Rittun

User's Guide

L-3320 Autosampler

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Rittun (Suzhou) Precision Measurement System Co., Ltd.

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If you have any problem or requirement when using our products or this manual, please contact Rittun.

E-mail: service@rittun.com
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Safety Requirement

General Safety Summary

Please review the following safety precautions carefully before putting the instrument into operation so as to avoid any personal injury or damage to the instrument and any product connected to it. To prevent potential hazards, please use the instrument only specified by this manual.

Use Proper Power Cord.
Only the power cord designed for the instrument and authorized for use within the local country could be used.

Ground the Instrument.
The instrument is grounded through the Protective Earth lead of the power cord. To avoid electric shock, it is essential to connect the earth terminal of the power cord to the Protective Earth terminal before connecting any inputs or outputs.

Connect the Probe Correctly.
If a probe is used, do not connect the ground lead to high voltage since it has isobaric electric potential as the ground.

Observe All Terminal Ratings.
To avoid fire or shock hazard, observe all ratings and markers on the instrument and check your manual for more information about ratings before connecting the instrument.

Use Proper Overvoltage Protection.
Make sure that no overvoltage (such as that caused by a thunderstorm) can reach the product, or else the operator might be exposed to the danger of electrical shock.

Do Not Operate Without Covers.
Do not operate the instrument with covers or panels removed.

Do Not Insert Anything Into the Holes of Fan.

Do not insert anything into the holes of the fan to avoid damaging the instrument.

Use Proper Fuse.

Please use the specified fuses.

Avoid Circuit or Wire Exposure.

Do not touch exposed junctions and components when the unit is powered.

Do Not Operate With Suspected Failures.

If you suspect damage occurs to the instrument, have it inspected by qualified service personnel before further operations. Any maintenance, adjustment or replacement especially to circuits or accessories must be performed by Rittun authorized personnel.

Keep Well Ventilation.

Inadequate ventilation may cause an increase of temperature or damage to the device. So please keep the instrument well ventilated and inspect the intake and fan regularly.

Do Not Operate in Wet Conditions.

In order to avoid short circuiting to the interior of the device or electric shock, please do not operate the instrument in a humid environment.

Do Not Operate in an Explosive Atmosphere.

In order to avoid damage to the device or personal injuries, it is important to operate the device away from an explosive atmosphere.

Keep Product Surfaces Clean and Dry.

To avoid the influence of dust and/or moisture in the air, please keep the surface of the device clean and dry.

Electrostatic Prevention.

Operate the instrument in an electrostatic discharge protective environment to avoid damage induced by static discharges. Always ground both the internal and external conductors of cables to release static before making connections.

Proper Use of Battery.

If a battery is supplied, it must not be exposed to high temperature or in contact with fire. Keep it out of the reach of children. Improper change of battery (note: lithium battery) may cause explosion. Use Rittun specified battery only.

Handling Safety.

Please handle with care during transportation to avoid damage to buttons, knob interfaces and other parts on the panels.

Safety Terms and Symbols

Terms Used in this Manual. These terms may appear in this manual:

WARNING

Warning statements indicate conditions or practices that could result in injury or loss of life.

CAUTION

Caution statements indicate conditions or practices that could result in damage to this product or other property.

Allgemeine Sicherheits Informationen

Überprüfen Sie diefolgenden Sicherheitshinweise sorgfältigumPersonenschädenoderSchäden am Gerätundan damit verbundenen weiteren Gerätenzu vermeiden. Zur Vermeidung vonGefahren, nutzen Sie bitte das Gerät nur so, wiein diesem Handbuchangegeben.

Um Feuer oder Verletzungen zu vermeiden, verwenden Sie ein ordnungsgemäßes Netzkabel.
Verwenden Sie für dieses Gerät nur das für ihr Land zugelassene und genehmigte Netzkabel.

Erden des Gerätes.
Das Gerät ist durch den Schutzleiter im Netzkabel geerdet. Um Gefahren durch elektrischen Schlag zu vermeiden, ist es unerlässlich, die Erdung durchzuführen. Erst dann dürfen weitere Ein- oder Ausgänge verbunden werden.

Anschluss einesTastkopfes.
Die Erdungsklemmen der Sonden sindauf dem gleichen Spannungspegel des Instruments geerdet. SchließenSie die Erdungsklemmen an keine hohe Spannung an.

Beachten Sie alle Anschlüsse.
Zur Vermeidung von Feuer oder Stromschlag, beachten Sie alle Bemerkungen und Markierungen auf dem Instrument. Befolgen Sie die Bedienungsanleitung für weitere Informationen, bevor Sie weitere Anschlüsse an das Instrument legen.

Verwenden Sie einen geeigneten Überspannungsschutz.
Stellen Sie sicher, daß keinerlei Überspannung (wie z.B. durch Gewitter verursacht) das Gerät erreichen kann. Andernfallsbestehtfür den Anwender die GefahreinesStromschlages.

Nicht ohne Abdeckung einschalten.
Betreiben Sie das Gerät nicht mit entfernten Gehäuse-Abdeckungen.

Betreiben Sie das Gerät nicht geöffnet.
Der Betrieb mit offenen oder entfernten Gehäuseteilen ist nicht zulässig. Nichts in entsprechende Öffnungen stecken (Lüfter z.B.)

Passende Sicherung verwenden.
Setzen Sie nur die spezifikationsgemäßen Sicherungen ein.

Vermeiden Sie ungeschützte Verbindungen.
Berühren Sie keine unisolierten Verbindungen oder Baugruppen, während das Gerät

in Betrieb ist.

Betreiben Sie das Gerät nicht im Fehlerfall.
Wenn Sie am Gerät einen Defekt vermuten, sorgen Sie dafür, bevor Sie das Gerät wieder betreiben, dass eine Untersuchung durch qualifiziertes Kundendienstpersonal durchgeführt wird.Jedwede Wartung, Einstellarbeiten oder Austausch von Teilen am Gerät, sowie am Zubehör dürfen nur von Rittun autorisiertem Personal durchgeführt werden.

Belüftung sicherstellen.
Unzureichende Belüftung kann zu Temperaturanstiegen und somit zu thermischen Schäden am Gerät führen. Stellen Sie deswegen die Belüftung sicher und kontrollieren regelmäßig Lüfter und Belüftungsöffnungen.

Nicht in feuchter Umgebung betreiben.
Zur Vermeidung von Kurzschluß im Geräteinneren und Stromschlag betreiben Sie das Gerät bitte niemals in feuchter Umgebung.

Nicht in explosiver Atmosphäre betreiben.
Zur Vermeidung von Personen- und Sachschäden ist es unumgänglich, das Gerät ausschließlich fernab jedweder explosiven Atmosphäre zu betreiben.

Geräteoberflächen sauber und trocken halten.
Um den Einfluß von Staub und Feuchtigkeit aus der Luft auszuschließen, halten Sie bitte die Geräteoberflächen sauber und trocken.

Schutz gegen elektrostatische Entladung (ESD).
Sorgen Sie für eine elektrostatisch geschützte Umgebung, um somit Schäden und Funktionsstörungen durch ESD zu vermeiden. Erden Sie vor dem Anschluß immer Innen- und Außenleiter der Verbindungsleitung, um statische Aufladung zu entladen.

Die richtige Verwendung desAkku.
Wenneine Batterieverwendet wird, vermeiden Sie hohe Temperaturen bzw. Feuer ausgesetzt werden.Bewahren Sie es außerhalbder Reichweitevon Kindern auf.UnsachgemäßeÄnderung derBatterie(Anmerkung:Lithium-Batterie)kann zu einer Explosion führen. VerwendenSie nur von RittunangegebenenAkkus.

Sicherer Transport.
Transportieren Sie das Gerät sorgfältig (Verpackung!), um Schäden an Bedienelementen, Anschlüssen und anderen Teilen zu vermeiden.

Sicherheits Begriffe und Symbole

Begriffe in diesem Guide. Diese Begriffe können in diesem Handbuch auftauchen:

WARNING
Die Kennzeichnung WARNING beschreibt Gefahrenquellen die leibliche Schäden oder den Tod von Personen zur Folge haben können.

CAUTION
Die Kennzeichnung Caution (Vorsicht) beschreibt Gefahrenquellen die Schäden am Gerät hervorrufen können.

Document Overview

Chapter 1 L-3320 Overview
This chapter provides an overview of the L-3000 HPLC system and briefly introduces the appearance and modules of the L-3320 autosampler.

Chapter 2 To Install L-3320
This chapter introduces the specifications, installation procedures as well as the installation notices of the autosampler.

Chapter 3 To Run L-3320
This chapter introduces how to run the autosampler and the use notices.

Chapter 4 Troubleshooting
This chapter lists the possible failures and the corresponding solutions of the instrument to help users to solve and analyze the common problems.

Chapter 5 Daily Maintenance
This chapter provides the operation methods of daily maintenance as well as guides users to replace some of the modules.

Chapter 6 Hardware Qualification
This chapter introduces the relative information of hardware qualification and guides users to perform hardware qualification.

Chapter 7 Appendix
This chapter provides the accossary list of the instrument and the warranty.

Content

Chapter 1 L-3320 Overview

Main topics of this chapter:

- L-3000 HPLC System Introduction
- L-3320 Introduction
  Appearance
  Modules

L-3000 HPLC System Introduction

L-3000 High Performance Liquid Chromatography (HPLC) system is a series of new products for compound separation and analysis, developed by **R**ittun on the basis of Chromatography Separation Principle – different compound has different solubility, distribution and absorption in two phases (the static is Stationary phase and the other is Mobile phase). Due to different mobile phase, the Chromatography is divided into two types: Gas chromatography (use gas as mobile phase) and Liquid chromatography (use liquid as mobile phase). With the improvement of separation technology, Liquid Chromatography theory develops continuously, from the Classical Liquid Chromatography to the current High Performance Liquid Chromatography. HPLC has more advantages, such as high pressure capabilities, speed, efficiency and sensitivity, matching with the column that has the advantage of high resolution, sensitivity and reuse and is widely used in analytical chemistry, organic chemistry and biochemistry.

The HPLC system is generally composed of pump, sampler, column and data processing equipments. Besides, such devices for example, solvent organizer, autosampler, column oven and fraction collector could also be used additionally to meet various requirements.

Rittun L-3000 HPLC system mainly consists of L-3100 series solvent organizer (L-3100, L-3120 or L-3140), L-3200 series pump (L-3210, L-3220 and L-3245), L-3320 autosampler, L-3400 column oven, L-3500 UV-Vis Detector, column and the workstation (UltraChrom or Rittun CWS). You can also use other devices additionally to improve the quality and efficiency of sample separation and analysis.

Working Process

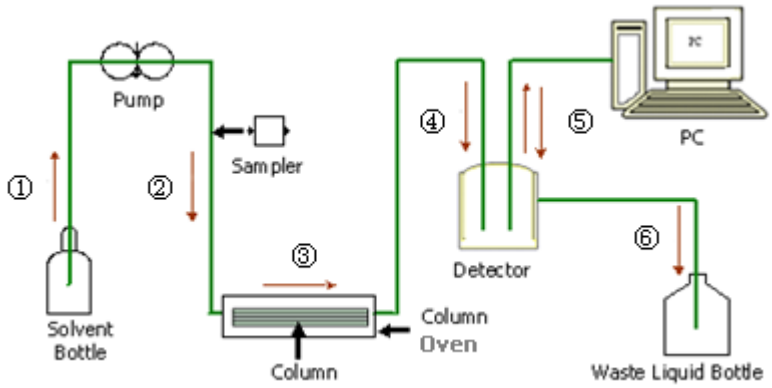


Figure 1-1 Working process of L-3000 HPLC system

Figure 1-1 shows the working process of the L-3000 HPLC system. The mobile phase and sample flow through all components in the direction of arrows to enable the L-3000 HPLC system to perform sample analysis and separation.

1. Driven by pump, the mobile phase is transported from the solvent bottle into the whole system against the resistance from column.
2. The sample to be separated is injected into the system by the sampler and then transmitted into the column carried along by the mobile phase.
3. The sample is separated because different compound has different adsorption on the stationary phase.
4. The separated compounds then flow into the detector. As compounds with different concentrations have various absorptions for monochromatic light, different photo signals are input into the photoelectric receiver and are converted into the related electric signals.
5. The converted electronic signals are transmitted into a data processing system such as PC for qualitative and quantitative analysis and separation.
6. The waste flow into the waste bottle.

Applications

The HPLC system applies to chemical compound separation and analysis, particularly the large molecular compounds, polar compounds, ionic compounds and thermal instability compounds. The system can be widely used in most industries such as pharmaceutical analysis; health epidemic prevention; environmental monitoring; farming, forestry, animal husbandry, sideline production and fishery; petrochemical industry; food inspection; quality test; teaching & researching.

L-3320 Introduction

The autosampler which is used to transport the material (sample) to be separated into the fluid system, is an important component of the HPLC system. The whole process of the injection is as follows: the sample needle enters the sample vial and aspirates the sample into the loop driven by the injection system; then, the injection valve switches to the fluid port and the sample is transported into the fluid system for separation and analysis. The wash system runs when the injection is finished to remove the remained sample in the sample needle and tubings, providing a clean fluid for the next injection.

L-3320 is a complete autosampler with flexible configurations and small size. Standard high or low plate or vial tray can be used. The sampling compartment can house two different well plates at the same time. It features quick injection, high accuracy, good repeatability, low cross contamination as well as easy maintenance. L-3320 configured with partial loopfill injection, full loop injection and μL pickup injection, can provide quick and efficient injection. It, in combination with the chromatography work station, largely improves the automation of the whole system and is an important component of the L-3000 HPLC system.

Appearance

Front Door

L-3320 is configured with a clear front door. The front door is connected to the chassis through magnets and can be easily disassembled. To disassemble the front door, open the front door in the arrow direction in Figure 1-2, push the joint part of the front door and the chassis upwards to separate the magnetic hinge and the chassis. To install the front door, press the magnetic hinge into the front door, align it with the installation position at the front panel of the chassis and then release the magnetic hinge to make it insert into the front panel.

When the front door is removed (as shown in Figure 1-3), you can view the syringe, injection valve, loop and the tubing connections, providing facilitate for timely cleaning, maintenance and change of parts.

An instrument running state VFD (Vacuum Fluorescent Display) is configured at the upper right corner of the front door (as shown in Figure 1-2). No matter whether the front door is removed, the operator can view the running state of the instrument through the VFD. When the autosampler is started, the VFD will display the current running state of the whole system.

Vertical View

As shown in Figure 1-2, there are four round grooves (corresponding to the four round rubber feet at the bottom of the case of other instruments of the system) at the four corners of the top of the autosampler and when the round rubber feet are inserted into the grooves, the whole system would be in relatively stable state.

