

2 Attach a male nut and a ferrule to the sample loop, and secure to the joint on the needle side using a spanner.

3 After attaching the part positioned with the sample loop's band to the hook in the back on the left side of the autosampler's interior, secure the sample loop to the hooks at the center and front in sequence.

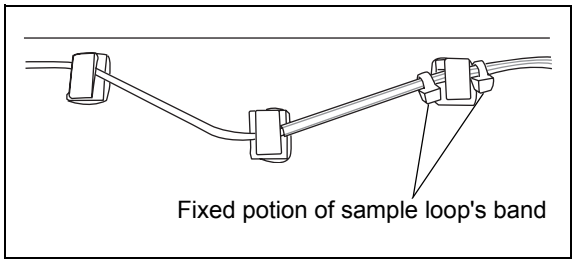


Fig. 7.43

⚠CAUTION

Verify that the fixed portion of sample loop band is lower and the sample loop do not hang loosely behind the Z mount. If the sample loop hangs loosely, it may interfere with the instrument operation and become damaged.

4 Attach the sample loop to the hooks (2 places) on the back of the front upper section of the plastic cover inside the instrument.

5 Secure the sample loop, with the male nut and ferrule attached, to port 1 of the high-pressure valve.

6 Adjust the plumbing for the sample loop attached to port 1 in the way shown on the right.
Bend the tubing downwards along the right of the high-pressure valve and route it at the right of the plastic cover.

7 Replace the Z mount cover, and tighten with its screws.

8 Replace the panel F, and close the front door.

9 Reinsert the power plug and turn the power ON.

10 During initialization, open the panel at the top right of the autosampler and make sure that there is no interference between the sample loop and other parts. In particular, make sure that there is no interference between port 1 of the high-pressure valve and the bottom of the Z mount.

⚠ CAUTION

When using the 50 μL sample loop ASSY, set [SAMPLE SPEED] to 5 μL/sec or less.
☞ "[SAMPLE SPEED]" P. 84
If the speed is fast, the injection volume may not be precise or may be reproducible.

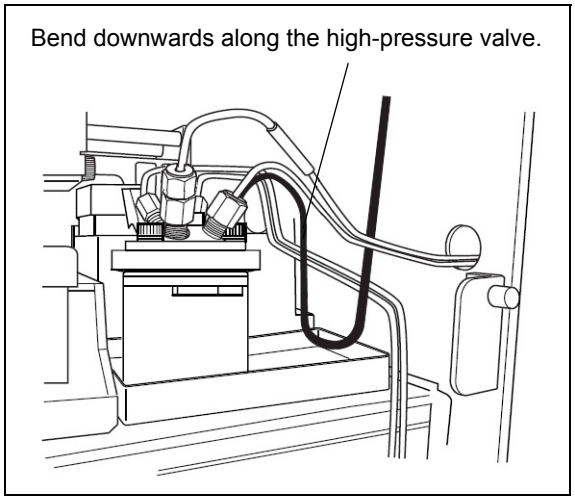


Fig. 7.44

7.8 Inspecting (Replacing) and Ultrasonic Bath Cleaning of Suction Filter

Necessary parts

Part	Type	Part No.
Suction filter SUS	Consumable part	S228-45707-91

7.8.1 Removing the Suction Filter

1. Turn the power switch OFF.
2. Unplug the instrument.
3. Pull the suction filter out of the tubing.
4. Put the suction filter in a bath of 2-propanol, and clean with an ultrasonic cleaning device for 5 minutes.

7.8.2 Installing the Suction Filter

1 Insert the suction tubing into the suction filter.

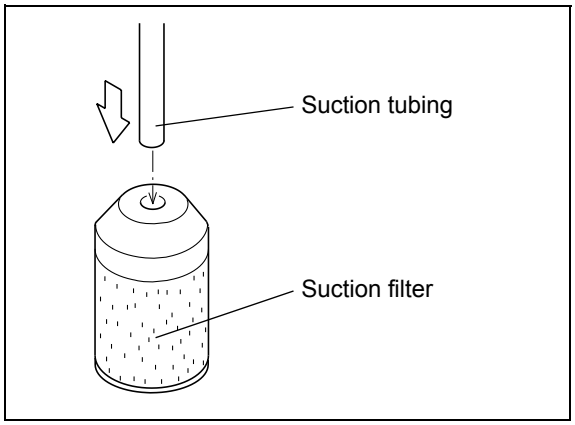


Fig. 7.45

2 Plug in the instrument.

3 Turn the power switch ON.

4 Use a manual syringe and draw three kinds of rinse solution from the reservoir into three flow lines.

5 Press (purge) to start purging of the flow lines.

6 Check that air bubbles do not accumulate inside the solvent tubing.
If bubbles build up, remove the suction filter using the above procedure and replace it with a new one.

7.9 Replacing Needle

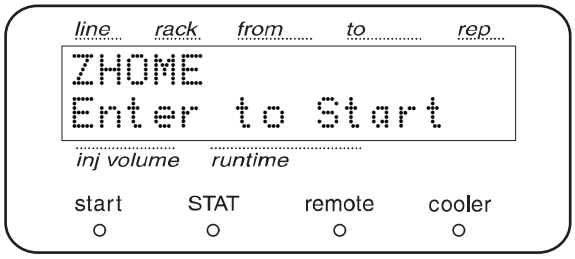
Necessary parts

Part	Type	Part No.	Remark
Needle Coating 30A	Consumable part	S228-41024-95	As an identification mark, a red tube is attached to the bottom of the needle. A ferrule and male nut are provided.