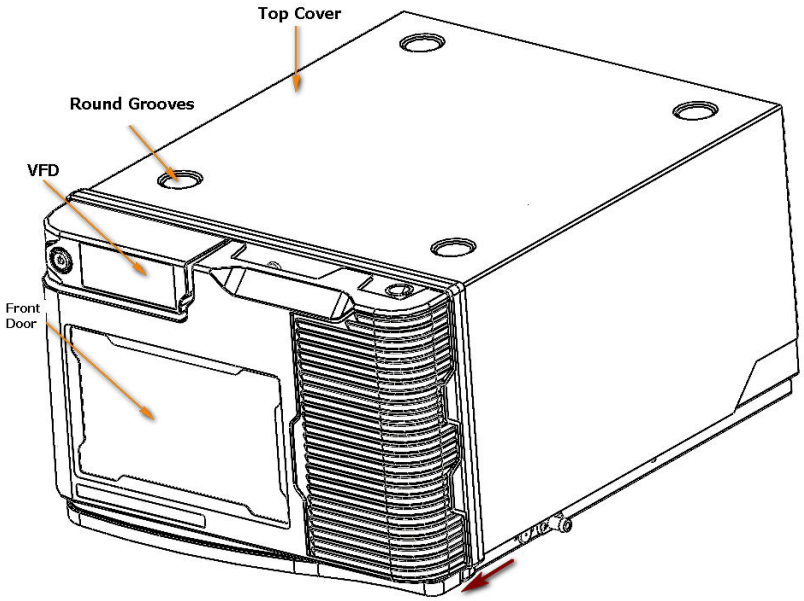


Figure 1-2 Vertical View of L-3320

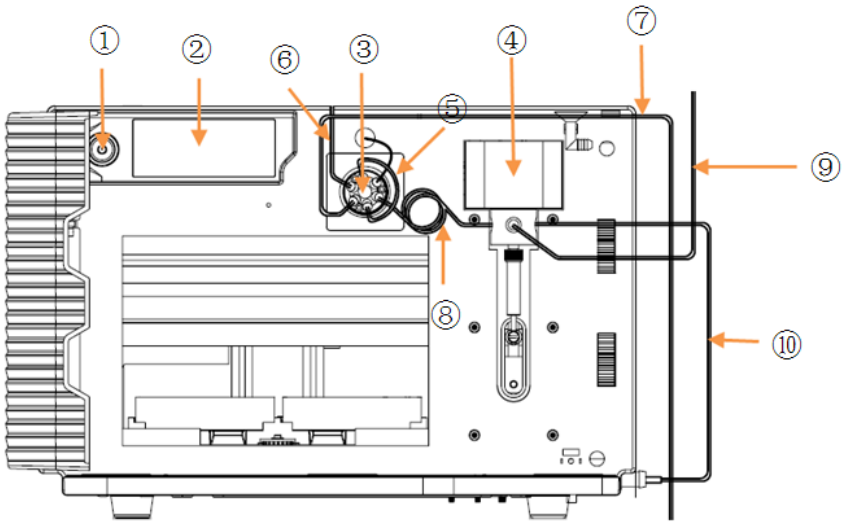


Figure 1-3 Front View of L-3320 (with front door open)

Table 1-1 Part List of L-3320 (with front door open)

No.	Description	No.	Description
①	Power Switch	⑥	Tubing for connecting the pump
②	VFD	⑦	Tubing for connecting the column
③	Injection Valve	⑧	Buffer Tubing
④	Syringe	⑨	Tubing for connecting the wash solvent
⑤	Loop	⑩	Waste Tubing

Upward View

As shown in Figure 1-4, there are four round rubber feet at the bottom of the instrument chassis which make the autosampler be easily fitted into the system to stabilize the whole system. Apart from these, at the bottom of the chassis, there are also vents (used to prevent the increased temperature from influencing the normal operation of the instrument), a leakage tubing and tray.

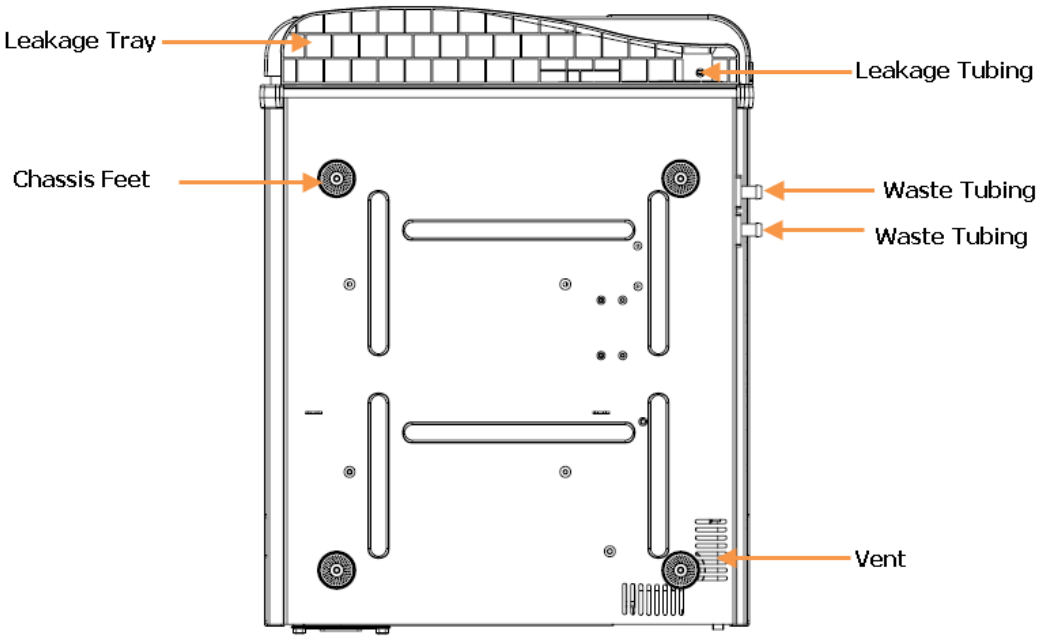


Figure 1-4 Upward View of L-3320

Rear Panel

As shown in Figure 1-5, the rear panel of the autosampler provides power socket (include fuse), fan (release the heat inside the chassis), interface board interface (option), main control panel interface and ground screw.

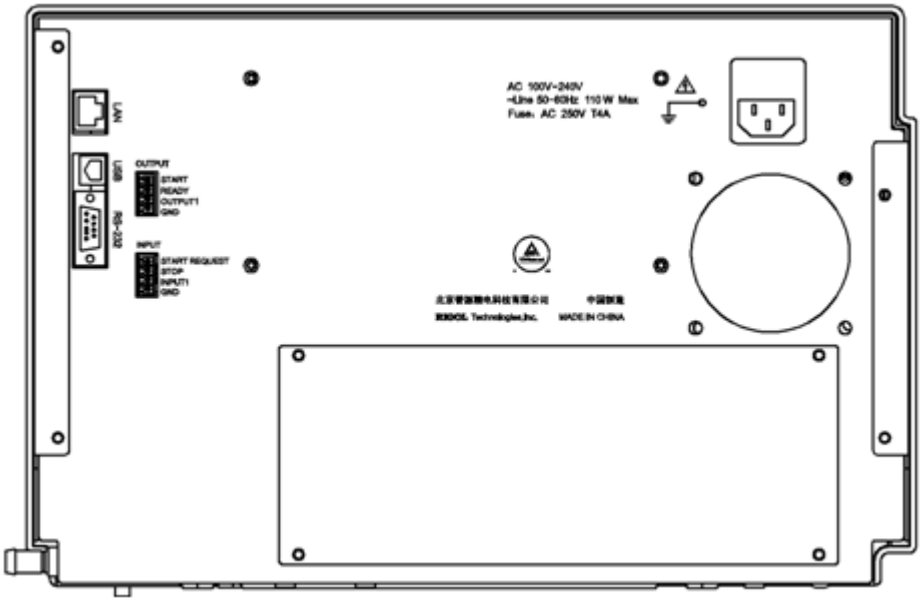


Figure 1-5 Rear Panel of L-3320

Modules

As shown in Figure 1-6, L-3320 mainly consists of seven parts.

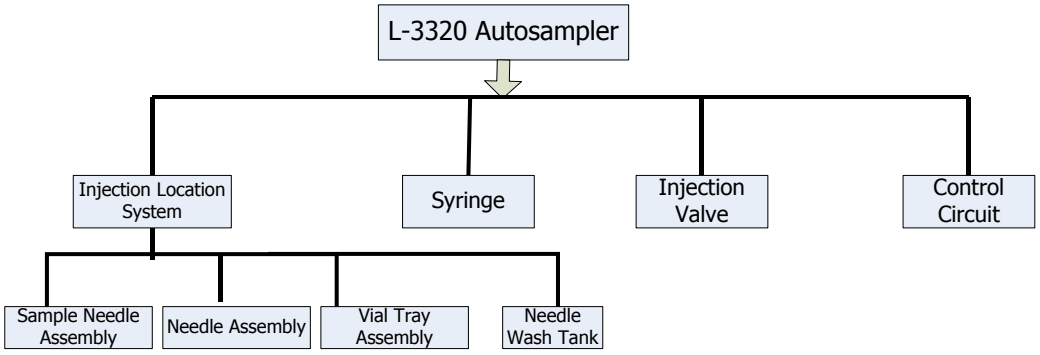


Figure 1-6 Parts of L-3320

1. Injection Location System

The injection location system mainly consisting of the sample tray assembly, needle assembly, wash position and sample needle assembly, is mainly used to locate the sample needle. Wherein, the sample tray assembly controls the Y-direction (front and back) movement of the sample; the needle assembly mainly controls the X-direction (left and right) movement as well as the Z-direction (up and down) movement of the sample needle; the wash position is mainly used to wash the sample needle (include washing the interior and exterior surfaces of the needle); the sample needle assembly is mainly used to pierce the septa and aspirate the sample.

2. Syringe

The syringe is used to aspirate the sample from a vial into the sample loop. The buffer tubing between the syringe and the injection valve prevents contamination of the syringe.

3. Injection Valve

In injection mode, disconnect the sample loop and the fluid system and when the injection is finished, connect the sample loop to the fluid system.

4. Control Circuit

The control circuit board controls the running of all the parts of the autosampler, including signal input, motor and motor position inspection and control, display and leakage inspection and control as well as system power control. Besides, the control circuit board provides various interfaces for connecting external devices as shown in Figure 1-7.

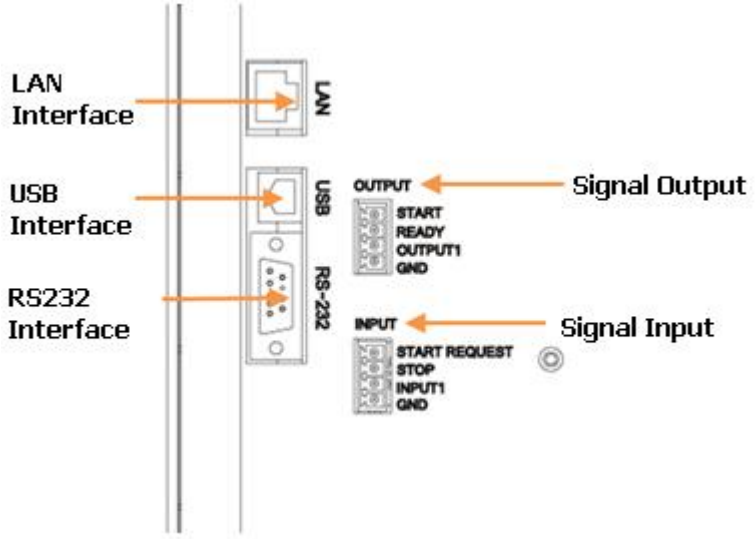


Figure 1-7 Interfaces on the Control Circuit Board

Chapter 2 To Install L-3320

Main topics of this chapter:

- Installation Preparations
  Working Environment and Place Requirement
  Characteristics
  Specifications
  Unpacking & General Inspection
- To Install the Instrument
  Preparations
  To Connect the Fluid System
  Waste Tubing
  To Connect other Devices
  Wash Solvent and Syringe Rinse
  Syringe
  Sample Handling
  To Connect to Power
  Power-on Self-test
- Instrument Optimization
  Parameter Optimization

Installation Preparations

You need to get a basic understanding of the working environment, place requirement, characteristics, specifications and the precautions before installing the autosampler to avoid hazard and ensure the safe, correct and efficient operation of the instrument.

Working Environment and Place Requirement

1. Temperature

Temperature fluctuation would affect the performance specifications of the autosampler. To acquire the optimum performance, place the instrument under constant environment temperature with less than 2℃/hour fluctuation and away from the vents of the air conditioner and other wind source.

2. Relative Humidity

The device should be placed under a relative humidity ranging from 20% to 80% in order to avoid being influenced by dusts or moistures as under high humidity, moisture may easily attach to the instrument surface and interior and corrode those metal parts or cause circuit damages.

3. Power Supply

The instrument accepts stabilized voltage supply: 100 VAC_{RMS} - 240 VAC_{RMS}, 50 Hz - 60 Hz.

WARNING

The instrument must be supplied with specified, stabilized source. Working in overvoltage may cause electric shock or instrument damages.

WARNING

It is necessary to remove the power cord from the AC inlet if you want to cut the power, otherwise the instrument will be still in power-on state, even if the power switch on the front panel of the instrument is turned off.

4. Power Cable

Please use the specific power cable offered by Rittun.

WARNING

The instrument is grounded through the grounding conductor of the power socket. To avoid electric shock, do not use a mains jack without grounding and make sure the instrument has been grounded properly before any connection.

5. Solvent Safety

Since most solvents are volatile and inflammable organic solvents, the instrument must be placed in a position with good ventilation (avoid convection of air) and without open flame. To guarantee the normal operation of the instrument and prolong its service life, the instrument should be operated away from inflammable, explosive and highly corrosive gases and be protected from electromagnetic interference etc).

6. Static Electricity

As the tubings of the HPLC system are usually rather thin, when the liquids most of which are inflammable and explosive organic solvents pass through the tubings at high flow rates, large amount of electrostatic charges would be generated. Thus, users must take precautions against static electricity during operation.

Safety Measures:

- Cover the cracks between solvent bottles and tubings using a cap or parafilm in order to prevent solvents from volatilization and keep the unit away from external static sparks.
- The operator should wear anti-static clothes and shoes.
- Maintain certain air humidity.

7. Workbench

The instrument needs to be placed on firm and level workbenches with more than 70 cm depth, more than 200 cm width, 10 cm clearance at the front and back as well as 5 cm clearance on each side of the body for easy operation.

Characteristics

Items	Specifications
Sample Capacity	2 standard 48-vial tray or 2 standard 96-well or 2 standard 384 -well
Loop Volume	Standard 100μL; 10μL, 20μL and 50μL optional
Syringe Volume	Standard 250 μL; 100 μL optional
Injection Mode	Full loop, partial loopfill and μL pickup mode
Reproducibility	Full loop injection: RSD≤ 0.3% Partial loopfill injection: RSD≤0.5% μL pickup injection: RSD≤1%
Cross Contamination	< 0.02% (after programmed needle wash)
Max Injection Volume	Full loop injection = loop volume Partial loopfill injection = 1/2 of loop volume μL pickup injection = (loop volume - 3 × needle volume)/2

Specifications

Items	Specifications
Weight	17 kg
Dimensions (L×W×H)	453 mm×359 mm×244 mm
Power Voltage	100 VAC_{RMS} to 240 VAC_{RMS}
Power Frequency	50 Hz to 60 Hz
Power	110 W_{MAX}
Operation Environment Temperature	15 ℃ to 35 ℃ (with less than 2 ℃/hour temperature variation)
Operation Relative Humidity	20% RH - 80% RH

Unpacking & General Inspection

When you get a new Rittun L-3320 autosampler, please inspect it following the steps below.

1. Inspect the instrument

Inspect the shipping container. In case of any damage, keep the damaged shipping container or cushioning material. After taking the instrument out of the container, check the appearance of the instrument. If it is damaged, do not connect the instrument. If the instrument is received in apparent good order and condition, ask Rittun specified installation engineer to install the instrument and test its performance.

If the instrument is damaged, or the instrument can not work normally, or the instrument does not pass the performance test, please contact the local distributor or the local office of Rittun.

The consigner or carrier shall be liable for the damage to instrument resulting from shipment. Rittun would not be responsible for free maintenance/rework or replacement of the unit.

CAUTION

The new instrument must be unpacked and connected only by Rittun assigned engineer. Any related works should not be done when installation engineer is absent.

Unpacking Steps:

- Clip the protective bandage around the package and open the package (please keep the package for future use).
- Take out all the accessories including the power cord, connecting cable, packing list, accessory box, User's Guide and warranty.
- Gingerly take out the unit along with the protective foam at the bottom and place it on a horizontal workbench.
- Clip the bandage on the moving part of the injection location system.

2. Check the accessories

Take out all the accessories after unpacking and carefully check the spare parts against the packing list. If your contents are incomplete or damaged, please contact the local distributor or the local office of **R**ittun.

To Install the Instrument

Installers can install the autosampler when the environment and installation place requirements are met and the instrument has passed the general inspection.

Preparations

The fluid connectors, installation tools and tubings (already been cut, you can also cut them by yourself) are provided in the accessory box of L-3320.

CAUTION

Watch your hands when you cut the tubing. You are recommended to use the tubings (already been cut) provided in the accessories.

CAUTION

Do not connect tubings or devices that would cause the back pressure of the flow cell to exceed 1000psi to avoid damaging the flow cell.

WARNNING

Make sure the instrument has been turned off and the power cable has been removed from the AC supply before you connect the fluid system.

To Connect the Fluid System

As shown in Figure 2-1, the fluid system connections of L-3320 consist of three parts: ① the connection between the outlet of the pump and the inlet of the autosampler; ② the connection between the outlet of the autosampler and the column; ③ the connections of the wash solvent tubing and the waste tubing of the syringe.

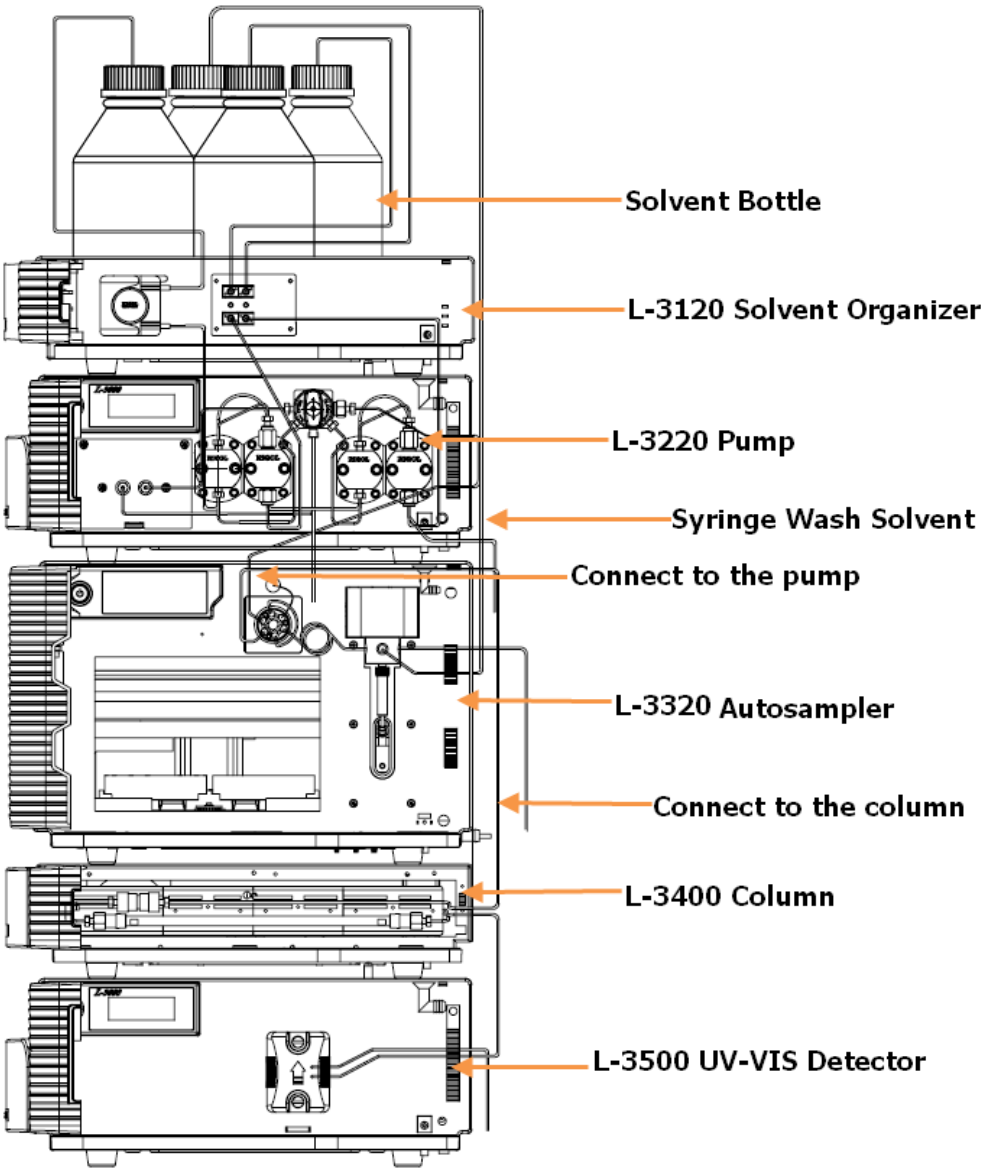


Figure 2-1 Fluid Connections of L-3000 HPLC System

① The connection between the outlet of the pump and the inlet of the autosampler:
- Open the front door of the autosampler to expose the injection valve (the exposed ports of the pump and injection valve are sealed with plugs when the instrument leaves factory to prevent dust from getting in).
- Take off the plugs and connect the outlet of the pump with the inlet of the injection valve (port 3) using the tubing (standard and provided randomly).

② The connection between the outlet of the autosampler and the column:
- Take off the plugs at the inlet of the column and the outlet of the injection valve.
- Connect the outlet of the injection valve (port 2) with the inlet of the column using the tubing (standard and provided randomly).

③ Take off the two plugs on the syringe and install the wash solvent tubing and the waste tubing to the corresponding positions (refer to Figure 1-3).

The fluid connections should be as shown in Figure 2-2 after the fluid is installed.

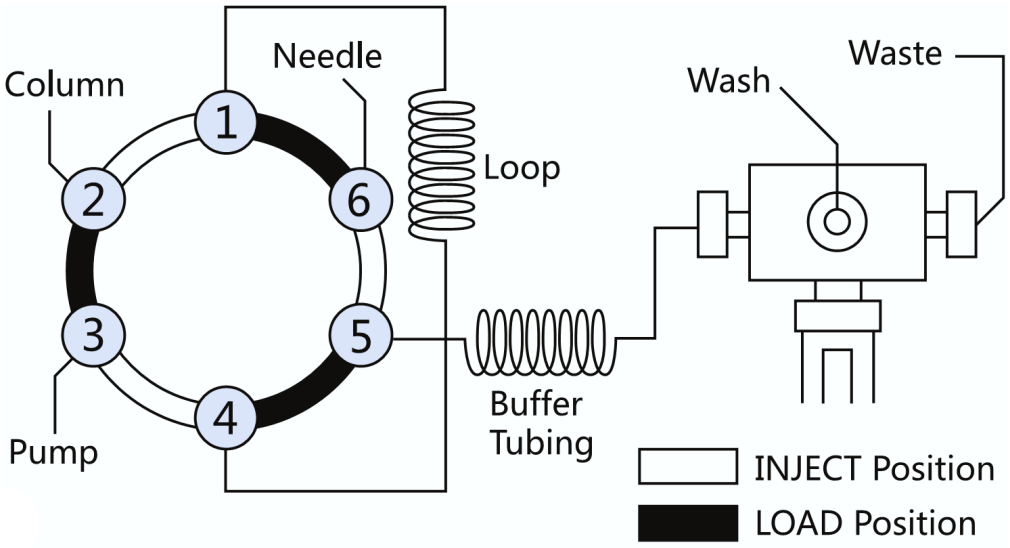


Figure 2-2 Fluid of L-3320

Table 2-1 L-3320 Standard Tubings

Name	Maretial/Dimensions
Standard sample needle and tubing (23.5 µL)	Dimensions: 82 mm × 0.8 mm OD × 0.25 mm ID ETFE (Tefzel): 400 mm × 1/16" OD ×0.25 mm ID
Buffer tubing from the injection valve to syringe valve (500 µL)	ETFE (Tefzel): 637 mm × 1/16" OD × 1.0 mm ID
Tubing from the wash solvent bottle to syringe valve	FEP: 600 mm × 1/8" OD ×1/16" ID
Tubing from the bulk head straight union to needle wash tank	Emulsion Hose: 200 mm × 11.2mm OD × 8.0 mm ID
Tubing from the pump to injection valve	Stainless Steel Pipe: 350 mm×1/16" OD×0.07" ID (L-3210, L-3220, L-3225, L-3240) 450 mm × 1/16" OD ×0.07" ID (L-3245)
Two-way tubing from the outlet of the injection valve to the preheat tube of the column	Stainless Steel Pipe: 300 mm × 1/16" OD ×0.007" ID

Note the following if you need to install new tubing:

- Insert tubing ends always flush with ferrule ends.
- Do not overtighten the nuts, as this may cause blockage in the flow path.
- Make sure that you always use tubing volumes that are suitable for use with the other items in the flow path.

Waste Tubing

Make the following connections for disposal of waste liquids.

- Connect the drain tubing (in the L-3320 tool package) to the right-hand drain hose connector (as shown in Figure 2-3).
- Place the other end of the drain tubing in the waste solvent bottle (on the floor).

All the wash solvent and the sample solvent that is not injected will be removed through this tubing.

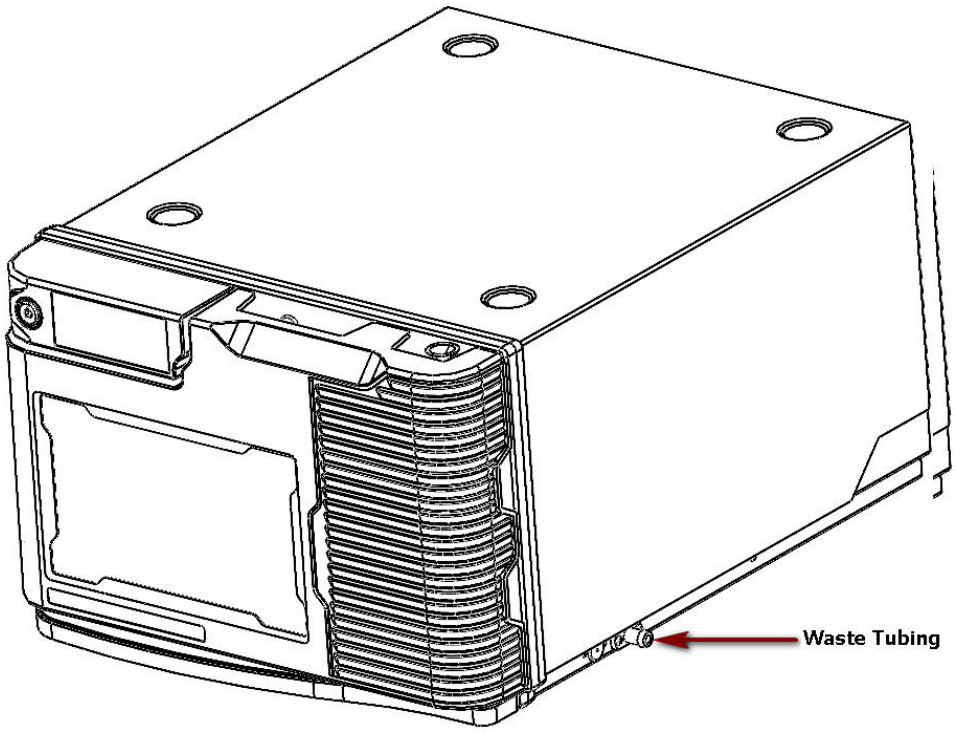


Figure 2-3 L-3320 Autosampler

To Connect other Devices

Besides the autosampler, a basic HPLC system also requires the column, pump, detector and data processing device (such as PC). You can also connect the column oven, solvent organizer (include vacuum degasser and peristaltic pump), hand-hold controller and fraction collector to improve the stability and reproducibility of the measurement and realize high degree of automation. The connecting methods of the devices required for a basic HPLC system are introduced below.

1. **Connect the column**
   The column which is used to separate the sample, is an important component of the HPLC system. The inlet of the column is connected to the autosampler and the outlet to the flow cell of the detector.

2. **Connect the pump**
   The pump which is used to transport the sample, is an important component of the HPLC system. The autosampler is connected to the pump via the column (for the detailed connecting method, refer to the Pump User's Guide).

3. **Connect the data processing device**
   L-3320 provides three kinds of basic interfaces (USB, RS-232 and LAN). You can use any of them to connect the data processing device (such as PC).

Wash Solvent and Syringe Rinse

Use a clean bottle for the wash solvent and place it onto the tray of the solvent organizer. The wash solvent is mainly used to wash the sample residues on the buffer tubing and the sample needle. You are recommended to use a mixture of distilled water and isopropanol (80 /20%) or mobile phase as wash solvent. Besides, you are recommended to change the wash solvent if the current one can not remove the residues ideally. Before using the wash solvent, degas the solvent with helium or an ultrasonic bath. Do not use salts or buffer solutions as crystals may block or damage the system.

To fill the wash solvent tubing, execute the following steps.

- Place the end of the wash solvent tubing in the filled wash solvent bottle;
- Open the device monitor interface of the CWS work station;
- Click **Initial Wash** in the L-3320 monitor area and the wash solvent is aspirated from the wash solvent bottle and the wash solvent tubing and the syringe are filled.

Syringe

A 250 µL syringe is standard installed in L-3320. However, it is also possible to install a 100 µL syringe.

Note: the L-3320 will give the best results if all air is removed from the syringe. Execute an extra initial wash to remove air from the syringe.

Sample Handling

Take the following into account when handling samples.

- The liquid in the standard vials should not exceed the vial neck to leave some space for the air needle to introduce in air during injection;
- Do not fill vials/wells to the edge. If you do, sample will be forced into the air needle, risking cross-contamination of samples and soiling the needles;
- It is important that seals and capmats are airtight to prevent air bubbles from forming and to block evaporation of volatile samples. The following seal types are recommended:
  Standard (low) well plate: sealing tape
  Deep Well plates: pierceable capmats (pre-slit or silicon) or sealing tape
  Vials: standard septa (thin types); do not use vials with hard caps that are not designed for being pierced by a sample needle
- Filtering the eluent with 0.2 µm filter will considerably reduce the risk of clogging. The same applies for the samples;
- Make sure you use the appropriate filter material for sample.