7.9.1 Before Replacing the Needle

1 From the initial screen, press (↑). The [Z HOME] screen is displayed.

2 Press **enter**. The needle rises to the highest position and then moves to the center of the autosampler.



3 Turn the power switch OFF.

4 Unplug the instrument.

7.9.2 Removing the Needle

1 Open the front door and remove front panel from the autosampler.

2 Loosen the 3 mounting screws, and pull the Z mount's cover forward to remove it.

3 Unscrew and remove the male nut of the needle with a wrench.

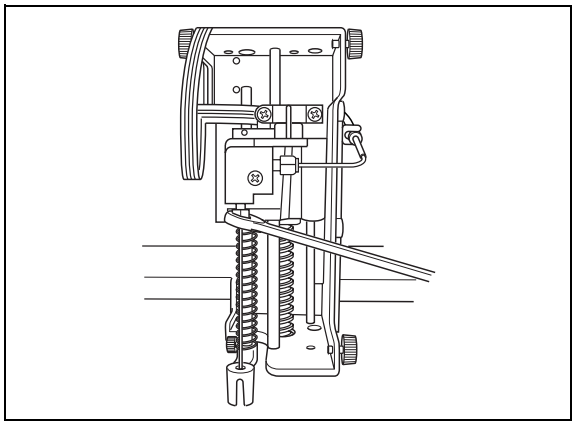


Fig. 7.46

7.9.3 Installing the Needle

1 Attach the male nut and the ferrule to a new needle, finger-tighten the male nut, and further turn it 180 degrees using a wrench.

⚠CAUTION

- Insert the needle fully into the connection, then tighten with a wrench. If the needle is not inserted fully in the hole, a dead volume is created resulting in peak diffusion or cross-contamination.
- Tighten the nut well. A loose fitting mary leak.
- Be sure to use the correct ferrule (Ferrule 1.2F) that cones with the new needle. Using a normal size ferrule (Ferrule 1.6F) may cause a leak.

2 Replace the Z mount cover, with its screws.

3 Reinstall the panel F.

4 Plug the instrument.

5 Turn the power switch ON.

6 Open the autosampler's right cover, and check the position at which the needle is lowered into the injection port. Adjust the needle position if it is incorrect.

☞ "[ADJUST INJ PORT]" P. 151

NOTE

In the event that contamination increases after the original needle is re-attached following maintenance, replace the needle with a new one.

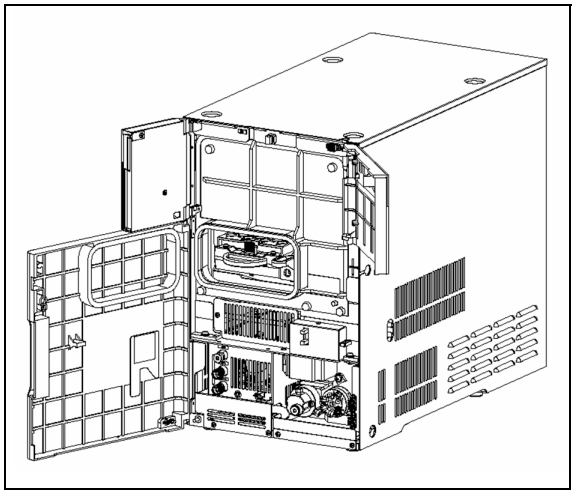


Fig. 7.47

7.10 Replacing Fuse

⚠WARNING

- Before replacing fuses, turn off the power and unplug the instrument.
- For replacement, only use fuses of the correct type and rating.

Failure to heed the above could result in fire, electric shock or short circuits.

The correct rating of the fuses is:

Necessary parts

Part	Type	Part No.
Fuse 218 06.3	Replacement part	S072-02004-24

1 Turn the power OFF.

2 Remove the fuse holder at the back of the autosampler using, for example, a flat-head screwdriver.

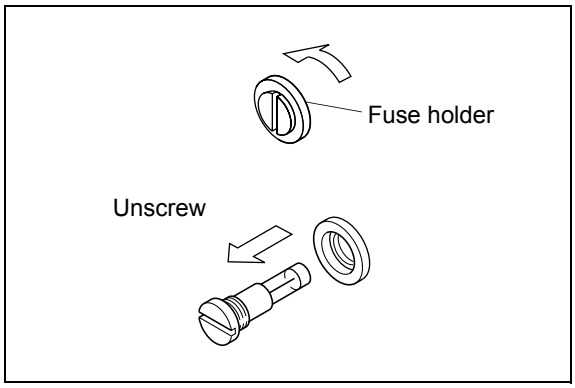


Fig. 7.48

3 Place new fuses into the fuse holder.

4 Push the fuse holder in and fix by using a flat-head screwdriver.

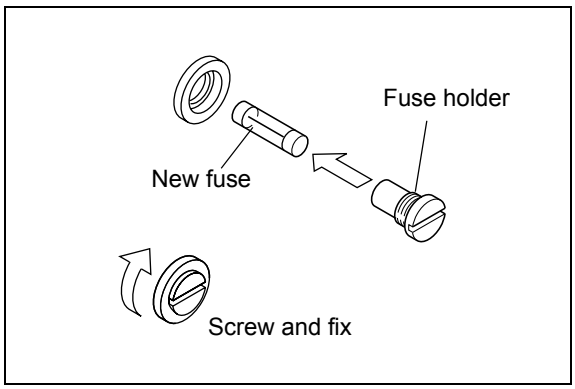


Fig. 7.49

7.11 Replacing Panel F

Necessary parts

Part	Type	Part No.
Panel F	Replacement part	S228-50487-92

1 Turn the power OFF.

2 Open the door.

3 Loosen the panel mounting screws with a screw driver, slide the panel right, and pull it forward to detach it. Remove the panel F.