CAUTION

When connecting the fluid, make sure that the tubings, connectors and parts are firmly connected to avoid leakage.

To Connect to Power

- Turn off the power switch at the front panel of the autosampler.
- Connect one end of the power cord to the power socket at the rear panel of the instrument and connect the other end to the specified AC line.

Note: to avoid danger, please refer to "**Working Environment and Place Requirement**".

Power-on Self-test

After connecting the instrument, please perform power-on self-test of the autosampler following the steps below.

- Connect the autosampler and PC using the data cable and power on both.
- Start the autosampler and PC, start the CWS work station on the PC and set the "communication port" to build the communication between the instrument and PC.
- The autosampler performs self-test. "SELFCHECKING" is displayed on the VFD during the self-test. "ERROR CODE:######" will be displayed if error occurs during the self-test. The self-test lasts for about 1 min and the state message will be displayed in the instrument state interface of the work station. The return state is displayed through the PC.

Instrument Optimization

You can improve the detect performance of the instrument by optimizing the instrument parameters.

Parameter Optimization

Parameter optimization mainly consists of:

1. **Select the injection mode of the sample**
   To get the highest injection precision, full loop injection should be used. Partial loopfill injection mode can fulfill most of the requirements. If the sample is rather precious and rare, μL pickup injection mode can be used to save the sample.

2. **Set the needle wash**
   The default needle wash program is applicable to most of the applications. If the concentration of the sample injected is rather large, increase the number of needle washes properly.

Chapter 3 To Run L-3320

Main topics of this chapter:

- Precautions
- To Run the Autosampler
- Injection Mode and Principle
  Full Loop Injection
  Air Segment with Full Loop Injection
  Partial Loopfill Injection
  Air Segment with Partial Loopfill Injection
  µL Pickup Injection
  Air segment with µL Pickup injection

Precautions

Please read the precautions below before using L-3320 to ensure that the instrument can operate normally.

1. As the functions of the instrument are realized in combination with the CWS, make sure that the CWS is correctly install on the PC (for the installation and using method, refer to the CWS User's Guide) before running the work station.
2. Make sure that the fluid system of the instrument is correctly connected and check the tubings and connectors to avoid solvent leakage before turn on the instrument.
3. Do not open and close the front door repeatedly or touch and shake the fluid to avoid interfering the normal data information acquisition of the baseline.
4. After using the instrument, especially when buffer salt is used as the mobile phase or high-concentration sample is used, use pure water and organic phase to wash the tubings subsequently.
5. Shipping, storing or operating the injection valve below 0℃ with water in the fluid passages may cause failure of the sealing surfaces.
6. Due to the sealing material used by the motor of the standard injection valve, the mobile phase containing formic acid is not recommended for use.
7. Please filter or replace the needle wash liquid at a regular interval; as the sealing effect or service life of the distribution valve and injection valve will be affected when the needle wash liquid passing through the valves contains particular matter or has been contaminated.
8. Select the needle wash liquid according to the characteristics of the sample. Compare to the sample, the needle wash liquid should have equal or greater elution capacity; otherwise, there will be more sample residues.

To Run the Autosampler

Connect one end of the power cord to the AC line and the other end to the power socket at the rear panel of the instrument (at this point, the power switch should be in off state). Press the power switch at the upper left corner of the front panel, the VFD turns on and the welcome interface is displayed as shown in the figure below.

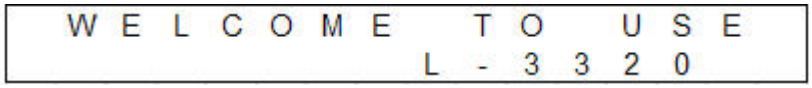


Figure 3-1 Welcome Interface

After about 3s, the instrument model, name and software version number will be displayed on the VFD as shown in the figure below.

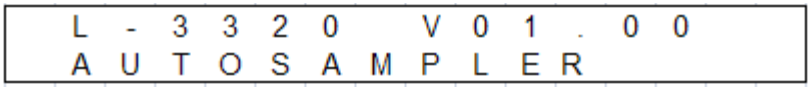


Figure 3-2 Instrument Information Display Interface

After about 3s, the instrument enters the self-test and the self-test item will be displayed on the VFD during the self-test as shown in the figure below.

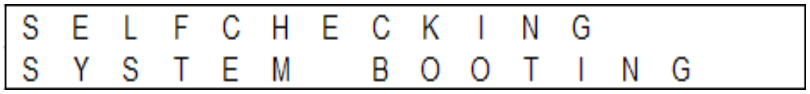


Figure 3-3 Instrument Self-test Interface

In this interface, the second line displays the self-test item (include the following 6 kinds). The VFD displays the running states of the self-test items in the table below one by one.

Table 3-1 Self-check Items

Self-check Item	Explanation
SYSTEM BOOTING	The self-check system starts
NEEDLE HOMING	The needle is initializing
TRAY HOMING	The sample tray is initializing
SEL. VAL. HOMING	The selection valve is initializing
SYRINGE HOMING	The syringe is initializing
INJ. VAL. HOMING	The injection valve is initializing

When self-check error occurs, the VFD will switch to the alarm display interface as shown in the figure below. For the alarm information, refer to "**Prompt Messages**".

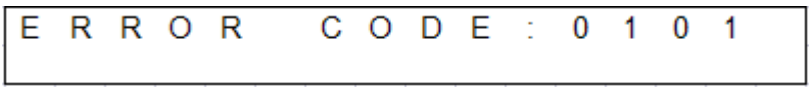


Figure 3-4 Alarm Interface

When the self-test passes, the VFD directly enters the main interface and switches between the main interface and secondary interface.

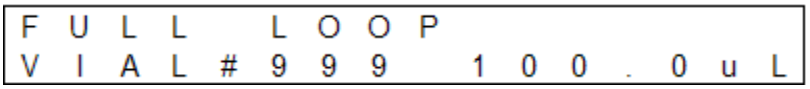


Figure 3-5 Main Interface

The first line of the main interface displays the injection mode (include 3 kinds as shown in the table below). The second line in the main interface displays the injection vial number and the injection volume.

Table 3-2 Injection Modes

Injection Mode	Explanation
FULL LOOP	Full Loop Injection
PARTIAL LOOPFILL	Partial Loopfill Injection
uL PICK-UP	μL Pickup Injection

The ready state of the secondary interface is as shown in the figure below.

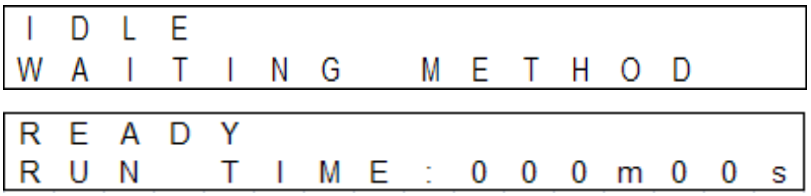


Figure 3-6 Secondary Interface

The first line in the secondary interface displays the main state (include 7 states as shown in the table below) of the system.

Table 3-3 Main States of the System

Main State	Explanation
IDLE	The system is ready
INITIAL WASHING	The system is performing initial wash
SAMPLE	The system is performing injection
WASHING	The system is performing wash
STOP	The system stops
DIRECT CONTROL	The system is controlling the movement directly
ERROR CODE:	The system is in error state

Injection is forbidden when the system is in error state.

The second line displays the secondary state (include 13 states) of the system as shown in the table below.

Table 3-4 Secondary States of the System

Secondary State	Explanation
NEEDLE MOVING	The needle is moving
FLUSH LOADING	The system is loading the flush liquid
CARRIER LOADING	The system is loading the transport liquid
AIR SEG. LOADING	The system is loading the air segment
SYRINGE WASHING	The system is washing the syringe
PRE RESET	The system is resetting
NEEDLE WASHING	The system is washing the needle
TRAY MOVING	The vial tray is moving to the front
TRAY HOMING	The vial tray is moving to the initial position
STOP…	The system is stopping
WARN:LEAK DETECT	Leakage warning
WARN:DOOR OPEN	Door open warning
WARN:DETECT FAIL	Vial missing warning

The functions of L-3320 that can be realized in combination with the CWS include:

- Full loop injection: maximum precision
- Partial loopfill injection: maximum flexibility
- µL Pickup Injection: zero sample loss

The maximum injection volumes are calculated with the following formulas.
Full loop injection = loop volume
Partial loop injection = 1/2 loop volume
µL Pickup Injection = (loop volume - 3 × needle volume)/2

Full loop gives maximum possible reproducibility ≤0.3%RSD. Partial loopfill gives maximum accuracy plus reproducibility ≤0.5%RSD; relatively smaller flush volumes can be programmed (the recommended minimum flush volume is 50µL), but will result in decreasing performance. µL Pick-up offers no sample loss, high accuracy, but slightly lower reproducibility (≤1%RSD).

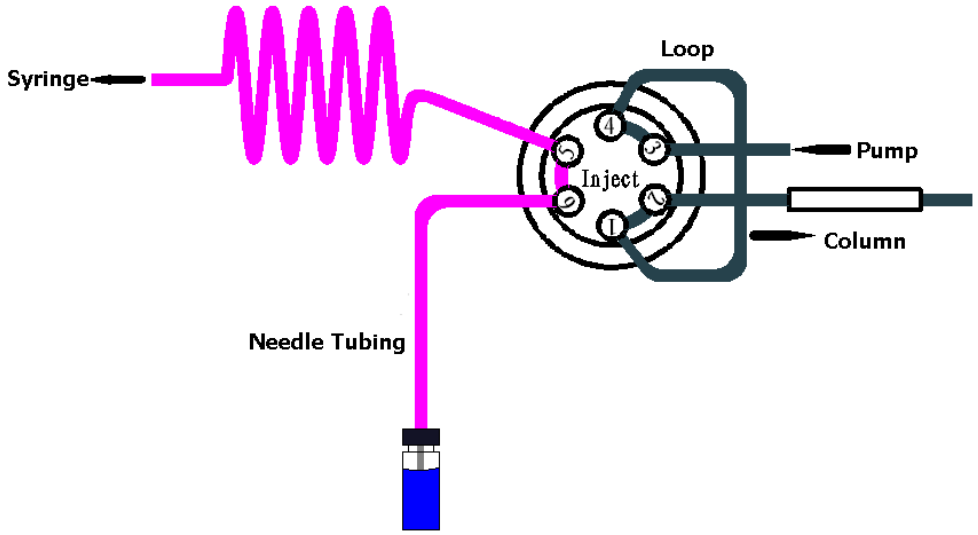
Injection Mode and Principle

Full Loop Injection

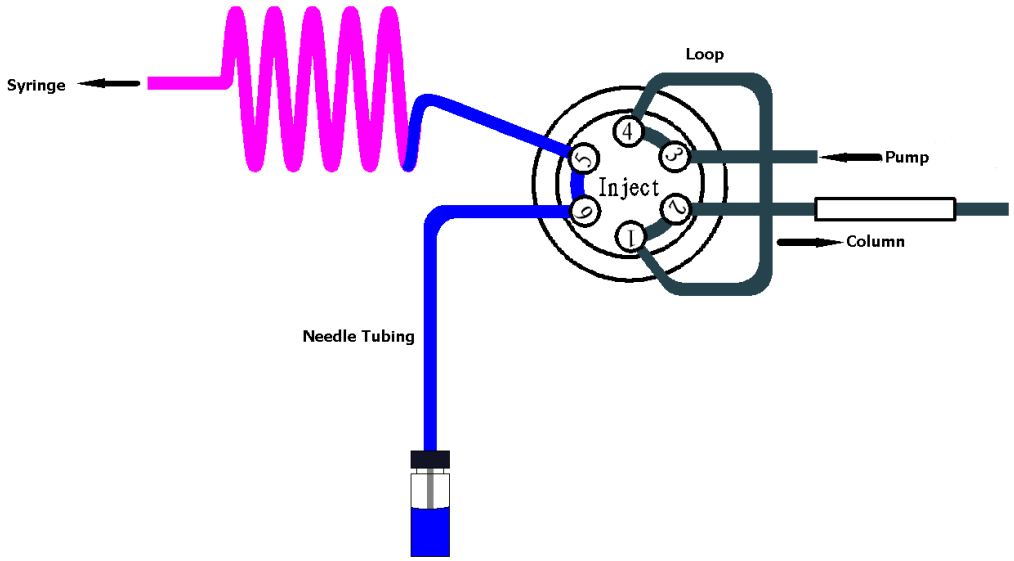
The sample loop is completely filled with sample. This type of injection results in extremely good reproducibility.

The steps of full loop injection are as follows.

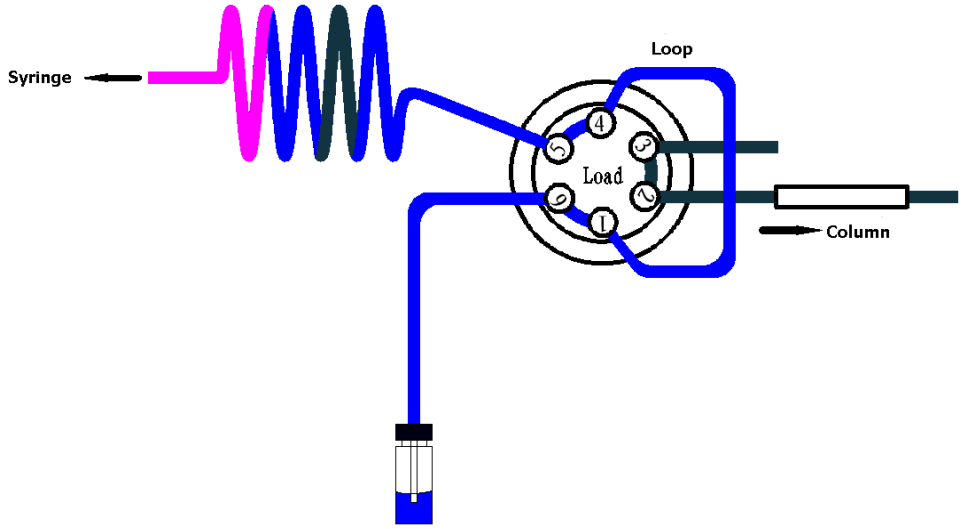
- The initial state: the injection valve is in Inject position. The sample needle with air needle has entered the vial or well.



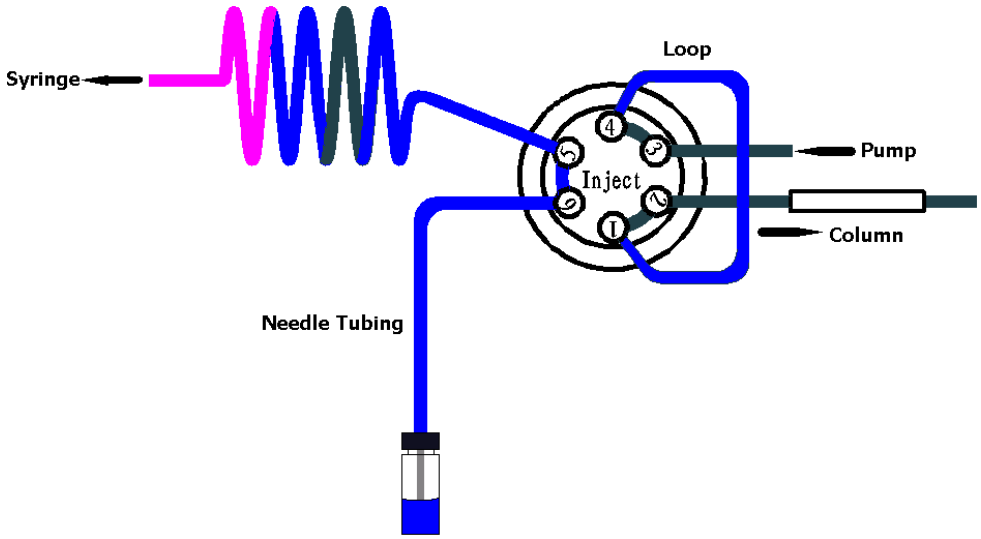
- The injection state: the injection valve is in Inject position. The sample needle with air needle has entered the vial or well. The syringe aspirates the "flush volume" from the sample vial/well to fill the needle tubing and part of the buffer tubing and remove the wash solvent.



- The injection valve is switched to Load. The sample loop is quantitatively filled by transporting a number of times the loop volume (2.5 times of the loop volume) of sample.



- The injection valve switches to the Inject position. The mobile phase flows through the loop and the sample is transported to the column. The analysis starts.

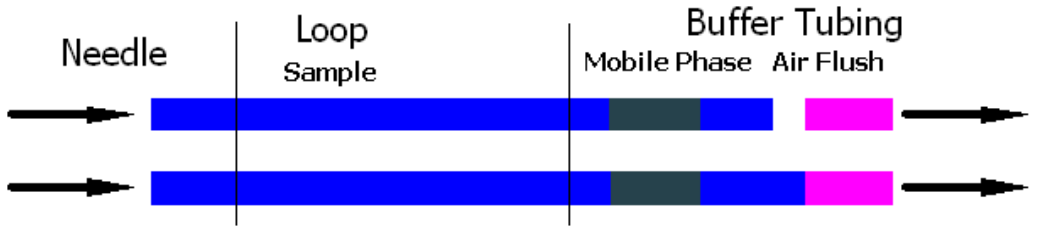


- A wash routine is performed after each injection.

Air Segment with Full Loop Injection

An air segment of 5 µL can be used to reduce the amount of flush volume. This air segment is at the front of the flush volume and will not be injected.

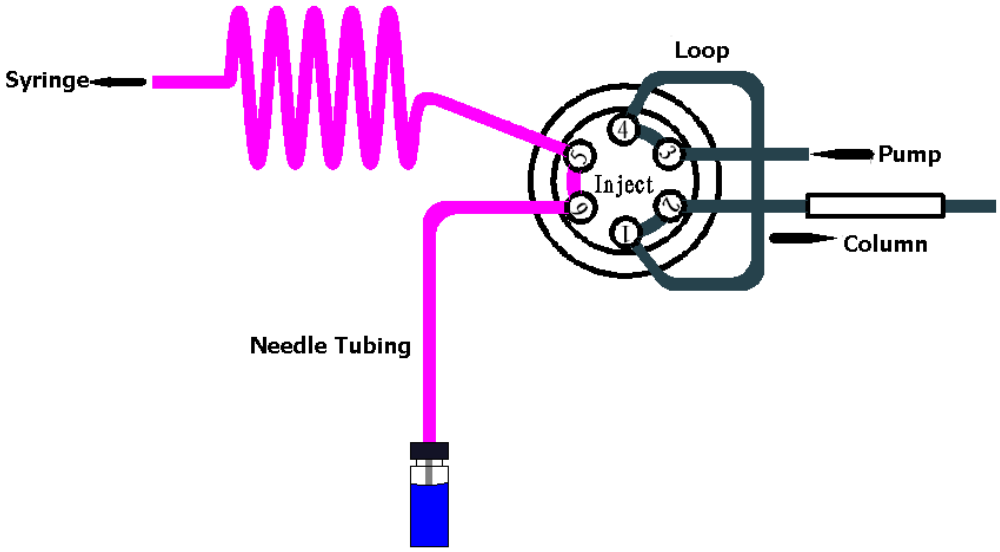
With a standard needle, the flush volume must be a minimum of 45 µL for injections with air segment and 50 µL for injections without air segment. If samples are highly viscous, it may be necessary to program larger flush volumes and reduce the syringe speed for better performance.



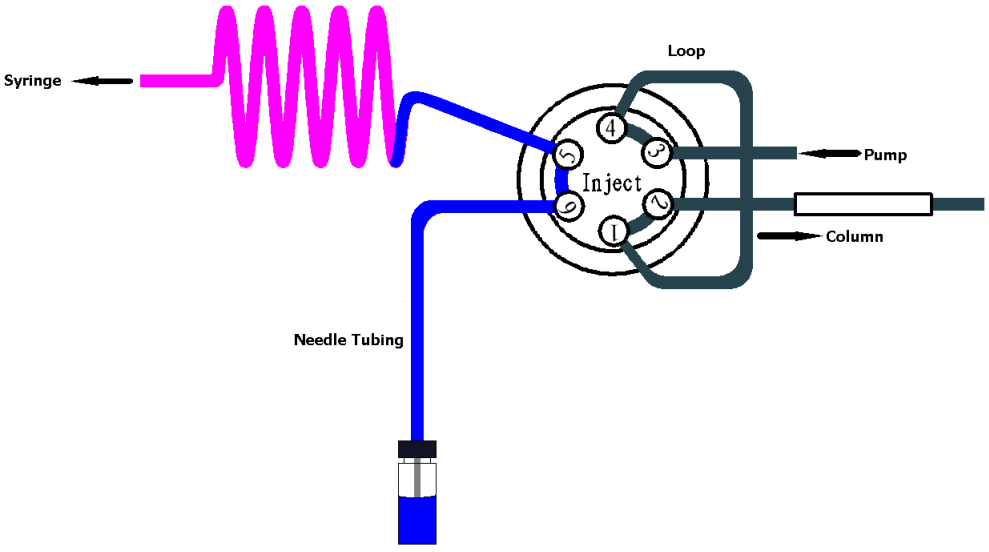
Partial Loopfill Injection

The steps of partial loopfill injection are as follows.

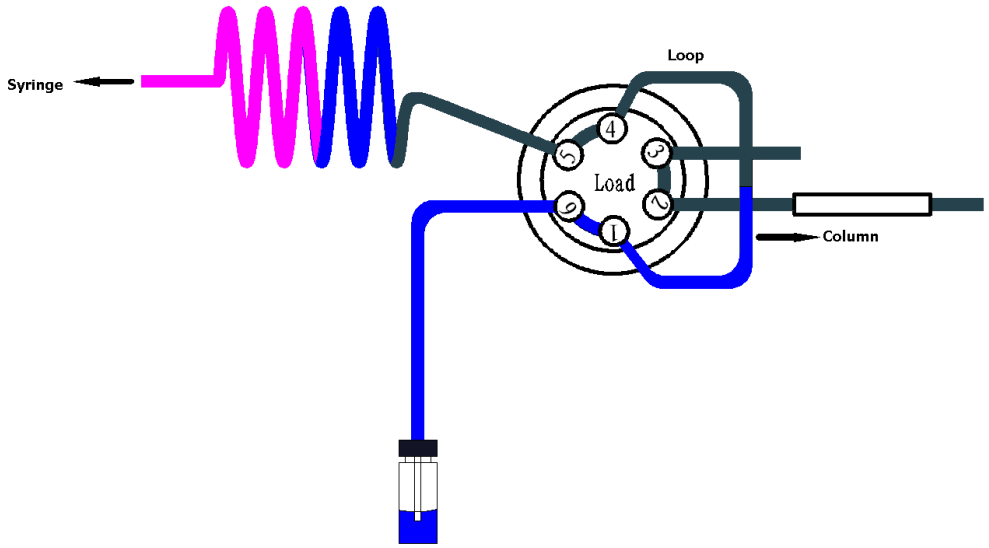
- The initial state: the injection valve is in Inject position. The sample needle with air needle has entered the vial or well.



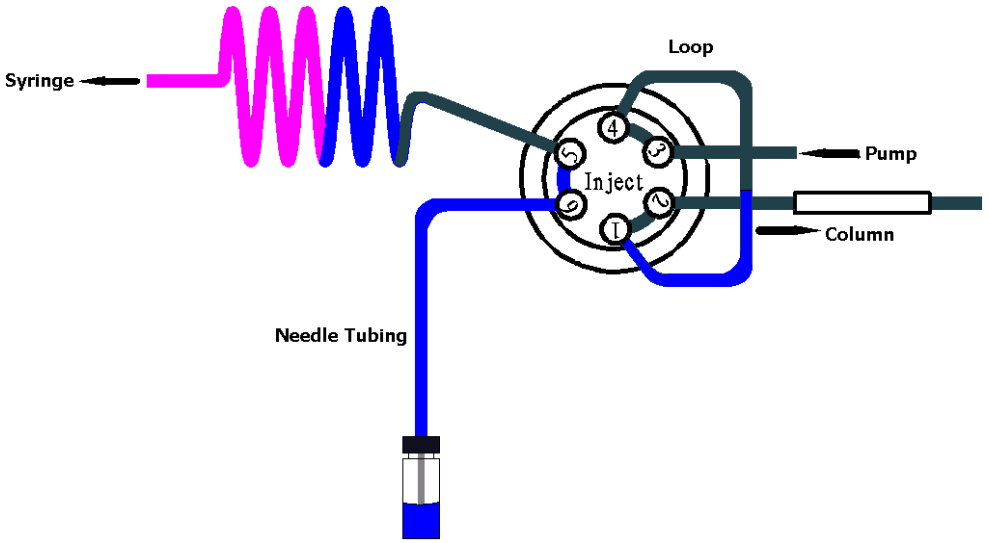
- The injection state: the injection valve is in Inject position. The syringe aspirates the "flush volume" from the sample vial to fill the needle tubing and part of the buffer tubing with sample and remove the wash solvent.



- The injection valve switches to Load and the programmed injection volume is aspirated into the sample loop (set through the software).



- The injection valve switches to Inject. The mobile phase flows through the loop and the sample is transported to the column. The analysis starts.

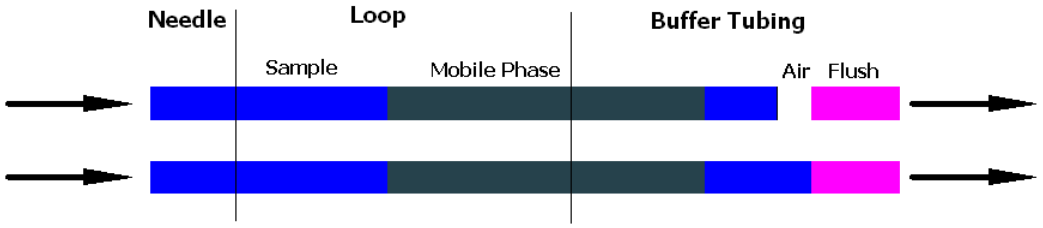


If an injection from the same vial and no wash routine is programmed, the next injection sequence will start with a flush of 50% of the programmed flush volume. Otherwise, it will start with a flush of the programmed flush volume. If the withdrawal of sample for the next injection exceeds the total volume of the sample buffer tubing, the buffer tubing is rinsed before the next injection. The next injection will start with the programmed flush (set via the software).

Air Segment with Partial Loopfill Injection

An air segment can be used to reduce the amount of flush volume. The air segment is at the front of the flush volume and will not be injected.

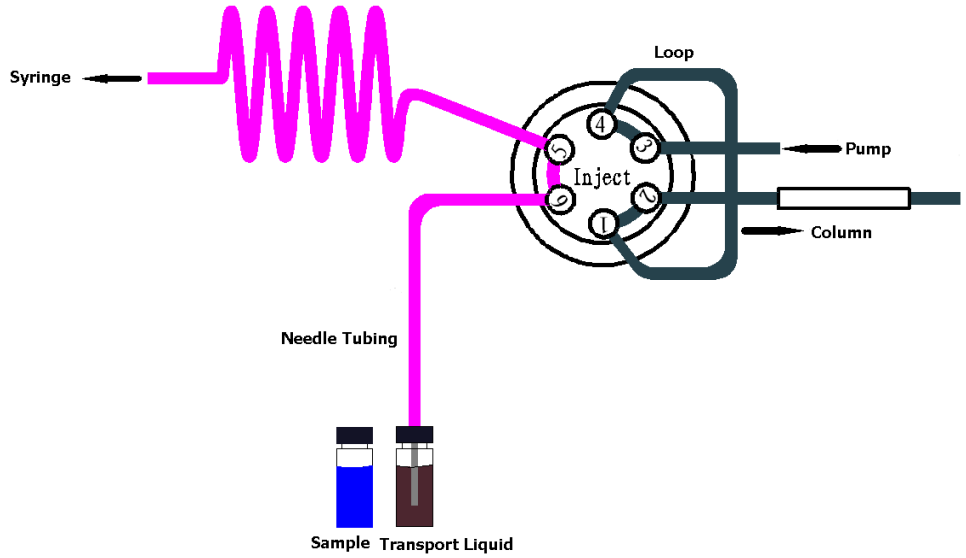
With a standard needle, the flush volume must be a minimum of 45 µL for injections with air segment and 50 µL for injections without air segment. If the samples are highly viscous, it may be necessary to program larger flush volumes and reduce the syringe speed for better performance.



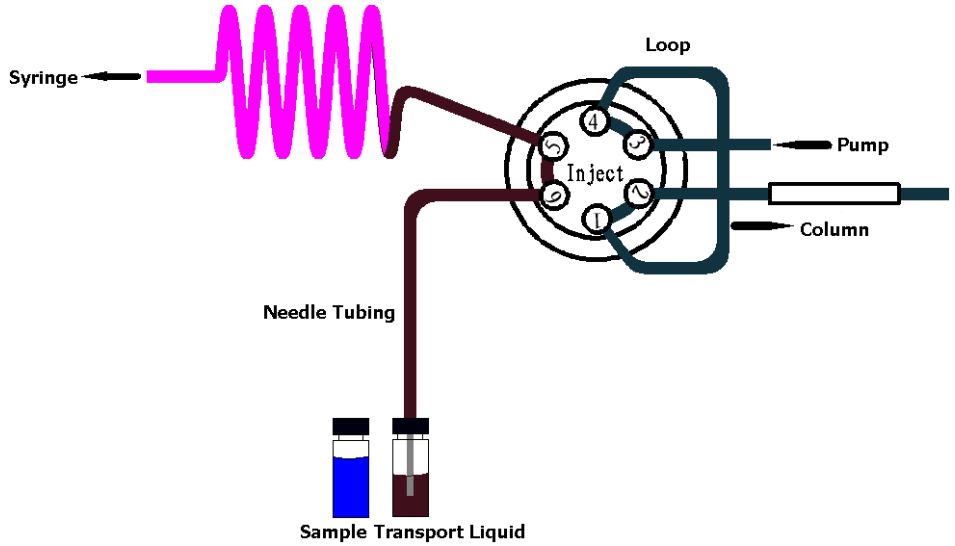
µL Pickup Injection

The steps of µL Pickup Injection are as follows.