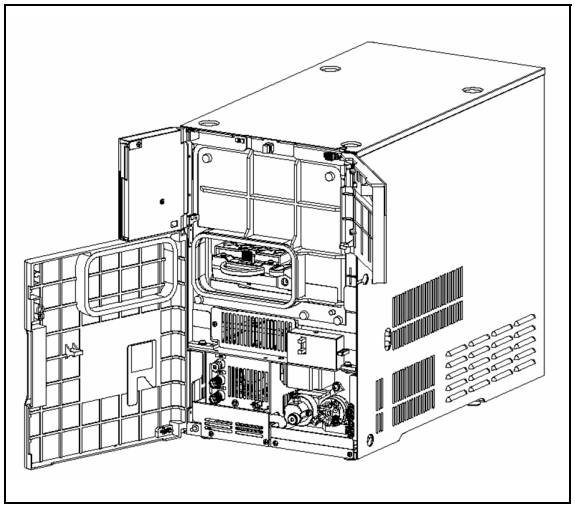


Fig. 7.50

4 Install a new panel F.

⚠CAUTION

Install the panel F to seal the enclosure.
Or, the condensation could be formed when air enters the sample cooler.

5 Close the door.

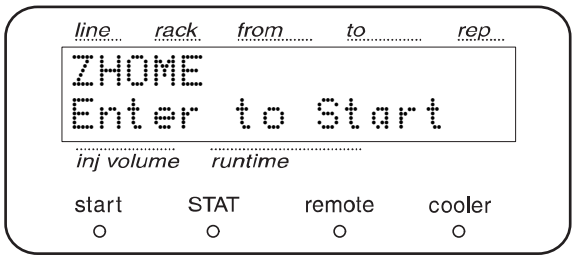
7.12 Replacing Rinsing Port Cap

Necessary parts

Part	Type	Part No.	Remark
Rinsing port cap (NO HOLE) 10 pieces per kit	Consumable part	S228-48331-91	When rinse solution contains highly-volatile acid (Eg.: formic acid, TFA, acetic acid, etc.)
Rinsing port cap 10 pieces per kit	Consumable part	S228-48331-92	When using other rinse solutions (Even when the above mentioned rinse solution is used, use rinsing port caps with hole if there is a problem of cross-contamination.)

7.12.1 Before Replacing the Rinsing Port Cap

1 From the initial screen, press (⇑). The [Z HOME] screen is displayed.



2 Press **enter**. The needle will go up to the top and move to the center of the instrument.

3 Turn the power switch OFF.

4 Unplug the power plug from the outlet.

7.12.2 Replacing the Rinsing Port Cap

1 Remove the rinsing port cover from the rinsing port.

2 Remove the two caps on the rinsing port cover.

3 Install the new caps on the rinsing cover.

⚠CAUTION

Fit the caps firmly all the way in. If they are loose, they may touch the Z mount.

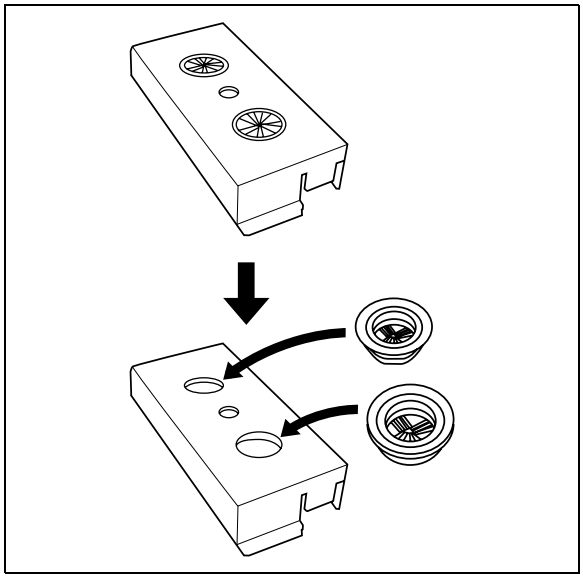


Fig. 7.51

4 Restore the rinsing port cover to its original place.

⚠CAUTION

Fit the cover firmly all the way in. If it is loose, it may touch the Z mount.

5 Move the Z mount gently with your hand, and make sure it does not touch the rinsing port cover. (gap of 1 mm min)

6 Restore the panel F to its original place.

7 Put the power plug into the outlet.

8 Turn the power switch ON.

9 Press the **rinse** key, and make sure the rinsing is done without any problem. (gap of 1 mm min.)

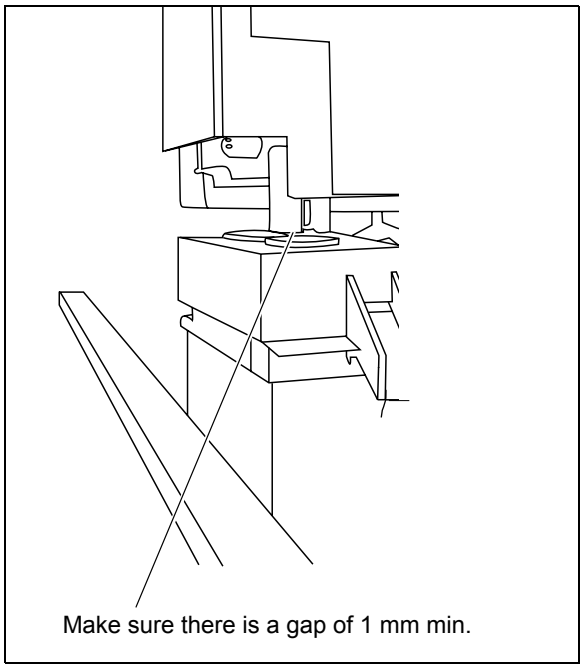


Fig. 7.52

7.13 Rinsing Flow Lines

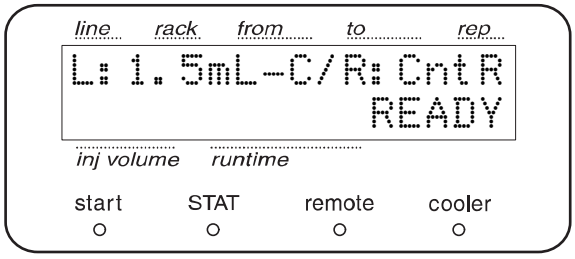
If there is clogging inside the needle or the sample loop, or if there is contamination on the needle surface, the inside and outside of the needle can be rinsed with the mobile phase.