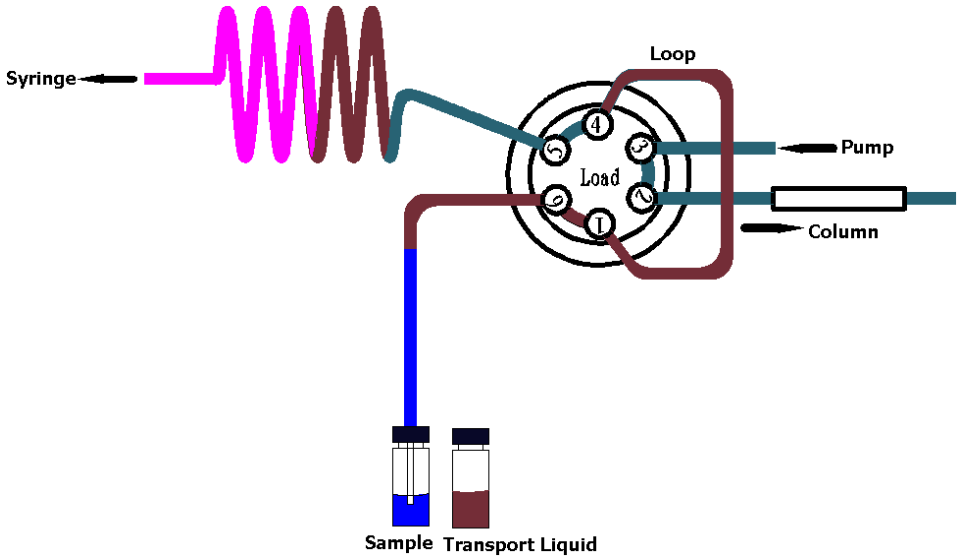
- The initial state: the injection valve is in Inject position. The sample needle with air needle has entered the transport position.



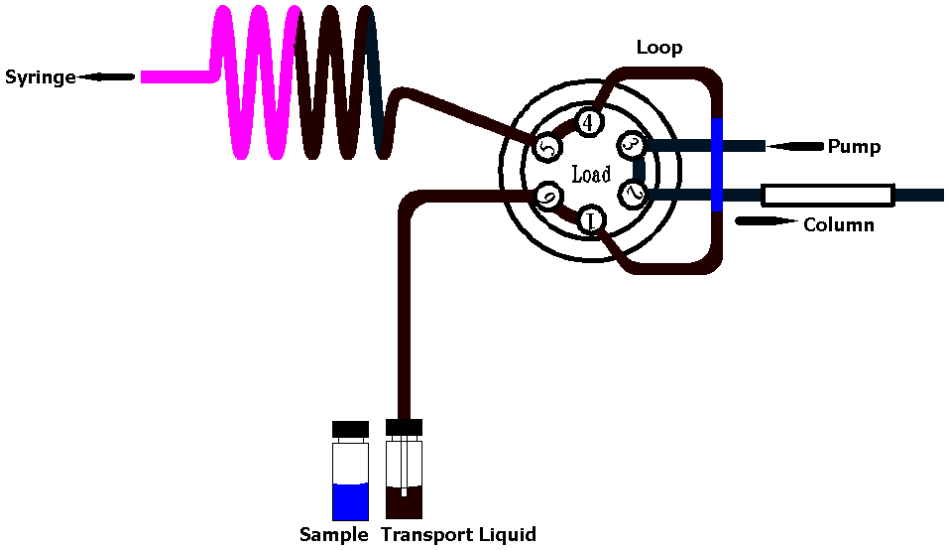
- For the first injection, the syringe aspirates a transport plug from the transport position to fill the needle tubing and part of the buffer tubing with transport liquid and remove the wash solvent.



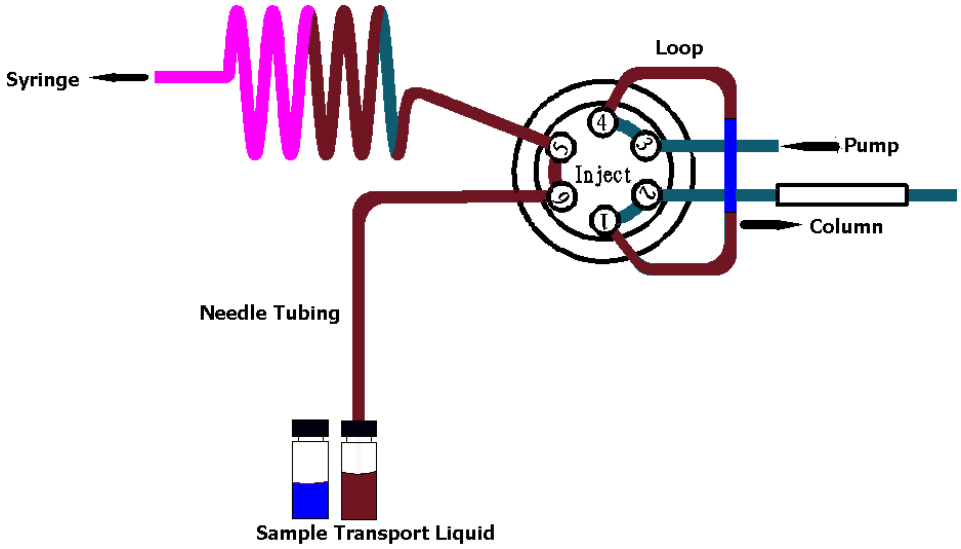
- The needle moves from the transport position to the sample vial. The injection valve switches to Load position. The programmed injection volume is aspirated from the sample vial.



- The sample needle moves back to the transport position. A second transport plug is aspirated. The sample is quantitatively transported into the loop.



- The injection valve switches to Inject. The sample loop is now part of the HPLC mobile phase flow path and the sample is transported to the column. The analysis timer starts.

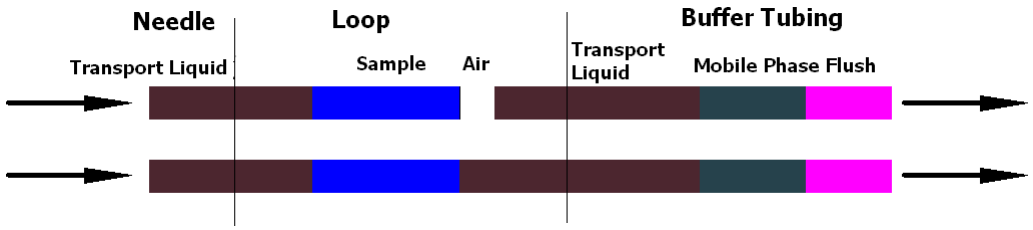


- The sequence is repeated for each injection.

Air segment with µL Pickup injection

If an air segment has been programmed, it appears at the front of the first plug of transport liquid and at the front of every sample plug.

In this injection mode, the air segment at the front of the sample plug is injected into the HPLC system.



Chapter 4 Troubleshooting

Main topics of this chapter:

- Troubleshooting
- Prompt Messages

Troubleshooting

The commonly encountered failures and their solutions are listed below to help users to analyze and solve the common problems. If the problem remains still or the problem is not included in the table below, please contact the local office of Rittun.

Table 4-1 Troubleshooting

Failure	Reason	Solution
The instrument does not start when power switch is pressed	Loose connection	Re-plug out/in the mains connection
	Faults in the power cable	Replace a same cable
	Substandard power supply	Use the specified power supply
	Fuse was burn out	Replace a same fuse
The **reproducibility** is not according to the specification	Air in the flow path	Perform one or more initial washes until the air bubbles are removed
	Leaking syringe	Make sure whether the syringe is correctly installed
	Dead volumes in tubing connections	Redo connections with new ferrules and nuts
The blank gives Peaks	Solubility problem	You can either modify your sample, or accept carry-over
	Bad match between sample characteristics and hardware	Check hardware: **Needle:** perform an extra wash (to wash the inside and outside needle) **Valve:** replace the injection valve **Tubing:** install different tubing (Steel, PEEK) between autosampler and column, or use different wash solvents
	The tubing is polluted	Use a new tubing

Table 4-1 Troubleshooting (continue)

Failure	Reason	Solution
No injection takes place	Blockage in flow path	Operate as follows: 1. Disconnect the needle from valve; 2. Perform a manual wash; 3. If solvent flows out from the injection port, check the needle; if no solvent flows from the injection port, disconnect the buffer tubing from valve; 4. Perform a manual wash; 5. If solvent flows out from the open end: check the rotor seal; if not: disconnect the buffer tubing from syringe valve; 6. Perform a manual wash; 7. If solvent flows out from the syringe valve: check the buffer tubing; if not, check for over-tightened connections in the entire flow path and check the syringe valve.
	Leakage in the injection valve	Operate as follows: 1. Disconnect the needle tubing and buffer tubing; 2. Connect the port 3 to a HPLC pump; 3. Block port 2; 4. Start the pump at a low flow; 5. Observe ports 5 and 6 for leakage; 6. If leakage occurs at ports 5 and 6, check the rotor seal; if not, recheck with manual valve.

Prompt Messages

L-3320 provides diagnosis function. During the operation, prompt messages will be displayed on the VFD if the instrument detects error. The prompt messages that might be displayed as well as their reasons and solutions are listed below.

1. **0101**
   Type: error
   Reason: fail to find the horizontal initial position of the needle.
   Solution: check the horizontal plate, optical coupler and the connection of the optical coupler of the needle. Make sure they are in normal condition and perform self-check again.

2. **0102**
   Type: error
   Reason: the horizontal movement of the needle times out.
   Solution: perform self-check again in the diagnosis and maintenance dialog box.

3. **0103**
   Type: error
   Reason: the horizontal motor of the needle is abnormal.
   Solution: perform self-check again in the diagnosis and maintenance dialog box.

4. **0104**
   Type: error
   Reason: the chip of the horizontal motor of the needle is abnormal.
   Solution: perform self-check again in the diagnosis and maintenance dialog box. If the error remains, please contact **R**ittun.

5. **0105**
   Type: error
   Reason: optical coupler signal is detected at non-optical coupler position in the horizontal direction of the needle.
   Solution: make sure that there is not obstacle in the moving direction and the optical coupler is correctly connected; perform self-check in the diagnosis and maintenance dialog box. If the error remains, please contact **R**ittun.

6. 0106

Type: error

Reason: do not detect optical coupler signal at the optical coupler position in the horizontal direction of the needle.

Solution: make sure that there is not obstacle in the moving direction and perform self-check in the diagnosis and maintenance dialog box. If the error remains, please contact **R**ittun.

7. 0107

Type: error

Reason: the distance error when returning to the optical coupler position in the horizontal direction of the needle is large.

Solution: make sure that there is not obstacle in the moving direction and perform self-check in the diagnosis and maintenance dialog box. If the error remains, please contact **R**ittun.

8. 0201

Type: error

Reason: fail to find the initial position of the vial tray.

Solution: check the plate, optical coupler and the optical coupler connection of the vial tray. Make sure they are in normal condition and perform self-check again.

9. 0202

Type: error

Reason: the vial tray movement times out.

Solution: perform self-check again in the diagnosis and maintenance dialog box.

10. 0203

Type: error

Reason: the motor of the vial tray is abnormal.

Solution: perform self-check again in the diagnosis and maintenance dialog box.

11. 0204

Type: error

Reason: the motor chip of the vial tray is abnormal.

Solution: perform self-check again in the diagnosis and maintenance dialog box.

If the error remains, please contact **R**ittun.

12. 0205

Type: error

Reason: optical coupler signal is detected at non-optical coupler position in the tray direction.

Solution: make sure that there is not obstacle in the moving direction and the optical coupler is correctly connected; perform self-check in the diagnosis and maintenance dialog box. If the error remains, please contact **R**ittun.

13. 0206

Type: error

Reason: do not detect optical coupler signal at the optical coupler position in the tray direction.

Solution: make sure that there is not obstacle in the moving direction and perform self-check in the diagnosis and maintenance dialog box. If the error remains, please contact **R**ittun.

14. 0207

Type: error

Reason: the distance error when returning to the optical coupler position in the tray direction is large.

Solution: make sure that there is not obstacle in the moving direction and perform self-check in the diagnosis and maintenance dialog box. If the error remains, please contact **R**ittun.

15. 0301

Type: error

Reason: fail to find the vertical initial position of the needle.

Solution: check the vertical plate, optical coupler and the optical coupler connection of the needle. Make sure they are in normal condition and perform self-check again.

16. 0302

Type: error

Reason: the vertical movement of the needle times out.

Solution: perform self-check again in the diagnosis and maintenance dialog box.

17. 0303

Type: error

Reason: the vertical motor of the needle is abnormal.

Solution: perform self-check in the diagnosis and maintenance dialog box.

18. 0304

Type: error

Reason: the chip of the vertical motor of the needle is abnormal.

Solution: perform self-check again in the diagnosis and maintenance dialog box. If the error remains, please contact **R**ittun.

19. 0305

Type: error

Reason: optical coupler signal is detected at non-optical coupler position in the vertical direction of the needle.

Solution: make sure that there is not obstacle in the moving direction and the optical coupler is correctly connected; perform self-check in the diagnosis and maintenance dialog box. If the error remains, please contact **R**ittun.

20. 0306

Type: error

Reason: do not detect optical coupler signal at the optical coupler position in the vertical direction of the needle.

Solution: make sure that there is not obstacle in the moving direction and perform self-check in the diagnosis and maintenance dialog box. If the error remains, please contact **R**ittun.

21. 0307

Type: error

Reason: the distance error when returning to the optical coupler position in the vertical direction of the needle is large.

Solution: make sure that there is not obstacle in the moving direction and perform self-check in the diagnosis and maintenance dialog box. If the error remains, please contact **R**ittun.

22. 0401

Type: error

Reason: fail to find the initial position of the distribution valve.
Solution: check the connection of the optical coupler of the distribution valve. Make sure it is in normal condition and perform self-check again.

23. 0402

Type: error
Reason: the distribution valve movement times out.
Solution: perform self-check again in the diagnosis and maintenance dialog box.

24. 0403

Type: error
Reason: the motor of the distribution valve is abnormal.
Solution: perform self-check in the diagnosis and maintenance dialog box.

25. 0404

Type: error
Reason: the motor chip of the distribution valve is abnormal.
Solution: perform self-check again in the diagnosis and maintenance dialog box. If the error remains, please contact **R**ittun.

26. 0501

Type: error
Reason: fail to find the initial position of the syringe.
Solution: check the vertical plate, optical coupler and the connection of the optical coupler of the syringe. Make sure they are in normal condition and perform self-check again.

27. 0502

Type: error
Reason: the syringe movement times out.
Solution: perform self-check again in the diagnosis and maintenance dialog box.

28. 0503

Type: error
Reason: the syringe motor is abnormal.
Solution: perform self-check again in the diagnosis and maintenance dialog box.

29. 0504

Type: error

Reason: the motor chip of the syringe is abnormal.

Solution: perform self-check again in the diagnosis and maintenance dialog box. If the error remains, please contact **R**ittun.

30. 0505

Type: error

Reason: optical coupler signal is detected at non-optical coupler position in the moving direction of the syringe.

Solution: make sure that there is not obstacle in the moving direction and the optical coupler is correctly connected; perform self-check in the diagnosis and maintenance dialog box. If the error remains, please contact **R**ittun.

31. 0506

Type: error

Reason: do not detect optical coupler signal at the optical coupler position in the moving direction of the syringe.

Solution: make sure that there is not obstacle in the moving direction and perform self-check in the diagnosis and maintenance dialog box. If the error remains, please contact **R**ittun.

32. 0507

Type: error

Reason: the distance error when returning to the optical coupler position in the moving direction of the syringe is large.

Solution: make sure that there is not obstacle in the moving direction and perform self-check in the diagnosis and maintenance dialog box. If the error remains, please contact **R**ittun.

33. 0601

Type: error

Reason: fail to find the initial position of the injection valve.

Solution: perform self-check again in the diagnosis and maintenance dialog box. If the error remains, please contact **R**ittun.

34. 0602

Type: error

Reason: the injection valve movement times out.

Solution: perform self-check again in the diagnosis and maintenance dialog box.

35. 0603

Type: error

Reason: the injection valve is abnormal.

Solution: perform self-check again in the diagnosis and maintenance.

36. 0604

Type: error

Reason: the motor chip of the injection valve is abnormal.

Solution: perform self-check again in the diagnosis and maintenance dialog box. If the error remains, please contact **R**ittun.

37. 0701

Type: error

Reason: the RAM check fails.

Solution: please contact **R**ittun.

38. 0702

Type: error

Reason: fail to write the RAM.

Solution: please contact **R**ittun.

39. 0703

Type: error

Reason: fail to read the RAM.

Solution: please contact **R**ittun.

40. 0704

Type: error

Reason: communication error of logic chip 1.

Solution: restart the instrument. If the error remains, please contact **R**ittun.

41. 0705

Type: error

Reason: communication error of logic chip 2.

Solution: restart the instrument. If the error remains, please contact **R**ittun.

42. 0706

Type: error

Reason: communication error of logic chip 3.

Solution: restart the instrument. If the error remains, please contact **R**ittun.

43. 0801

Type: alarm

Reason: invalid vial number.

Solution: check whether the vial tray specification is correct.

44. 0802

Type: error

Reason: invalid tray.

Solution: check whether the vial tray specification is correct.

Chapter 5 Daily Maintenance

Main topics of this chapter:

- Daily Maintenance
- Replaceable Parts
  To Replace the Injection Valve
  To Replace the Loop
  To Replace the Sample Needle Assembly
  To Replace the Air Needle
  To Replace the Syringe

Daily Maintenance

To ensure the optimum measurement effect of L-3320, perform regular check and maintenance of the instrument.

1. Check whether the tubings are bended and replace the bended tubings timely.
2. Check whether there are air bubbles and leakage in the tubings (include the buffer tubing, sample needle tubing and the syringe) and take timely solutions.
3. The wash solvent should be the same with the sample used to reduce the possibility of air bubble generation.

Safety Notices

Before any maintenance works, carefully read the safety instructions provided by your solvent supplier and prepare for precautions (such as wear a protect clothing, glasses or gloves) in order to avoid a significant risk of causing major injuries or illness because of solvent (especially poisonous and hazardous solvents) leakage when you try to open the unit tubing or connection.

Replaceable Parts

To Replace the Injection Valve

L-3320 is configured with an injection valve at the front panel for easier tubing connection and replacement. The replacement procedures are as follows (as shown in Figure 5-1).

1. Open the upper cover of the chassis and take the injection vale and bracket off from the front panel of the chassis;
2. Remove the 2 screws on the bracket and take off the injection valve to be replaced;
3. Fix a new injection valve onto the bracket using the screws;
4. Install the injection valve and the bracket onto the front panel of the chassis.

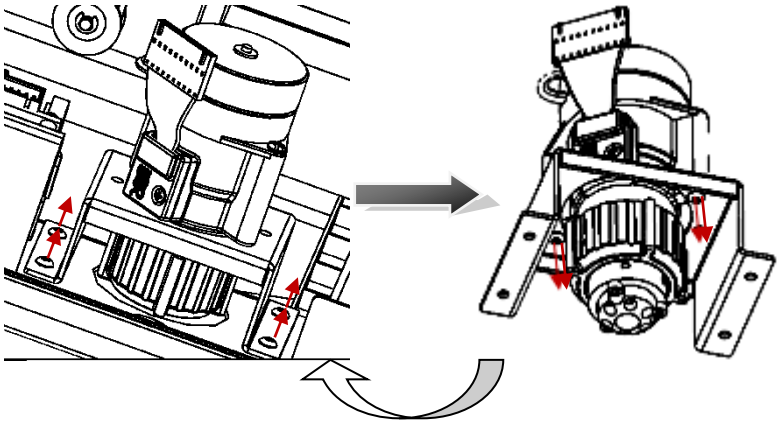


Figure 5-1 To Replace the Injection Valve of L-3320

Note: port 1 of the injection valve should point upward.

To Replace the Loop

L-3320 is configured with a 50 μL loop. A different sample loop size can be installed, but note that you will need the proper combination of syringe and tubing.

Take the following into account when you have installed a sample loop.

1. Connect the loop between port 1 and port 4 of the injection valve;
2. If loop with a different volume is installed, please enter the configuration setting interface to select the corresponding loop volume.

To Replace the Sample Needle Assembly

Execute the following steps to replace the sample needle (as shown in Figure 5-2).

1. Loosen the needle connection nut (number 3);
2. Loosen the nut (number 5) that connects the tubing (number 4) to port 6 of the injection valve;
3. Remove the sample needle by pulling it up from under the air needle;
4. Install a new needle and make sure the tubing end is connected to the sample needle end;
5. Fix the sample needle using nut 3;
6. Connect the other end of the needle connection tubing to port 6 of the injection valve using nut 5 and ferrule 6. Do not tighten too much as this may block the tubing;
7. Then, unscrew the connecting nut 3 and take out the tubing to check whether the front of the tubing is flush with the ferrule (as shown in Figure 5-3).

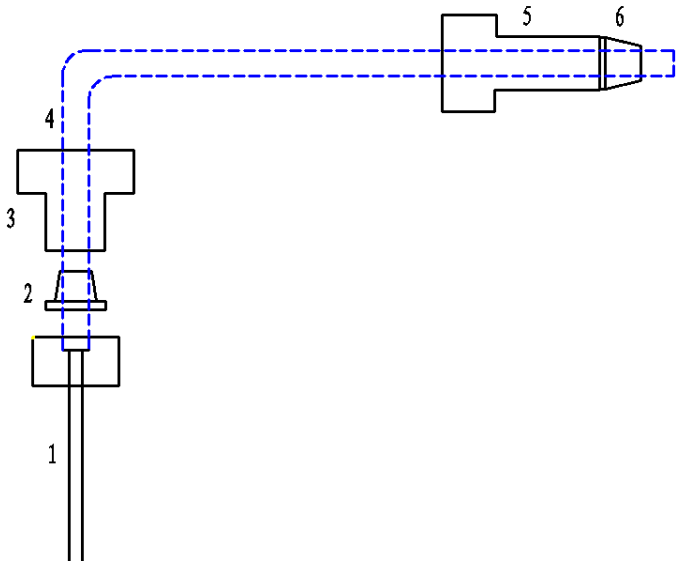


Figure 5-2 To Replace the Sample Needle Assembly of L-3320

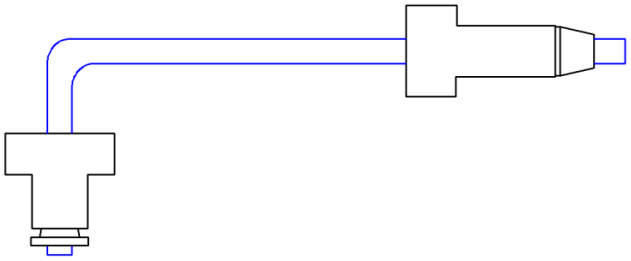


Figure 5-3 Correct Position Relation between the Sample Needle and Connectors

To Replace the Air Needle

Execute the following steps to replace the air needle (as shown in Figure 5-4).

1. Remove the sample needle (refer to "**To Replace the Sample Needle Assembly**");
2. Unscrew the locking nut to remove the air needle;
3. Install a new air needle;
4. Install the sample needle.

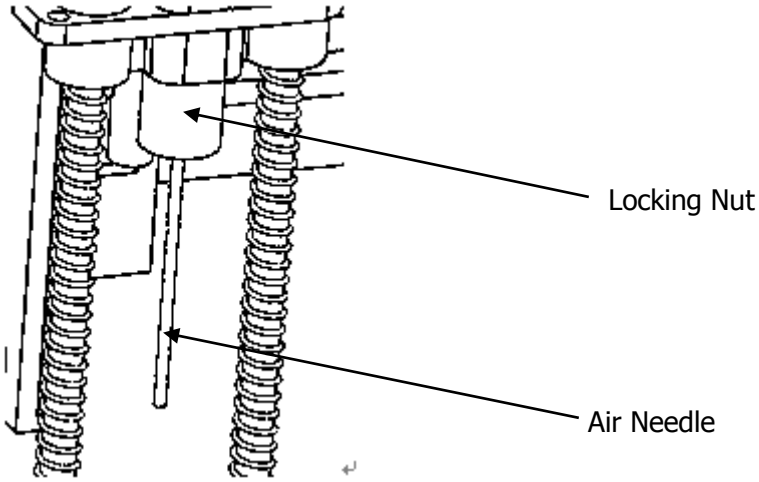


Figure 5-4 To Replace the Air Needle of L-3320

To Replace the Syringe

The schematic diagram is as shown in the figure below.

1. Selection valve;
2. Syringe glass tubing;
3. Syringe plunger;
4. Stationary shaft of the push rod.

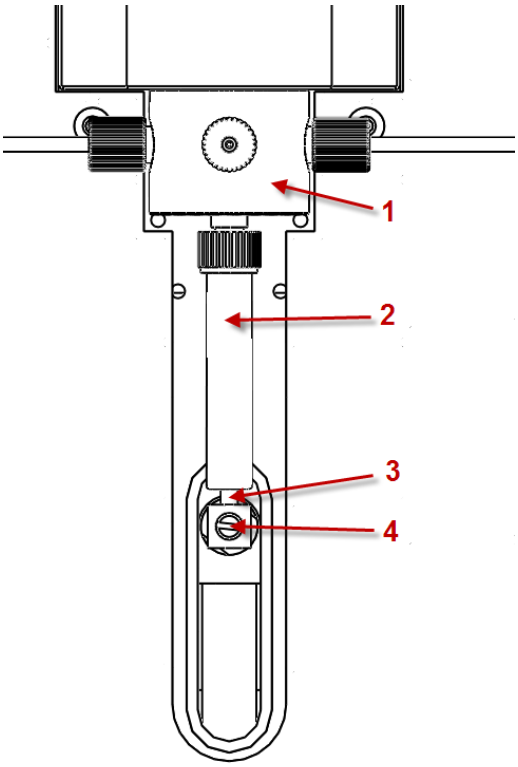


Figure 5-5 Syringe of L-3320

Execute the following steps to replace the syringe (as shown in Figure 5-6).

1. Click Change in the work station. Switch the distribution valve to the waste port and the syringe position to the initial position;
2. Remove the stationary shaft of the push rod using the straight screw driver;

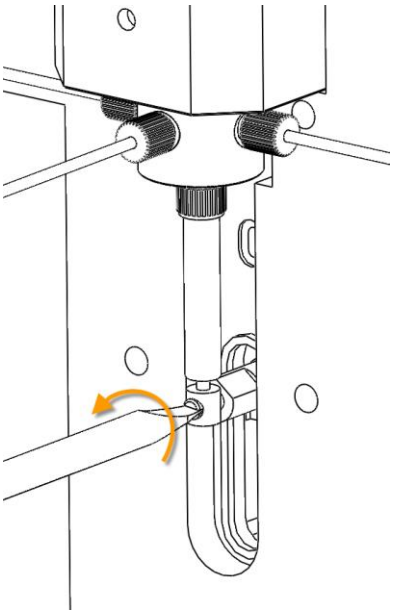


Figure 5-6 To Replace the Syringe of L-3320 (1)

3. Remove the connecting thread connecting the top of the syringe and the valve (as shown in Figure 5-7);

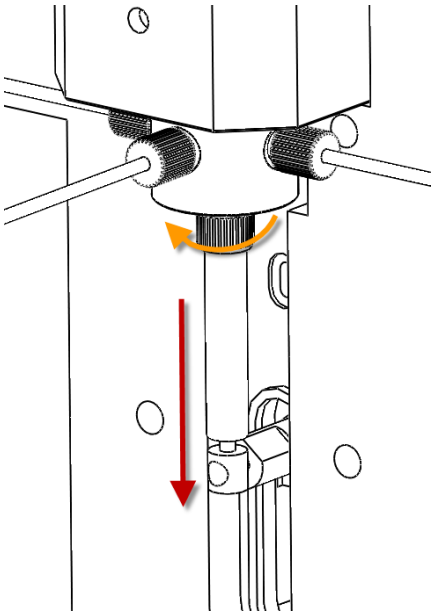


Figure 5-7 To Replace the Syringe of L-3320 (2)

4. Put the plunger seal part of the new syringe into the beaker filled with wash solvent or the mixture of isopropanol and water.

5. Insert the wet syringe plunger into the glass tubing of the syringe gently (as shown in Figure 5-8). Put the syringe into the beaker with the inlet facing downward and push the plunger up and down repeatedly to remove the air bubbles in the syringe inlet. If necessary, aspirate and drain solvent until the air bubbles in the syringe are removed and then fill the syringe with solvent.

Figure 5-8 To Replace the Syringe of L-3320 (3)

6. Install the new syringe onto the valve (as shown in Figure 5-9). You can push the plunger of the syringe gently to extrude some drops of solvent to make sure that there is liquid film at the inlet of the syringe to prevent air bubbles from entering the syringe when installing. Screw the syringe into the connecting thread hole on the valve and tighten it.

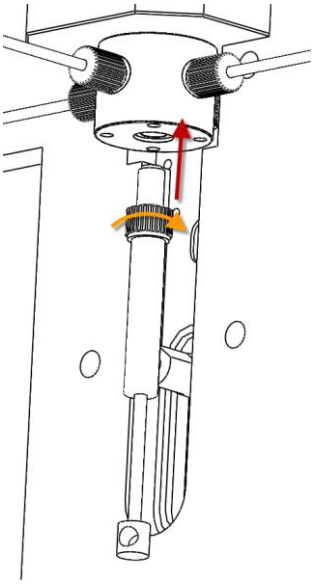


Figure 5-9 To Replace the Syringe of L-3320 (4)

7. Push the plunger of the syringe up to align the installation hole on the plunger with the connecting thread hole on the push rod of the syringe and the liquid in the syringe will be drained through the waste port. Then, screw the stationary shaft of the push rod into the connecting thread hole via the installation hole on the plunger of the syringe and tighten it using the screw driver (as shown in Figure 5-10).