7.13.1 Rinsing Needle and Sample Loop

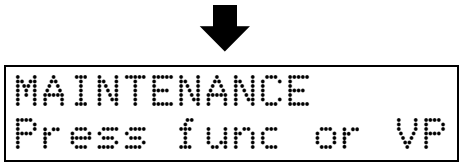
NOTE

[NDLE FLUSH] is allowed to use when total injection method (INJECTION TYPE: 0) is selected

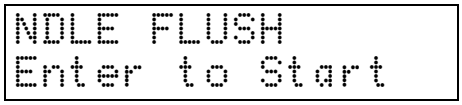
1 Press **CE** to display the initial screen.



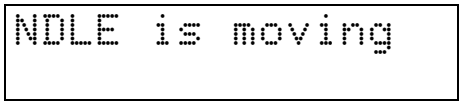
2 Press **VP** repeatedly until the Maintenance Information screen on the right is displayed.



3 Press **func** repeatedly until the [NDLE FLUSH] screen on the right is displayed.



4 Press **pump** on the pump.
Start pumping at 2 mL/min for 5 seconds and then stop pumping.



5 Press **enter**.
The screen on the right is displayed, the needle moves to the rinsing port, and the high-pressure valve switches to [INJ]. (The pump and the needle become connected.)

6 Pump mobile phase with the pump to wash away any clogging or contamination in the needle.

NOTE

Replace the needle if it is not possible to remove the clogging or the contamination.
"7.9 Replacing Needle" P. 262

7 When rinsing the inside of the needle is completed, stop the pump by pressing (**pump**).

8 Press (**enter**).
The needle returns to the injection port.

9 Press (**CE**) to return to the initial screen.

7.13.2 Reverse Rinsing of the Flow Lines

NOTE

If clogging is observed in the flow line inside the instrument with the total injection method, it may be possible to remove clogs by pumping when the SUS tubing HP IN is connected to No. 5 of the high-pressure valve and the preheat block or SUS tubing HP OUT is connected to No. 6 of the high-pressure valve in reverse.

1 Disconnect the inlet tubing from the high-pressure valve and the outlet tubing from the high-pressure valve and the column. Connect the inlet tubing to port 5 of the high-pressure valve.
For details on the procedure for removing or tightening UHPLC fittings at the outlet tubing, refer to the Nexera X2 SYSTEM GUIDE.
☞ Nexera X2 SYSTEM GUIDE, "2.3.4 Prior to Plumbing the High Pressure Flow Line"

2 Connect the end of the outlet tubing, originally connected to the column side, to port 6 of the high-pressure valve.
Mobile phase flows from the other side (originally connected to the high-pressure valve side) of the outlet tubing. Collect it in, for example, a beaker.

3 Pump 2-propanol into the autosampler from the solvent delivery module at 2-5 mL/min.

4 Return the plumbing to the original state.

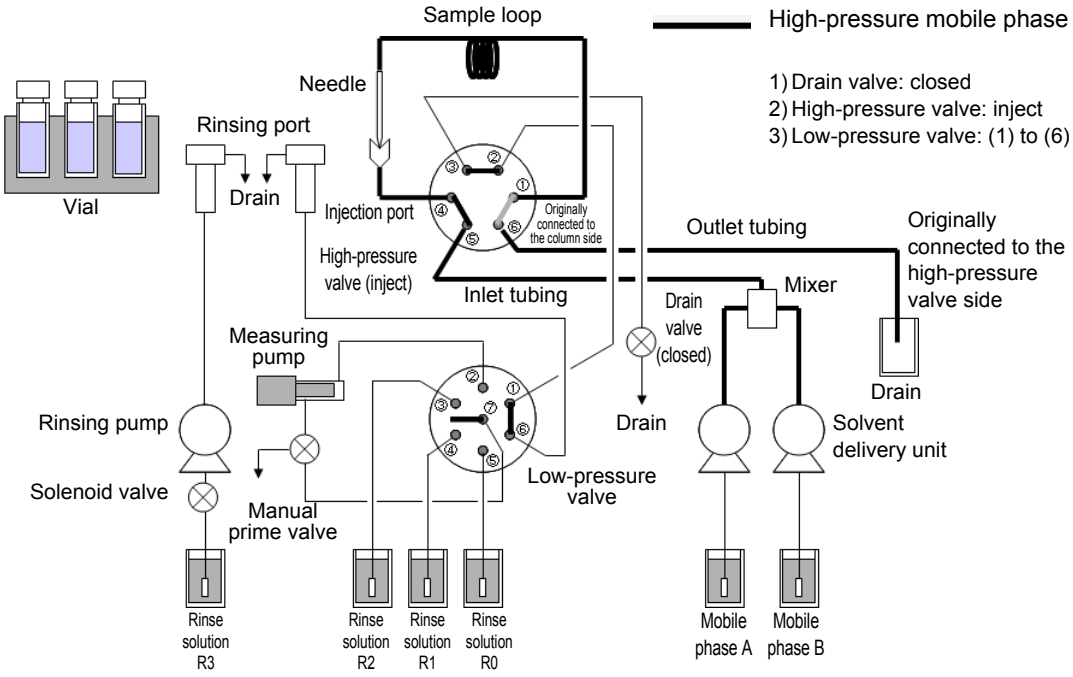

Sample loop
High-pressure mobile phase
1) Drain valve: closed
2) High-pressure valve: inject
3) Low-pressure valve: (1) to (6)
Needle
Rinsing port
Vial
Drain
Injection port
Originally connected to the column side
Outlet tubing
Originally connected to the high-pressure valve side
High-pressure valve (inject)
Inlet tubing
Mixer
Drain valve (closed)
Measuring pump
Drain
Rinsing pump
Solvent delivery unit
Solenoid valve
Low-pressure valve
Manual prime valve
Rinse solution R3
Rinse solution R2
Rinse solution R1
Rinse solution R0
Mobile phase A
Mobile phase B

Fig. 7.53

7.14 Replacing the Outlet Tubing

⚠CAUTION

Before replacing the clogged outlet tubing, first check the procedure "7.13.2 Reverse Rinsing of the Flow Lines". If the clogs remain after above action, replace the outlet tubing following the procedure below. For details about the tubing connection and precautions such as bends, refer to the Nexera X2 SYSTEM GUIDE.

Necessary parts

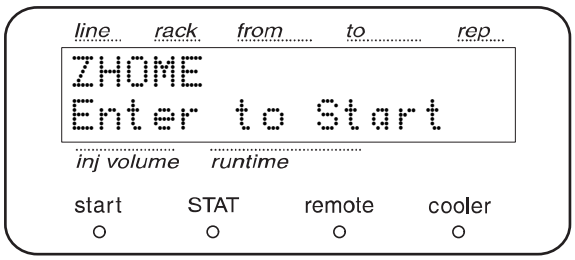
Part	Type	Part No.	Remark
SUS tubing HP OUT (0.1 × 600 mm)	Consumable part	S228-53184-91	ID 0.1 mm, length 600 mm (For the CTO-20A/20AC)
Preheat block (0.1 × 800 mm)	Consumable part	S228-52597-43	ID 0.1 mm, length 800 mm, with pre-heat block (For the CTO-30A)

NOTE

Depending on the column type or manufacturer, the connecting port shape may vary. In the event that cross contamination occurs or peaks are affected due to the difference in the shape when the column has been replaced, replace the outlet tubing with a new one.

7.14.1 Before Removing the Outlet Tubing

1 From the initial screen, press (↑). The [Z HOME] screen is displayed.



2 Press **enter**. The needle will go up to the top and move to the center of the instrument.

3 Turn the power switch OFF, and unplug the instrument.

4 Open the front door and the panels on the right and left, and remove panel F.

5 Remove the outlet tubing of the instrument from the column.
For details on the procedure for removing or tightening UHPLC fittings at the outlet tubing, refer to the Nexera X2 SYSTEM GUIDE.
☞ Nexera X2 SYSTEM GUIDE, "2.3.4 Prior to Plumbing the High Pressure Flow Line"

6 Move the column oven to the right and make space between autosampler and column oven.

7.14.2 Replacing the Outlet Tubing

1 Remove the outlet tubing from port No. 5 of the high-pressure valve. For details on the procedure for removing or tightening UHPLC fittings at the outlet tubing, refer to the Nexera X2 SYSTEM GUIDE.

Nexera X2 SYSTEM GUIDE, "2.3.4 Prior to Plumbing the High Pressure Flow Line"

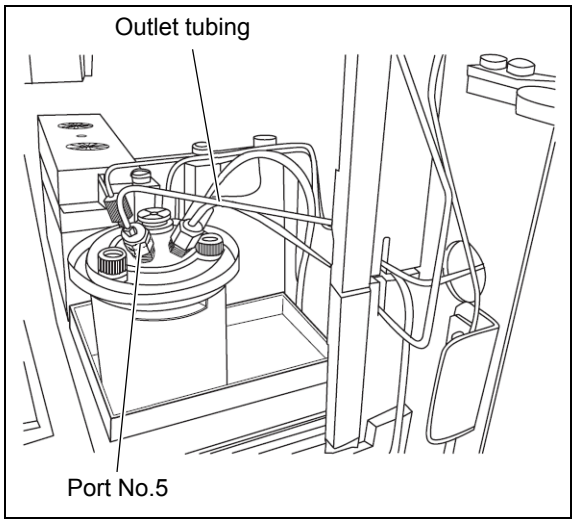


Fig. 7.54

2 Straighten the wound portion of the new outlet tubing as shown in the figure.

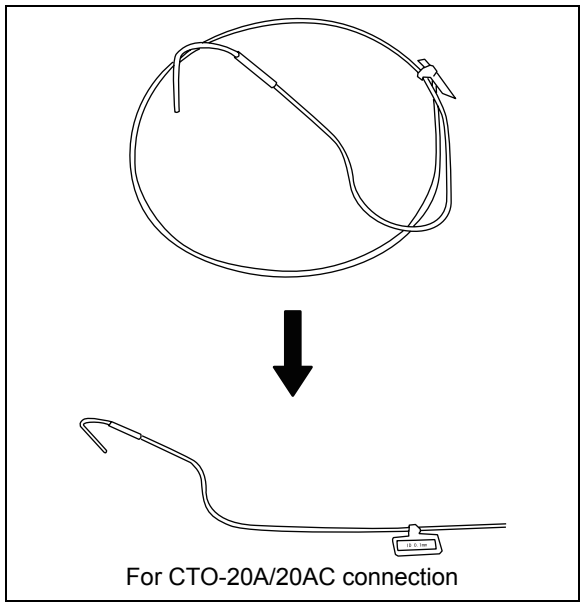


Fig. 7.55

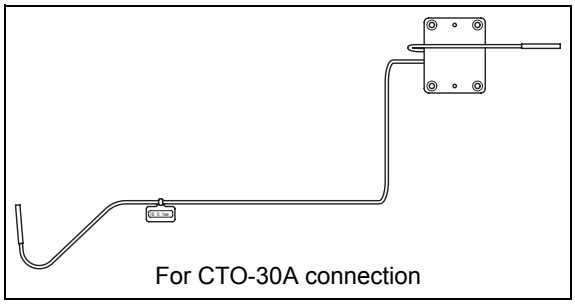


Fig. 7.56

3 Insert the end of the outlet tubing, equipped with the ID identification mark, from the outside into the round hole at the side panel.

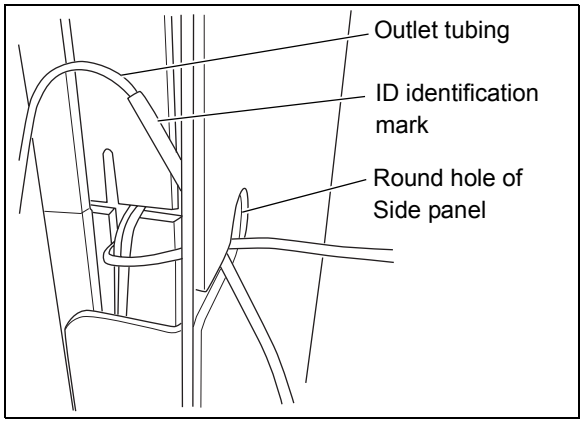


Fig. 7.57

4 Insert the outlet tubing into the round hole at the left side of the column oven.
Slide the ID identification tags necessary to properly install the tubing. After the tubing is installed, slide the tag back to the original position.

⚠CAUTION

Do not leave the tag inside the column oven.
The tag may melt because of the high temperature.

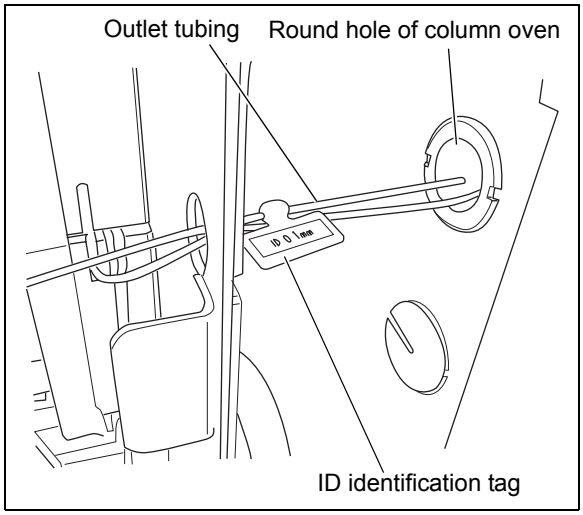


Fig. 7.58

5 Attach the UHPLC fitting to the end of the outlet tubing, and connect it to port No. 5 using a wrench.
Keep the tubing passing through the upper side of the "+" shaped slit on the plastic cover.

6 Attach the UHPLC fitting to the end of the outlet tubing, and connect the outlet tubing to the column.

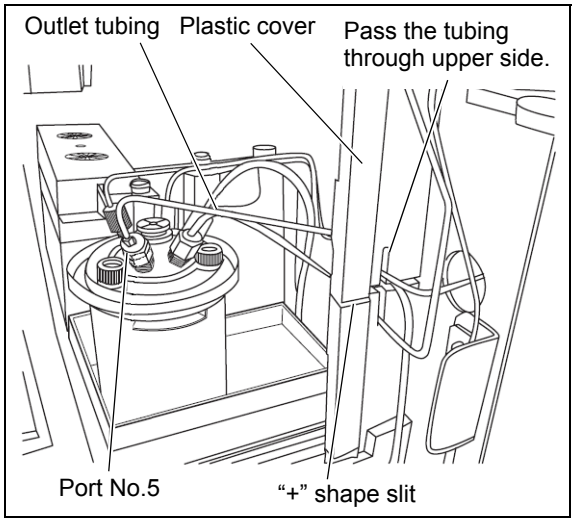
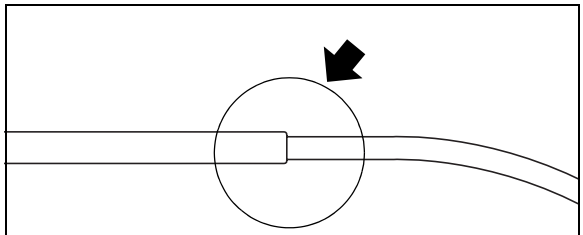


Fig. 7.59

7 Return the panel F.



The tubing is more likely to break at the ends where the diameter is reduced. Be careful not to bend the tubing more than 45 degrees.

Fig. 7.60

⚠CAUTION

If the tubing is bent at the same position repeatedly, the strength of tubing will be lowered and a rupture or cracks may occur, which could result in mobile phase leaks.
When bending the SUS tubing, making a bending radius (curvature radius) too small will deform the inner diameter of the tubing, and this could cause clogging or pressure increases in the tubing. Do not bend the tubing excessively, such as pinching it using pliers or similar tools and bending it to an acute angle. Also, do not bend and straighten out the same portion repeatedly. This will weaken the tubing, with the risk that it will break.

7.15 Exterior Cleaning

If the instrument cover or front panel becomes dirty, wipe it clean with a soft dry cloth or tissue paper.
For persistent stains, clean the exterior using the following procedure.

1. Dip a piece of cloth in a dilute neutral detergent and twist firmly to remove excess liquid. Use this cloth to scrub the soiled area of the exterior surface of the instrument.

2. Dip a piece of cloth into water and twist firmly to remove excess liquid. Use this cloth to wipe away all the remaining detergent. Use a dry cloth to remove all moisture from the exterior surface of the instrument.

⚠CAUTION

Do not allow spilled water to remain on the instrument surface, and do not use alcohol or thinner-type solvents to clean the surfaces. These can cause rusting and discoloration.

7.16 Maintenance for Long Periods without Use

If the autosampler is not used for a long period, raise the needle to prevent a reduction in the service life of the needle seal.

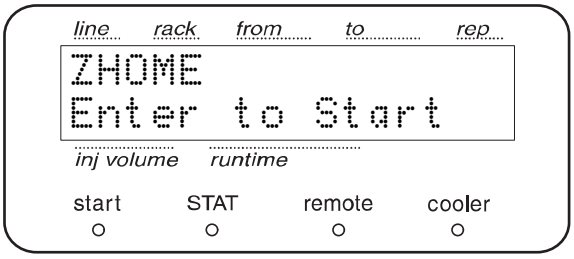
7.16.1 Moving the Needle

1 Turn the power ON.

2 From the initial screen, press (↑). The [Z HOME] screen is displayed.

3 Press **enter**. The needle rises to the highest position and then moves to the center of the autosampler.

4 Turn the power OFF.



7.17 Maintenance for Continuous Use over Extended Periods

If the instrument is used with the sample cooler set at a temperature lower than the room temperature, dew condensation water may gather between the sample rack and the rack cooling plate.
In such a case, wipe the water as below.

1 From the initial screen, press (↑). The [Z HOME] screen is displayed.

2 Press **enter**. The needle rises to the highest position and then moves to the center of the autosampler.

3 Remove the sample rack.

4 Remove the panel F.
"7.11 Replacing Panel F" P. 265

5 Wipe water off the rack cooling plate with soft cloth or paper.

⚠CAUTION

If your hand contacts the needle, you may sustain an injury due to the tip of the needle.

6 Attach the panel F.

7 Mount the sample rack.

8 Press **enter**. The needle moves back to the injection port.

8 Technical Information

8.1 Specifications

8.1.1 Sample Injection

Item	Specification	
Injection system	Total injection, variable injection volume type (zero sample loss during injection) Loop injection, adjustable or fixed volume loop injection (option)	
Injection volume setting range	Total injection	0.1 - 50 μL (0.1 - 9.9 μL: 0.1 μL increments, 10 - 50 μL: 1 μL increments)
	Loop injection	Select either loop of 5 μL or 20 μL capacity. (0.1 - 9.9 μL: 0.1 μL increments, 10 - 20 μL: 1 μL increments)
Number of samples processed	175 (w/1 mL sample vials), 105 (w/1.5 mL sample vials), 50 (w/4 mL sample vials), 192 (w/2 microtiter plates each with 96 wells), 768 (w/2 microtiter plates each with 384 wells), 192 (w/2 Deep well MTP each with 96 wells), 768 (w/2 Deep well MTP each with 384 wells)	
Sample vials	1.5 mL glass, 1.1 mL glass, 1 mL glass, 4 mL glass, 0.3 mL glass (w/plastic spacers), 0.3 mL glass (4 mL vial storage type), 1 mL plastic, 0.2 mL plastic, 4 mL plastic, 96-well microtiter plate, 384-well microtiter plate, 96-well deep-well plate, 384-well deep-well plate	
Injection volume repeatability	Total injection	RSD ≤ 0.25 % (5 μL injected)
	Loop injection	RSD ≤ 1.0 % (5 μL loop used, partial loop method, 2 μL injected)
		RSD ≤ 1.0 % (20 μL loop used, partial loop method, 5 μL injected)
Carryover	Total injection	0.0015 % max. (Internal and external rinsing of the needle is not performed, caffeine 5 μL injected)
	Loop injection	0.005 % max. (Internal and external rinsing of the needle is performed, Caffeine injected both partial loop method and full loop method) (In case of partial loop method, 3 μL injected at 5 μL loop used, 5 μL injected at 20 μL loop used)
Injection volume accuracy	±1% (total volume injection, at 50 μL injection, n = 10)	
Number of repeat injections	1 - 30 times/sample	
Analysis time setting	0.01 minute steps (< 1000 minutes), 0.1 minute steps (> 1000 minutes)	
Number of sample table steps	100 steps max	

Item	Specification
Sample aspiration rate	0.1 - 15 µL/sec (0.1 µL/sec increments)
Rinse aspiration rate	Variable (1 to 35 µL/sec, 1 µL/increments)
Maximum pressure	130 MPa

8.1.2 Sample Cooler

Item	Specification
System	Direct cooling system (environment conditions: room temperature below 30 °C with humidity less than 70% when the cooler temperature is set to 4 °C), dehumidification function included
Range to set temperature	4 to 40 °C (Room temperature needs to be 30 °C or lower and the humidity 70% or less to go down to 4 °C.)
Temperature accuracy	±3 °C (±6 °C for microtiter plate and deep-well plate. Not cooled below 1 °C.)

8.1.3 Others

<table>
<tr><th>Item</th><th colspan="5">Specification</th></tr>
<tr><td>Liquid contact materials</td><td colspan="5">Stainless steel (SUS316L, SUS316), ceramic, PTFE, ETFE, FEP, GFP, sapphire, PEEK, Polyimide</td></tr>
<tr><td>Ambient temperature</td><td>4-35 °C</td><td colspan="4" rowspan="2">Cooling down to 4 °C is possible at a room temperature of 30 °C, humidity of 70% or lower.</td></tr>
<tr><td>Humidity range</td><td>20-85%</td></tr>
<tr><td>pH range</td><td colspan="5">1 to 9 (standard), 1 to 14 (option)</td></tr>
<tr><td>Dimensions</td><td colspan="5">W260 × H415 × D500 mm</td></tr>
<tr><td>Mass</td><td colspan="5">33 kg</td></tr>
<tr><td rowspan="4">Power supply</td><td>Part No.</td><td>Power Supply Voltage (indicated on the instrument)</td><td>Power Consumption</td><td>Frequency</td><td>Rated Breaking Capacity *</td></tr>
<tr><td>S228-45157-31
S228-45157-41</td><td rowspan="3">AC100 - 240 V</td><td rowspan="3">300 VA</td><td rowspan="3">50/60 Hz</td><td rowspan="3">63A</td></tr>
<tr><td>S228-45157-32
S228-45157-42</td></tr>
<tr><td>S228-45157-38
S228-45157-48
S228-45157-58</td></tr>
<tr><td>Installation Environment (IEC)</td><td colspan="5">Installation Category II
Pollution Degree 2
Altitude 2000 m or lower
Install inside the room.</td></tr>
<tr><td>Error display</td><td colspan="5">Exists (Error display and stop at the time of malfunction)</td></tr>
</table>

* Connect the instrument to a power outlet that is equipped with a circuit breaker that shuts off the current at the described value or less.

8.2 Maintenance Parts

8.2.1 Consumable Parts

Part	Part No.	Compatibility ① 30AC only ② XR, HT (UFLC) ③ SIL-20A Series	Remark
Needle seal 30A (standard)	S228-52253	①	pH1 to 9 Maximum withstand pressure 130 MPa
PEEK needle seal 30A (option)	S228-53178-91	①	pH1 to 14 (option) Maximum withstand pressure 66 MPa
Needle seal 30A kit	S228-52401-92	①	3 needle seals 30A (standard) and 1 housing needle seal 30A in a set
Needle coating 30A	S228-41024-95	①	With an identification mark (red tube) With a ferrule and male nut
Plunger seal	S228-35145	③	
Low-pressure valve rotor 30A	S228-51922	①	
Low-pressure valve stator 30A	S228-51663-01	①	
High-pressure valve rotor 30A	S228-52139	①	
High-pressure valve stator 30A	S228-48858-95	①	
Sample loop ASSY 50 μL	S228-45402-96	③	
Suction filter SUS	S228-45707-91	③	
Rinsing port cap set (NO HOLE)	S228-48331-91	③	Set of 10 rinsing port cap (S228-47973-01)
Rinsing port cap set	S228-48331-92	③	Set of 10 rinsing port cap (S228-47973-02)
Preheat block (0.1 × 800 mm)	S228-52597-43	①	For CTO-30A With pre-heat block
SUS tubing HP OUT (0.1 × 600 mm)	S228-53184-91	②	For CTO-20A/20AC
Vial detection spring	S034-01615-09	③	
UHPLC fitting	S228-56867-41	②	

8.2.2 Replacement Parts

■ Fuse

Part	Part No.	Remark
Fuse, 218 06.3	S072-02004-24	

■ Autosampler Base Parts

Part	Part No.	Compatibility ① 30AC only ② XR, HT (UFLC) ③ SIL-20A Series	Remark
Panel F	S228-50487-92	③	

■ High-Pressure Valve Parts

Part	Part No.	Compatibility ① 30AC only ② XR, HT (UFLC) ③ SIL-20A Series	Remark
Housing needle seal 30A	S228-52228	①	
Cap needle seal 30A	S228-51904	①	

■ Valve ASSY

Part	Part No.	Compatibility ① 30AC only ② XR, HT (UFLC) ③ SIL-20A Series	Remark
Measuring pump plunger	S228-35010-92	③	
Manual prime valve	S228-52608-91	①	
Packing LV,7P	S228-51923	③	

■ Tubing Parts

Part	Part No.	Compatibility ① 30AC only ② XR, HT (UFLC) ③ SIL-20A Series	Remark
Ferrule	S228-16000-10	③	
Male nut, 1.6MN	S228-16001	③	
Male nut, 1.6MN, W6	S228-16001-03	③	
SUS tubing HP IN (0.3 × 300 mm)	S228-53184-92	①	
SUS tubing HP IN (0.1 × 600 mm)	S228-53184-94	①	For loop injection method

■ Service Tools

Part	Part No.	Compatibility ① 30AC only ② XR, HT (UFLC) ③ SIL-20A Series	Remark
Syringe needle, lock type	S046-00002	③	
Syringe needle ASSY	S228-18216-91	③	
Syringe, L-208	S046-00038-01	③	
Adapter, for syringe	S228-15672-91	③	
Double-ended wrench 6 × 8	S086-03003	③	
Double-ended wrench 8 × 10	S086-03006	③	
Tool for needle seal XR	S228-50570	②	
Jig, rack teaching	S228-50895-91	③	
Allen wrench (3 mm)	S086-03804	③	
Allen wrench (4 mm)	S086-03805	③	
Seal installer tool	S228-25142-01	③	
Tool plunger	S228-34672-02	③	
Jig rotor	S228-48899-91	②	

■ Others

Part	Part No.	Compatibility ① 30AC only ② XR, HT (UFLC) ③ SIL-20A Series	Remark
Control vial rack cover	S228-45417-91	③	
1.5 mL sample vial cooling rack cover	S228-50759-91	–	105 vials

8.2.3 Maintenance Kit

We offer maintenance kit ASSY consisting of consumable parts and tubing parts shown below.

■ Maintenance Kit SIL-30AC (Part No. S228-45413-98)

Part Name	Part No.	Compatibility ① 30AC only ② XR, HT (UFLC) ③ SIL-20A Series	Q'ty	Remark
Needle seal 30A (standard)	S228-52253	①	1	Polyimide
High-pressure valve stator 30A	S228-48858-95	①	1	
High-pressure valve rotor 30A	S228-52139	①	1	
Low-pressure valve rotor 30A	S228-51663-01	①	1	
Low-pressure valve stator 30A	S228-51663-01	①	1	
Needle coating 30A	S228-41024-95	①	1	With an identification mark (black tube) With a ferrule and male nut
Sample loop ASSY 50μL	S228-45402-96	③	1	
Plunger seal	S228-35145	③	1	
Suction filter	S228-45707-91	③	1	
Set of 10 rinsing port cap (without holes)	S228-48331-91	③	1	
Set of 10 rinsing port cap (with holes)	S228-48331-92	③	1	
Vial detection spring	S034-01615-09	③	1	

Index

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