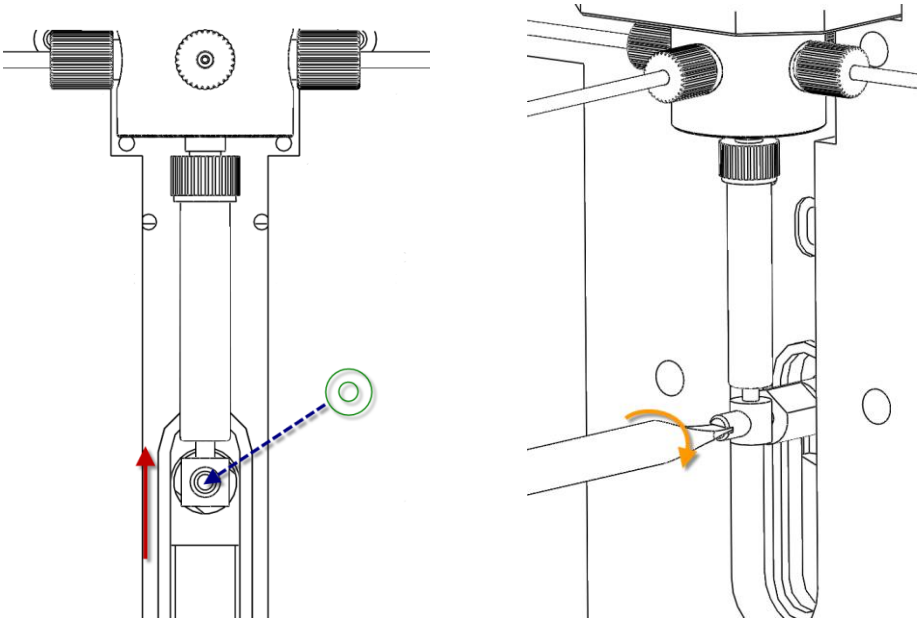


Figure 5-10 To Replace the Syringe of L-3320 (5)

8. Perform initial wash to remove the air bubbles in the syringe.
9. Check whether there are still air bubbles in the syringe. If yes, perform initial wash again until all the air bubbles are removed.

Chapter 6 Hardware Qualification

Topics of this chapter:

- Hardware Qualification Overview
  Hardware Qualification Implementation
  Hardware Qualification Precautions
  Hardware Qualification Requirements
- Hardware Qualification
  Autosampler Qualification
  System Qualification
- If the Qualification Fails

Hardware Qualification Overview

Hardware qualification has two types: component qualification and system qualification. The component qualification aims to individual components of the system and the system qualification is used to check whether the whole system works normally. To ensure the normal operation of the instrument system and the reliability of analytical data, you should regularly inspect every LC component and the whole LC system - execute regular hardware qualification from date of system installation until the end of service life and keep the records - as some vulnerable parts of the instrument may reduce the performance of the LC system after an appropriate interval of use. The hardware qualification is relevant with analysis, which needs to execute analytical methods validation and system suitability tests. Hardware qualification is the precondition for validation and test.

Hardware Qualification Implementation

1. **Daily Inspection**
   The daily inspection of the components and the HPLC system mainly focus on the maintenance of components in order to ensure the reliability of data analysis. Some inspections such as column loss and mobile phase adjustment should be performed during the system suitability test.

2. **Regular Qualification**
   Component and system qualifications should be performed at the time of the instrument installation and every 6-12 months after the installation (because the performance of the LC system will decline as time moves on).

3. **Maintenance Qualification**
   You need to re-certificate the component performance after system maintenance every time. The qualification type depends on the practical situation. If the module can't be qualified alone after maintenance, system qualification is required.
   Note: the maintenance information and hardware qualification result must be recorded for future reference.

Hardware Qualification Precautions

1. Environment

Abrupt change of the indoor environment would influence the instrument performance. Therefore, the instrument should be placed in indoor with small temperature fluctuation (< 2℃) and kept away from air flowing source.

2. Installation Position

The installation position is very important for correct qualification and the following requirements should be fulfilled:

- **Good Ventilation and away from Ignition Source**
  Please keep well-ventilated condition when flammable and poisonous solvent is used as the mobile phase. The room should be free from open fire and any other ignition source when flammable solvent is used.

- **Avoid Dust and Corrosive Gas**
  Do not install and use the instrument in environment with dust and corrosive gas as this may influence the service life and performance of the instrument.

- **Away from Strong Magnetic Field**
  Do not install the instrument in environment with heavy magnetic field. If the power cord is interfered by the heavy current noise, heavy-current protector is recommended.

- **Control the Room Temperature and Humidity**
  The room where you place the instrument should be kept at 15-35℃ and 30-75% relative humidity. Note the whole-day temperature will be as stable as possible and cannot exceed 5℃ of variation.

- **Place the Instrument at a Suitable Position**
  Make sure your instrument is placed away from vibration, direct sunlight, heat source and the air outlet of the air conditioner.

- **Adequate Installation Bench Space**
  The bench for instrument installation should have adequate weight capacity

to support the total weight of the HPLC system and relative accessories. The bench should be level, stable and has at least 700mm depth.

Note: if the above requirements are not fulfilled, the instrument might turns over sideways.
If components need to be installed side by side, ensure at least 30mm clearance between neighboring components.

Hardware Qualification Requirements

Please prepare desired devices and sample according to the system configuration of the instrument.

1. Test Equipment

The table below lists the test equipment required for hardware qualification. The unit must have traceable qualification and verified testing results.

Table 6-1 Test Equipment for Hardware Qualification

Equipment	Description
Electronic Balance	To inspect the accuracy of the injection volume of the autosampler. The balance must be calibrated with 0.001 g accuracy in weighing.

2. Standard Reagent

Please prepare the desired standard reagent for qualification according to the following table and prepare ultra-pure water and methanol for the mobile phase and perform ultrasonic degassing for about 20 minutes before using them.

Table 6-2 Standard Reagents for Hardware Qualification

Standard Reagent	Description
Pure Water	To detect the accuracy.
1.0×10^{-4}g/mL Naphthalin /Methanol Standard Solvent	To detect the precision, linearity and cross contamination.

3. Necessaries for Hardware Qualification

The table below lists the necessaries for hardware qualification. In addition, some other necessaries (such as the sample vial) might also be required.

Table 6-3 Necessaries of Hardware Qualification

Articles	Description
PEEK Tubing and Connector	To Connect the autosampler and other devices.
Column	Rittun C18: 5μm, 4.6mm×150 (250) mm,or other column with the same specifications.
Methanol : Water = 80:20	Mobile phase

Hardware Qualification

Autosampler Qualification

1. Inspection Condition

The following table lists the qualification inspection conditions of the autosampler.

Table 6-4 Qualification Inspection Conditions

Inspection Item	Description
Firmware Version Inspection	To inspect the version of the firmware.
Instrument Self-test	To inspect the 6 self-test items of the instrument.
Injection Accuracy	To inspect the injection accuracy.
Injection Precision	To inspect the reproducibility of multiple injections.
Linear Range	To inspect the linearity of the injection volume.
Cross Contamination	To inspect the cleanliness of the injection needle after the wash.
Leakage Sensor Inspection	To inspect the sensitivity of the leakage sensor.

2. Firmware Version Inspection

Purpose: check the version of the firmware.

Procedures:

① Turn on the power switch after the instrument is powered on;

② The VFD goes on and enters the welcome interface, as shown in the figure below.

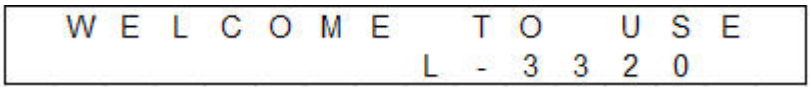


Figure 6-1 Welcome Screen

③ After about 3 seconds, the VFD displays the model number, name and program version number of the instrument, as shown in the figure below.

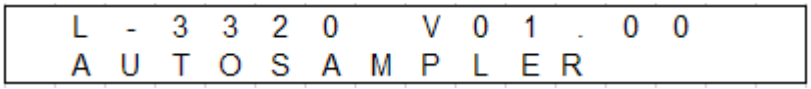


Figure 6-2 Instrument Information Display Interface

Criterion: The version number displayed is the same with the management number.

3. Instrument Self-check

Purpose: inspect whether the six basic internal functions of the instrument are normal.

Procedures:

① Wait for the self-check after the instrument firmware information is displayed;

② After about 3 seconds, the instrument enters self-check automatically. The VFD displays the self-check item the instrument is testing during the self-check as shown in the figure below.

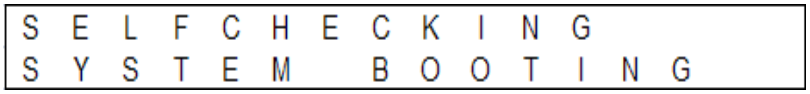


Figure 6-3 Instrument Self-test Interface

The second line on the above interface indicates the self-check items including 6 kinds as shown in the table below. The VFD tests the self-check items one by one.

Self-check Item	Description
SYSTEM BOOTING	The self-test system starts
NEEDLE HOMING	The needle is initializing
TRAY HOMING	The sample tray is initializing
SEL. VAL. HOMING	The distribution valve is initializing
SYRINGE HOMING	The syringe is initializing
INJ. VAL. HOMING	The injection valve is initializing

Criterion: When errors occur during the self-test, the VFD will switch to the alarm display interface (for details, please refer to "**Prompt Messages**"). If the self-test passes, the VFD enters the main interface and switches between the

main interface and the secondary interface.

4. Accuracy Inspection

Purpose: check the injection accuracy of the autosampler.

Procedure: use pure water as the solvent to be tested. Weigh the sample vial with water on the balance and get W1; inject continuously for 10 times (40μL flush volume and10μL injection volume), weigh the injection vial with the pure water and get W2; calculate the injection volume accuracy via formula (1).

Criterion: the injection accuracy of the autosampler is within ±5.0%.

$$A=\frac{W_1-W_2}{V\times\rho\times10}\times100\% \qquad \text{.....(1)}$$

Wherein,

A ——injection volume accuracy;

W_1——the weight of the sample vial with the pure water in the initial state (unit: g);

W_2 ——the weight of the sample vial with the pure water after the injection (unit: g);

V ——the volume of the solvent aspirated from the injection vial at each injection (unit: mL; here, V=0.05mL);

ρ ——the concentration of water under the current temperature (unit: g/cm^3).

5. Precision Inspection

Purpose: inspect the reproducibility of the injection volume of multiple injections.

Procedure: the mobile phase is Methanol/water (80/20), the flow rate is 1.0mL/min, the detect wavelength is 254nm, the injection volume is 10μL and the sample to be tested is 1.0×10^{-4}g/mL Naphthalin/Methanol standard solvent. Inject for 6 times to get the corresponding peak areas and calculate the reproducibility using the peak area data via formula (2).

Criterion: the RSD of the concentration should not be greater than0.5%.

$$\mathrm{RSD} = \frac{1}{\overline{X}} \sqrt{\sum_{i=1}^{n} (X_i - \overline{X})^2 / (n-1)} \times 100\% \qquad \text{..... (2)}$$

Wherein,

X_i ——the peak area got in the i[th] measurement;

$\overline{X}$ ——the arithmetic average of the results of n measurements;

i ——the number of the measurement;

n ——the total number of measurements (here, n=6).

6. Linearity Inspection

Purpose: inspect the linear range of the injection volume.

Procedure: use the 1.0×10^{-4}g/mL Naphthalin/Methanol standard solvent as the sample to be tested. The chromatographic conditions are the same as those in "Precision Inspection" except that the injection volumes are 5μL, 10μL, 15μL, 20μL and 25μL respectively. Draw the standard curve using the peak areas VS the injection volumes.

Criterion: the linearity of the autosampler should be no less than 0.999.

7. Cross Contamination Inspection

Purpose: inspect the cleanliness of the injection needle after the wash.

Procedure: inject the 1.0×10^{-4}g/mL Naphthalin/Methanol standard sample and blank solvent subsequently. Compare the peak areas of them and the percentage that the Naphthalin remained in the blank solvent takes up in the Naphthalin in the standard sample is the cross contamination.
Note: the needle wash liquid volume is 400μL.

Criterion: the cross contamination of the autosampler should be no greater than 0.05%.

8. Leakage Sensor Inspection

Purpose: inspect the sensitivity of the leakage sensor.

Procedure: turn on the autosampler and wet the leakage sensor with the syringe filled with water or wet cotton bud.

Criterion: the VFD displays the prompt message and the beeper sounds when the leakage sensor is wetted.

System Qualification

HPLC is made up of lots of components and the system qualification aims to verify the function of each single component and the performance of the whole system. System qualification which is the foundation of the LC system performance inspection aims to check if the LC system works normally.

Perform system qualification when the instrument is installed for the first time and then regularly. If any problem occurs during the operation, you can perform system qualification to determine whether the problem is on the LC system or on the analysis method. If the LC system passes the qualification, it can be assumed that the LC system works normally and the problem might be on the specific analysis method or the condition used. If the LC system does not pass the qualification, it is assumed that the LC system is abnormal.

1. Gradient LC System Qualification

Purpose: analyze to get the retention time and peak area of each injection. Compare the data obtained to verify the repeatability of the system. The repeatable data can be used for system qualification.

Requirements: pump, column oven, autosampler, detector, data processor

Procedures:

① Inspect all the cable connections in the HPLC system. For details, please refer to the detailed instructions of each module;

② Check if the pipe connection in the HPLC system is normal. The best pipe connection is to minimize the dead volumes outside the column;

③ Wash the new system stream using isopropanol and water respectively at the speed of 2 ml/min for more than 10 minutes;

④ After that, inject the mobile phase (methanol) into the solvent bottle and connect the column;

⑤ Set the flow rate to 1 ml/min. For setting procedures, please refer to the related context in **R**ittun Work Station User's Guide;

⑥ Set the temperature of the column oven to 40℃. For the setting procedures, please refer to the related context in **R**ittun Work Station User's Guide;

⑦ Set the detection wavelength to 254nm. For the setting procedures, please

refer to the related context in **R**ittun Work Station User's Guide;

⑧ Perform "auto zero" after the baseline is stable - inject 20 μL mobile phase and make sure there is no peak;

⑨ Inject continuously for 6 times you will then obtain the analysis data. Analysis conditions: flow rate: 1 ml/min; column oven temperature: 40℃; detection wavelength: 254 nm;

⑩ Obtain the relative standard deviation according to six analysis data from:

$$\mathrm{RSD} = \frac{1}{\overline{X}} \sqrt{\sum_{i=1}^{n} (X_i - \overline{X})^2 / (n-1)} \times 100\%$$

Wherein,

X_i——the retention time or peak area (or peak height) from the i^{th} measurement;

$\overline{X}$——the arithmetical average from n measurements;

i——the number of measurement;

n——the total number of measurements (here n=6).

Criterion:

The calculated RSD should be as follows.
Qualitative measurement RSD is no greater than 0.5%;
Quantitative measurement RSD is no greater than 1.0%.

2. Min Detectable Concentration Qualification

Purpose: inspect the minimum detectable concentration of the system.

Procedures:

① Connect the HPLC system (including the pump, detector, work station, column, injection valve etc.);

② Set the wavelength of the detector to 254nm, the response time (T_{90}) to 0.5s and the pump flow capacity to 1.0ml/min. For detailed operation, please refer to the related context in L-3320 Autosampler User's Guide;

③ Run the system and inject 20μL of 1.0×10^{-7}g/ml Naphthalin/Methanol solvent through the injection valve inlet after the values displayed on the detector become stable;

④ Collect chromatogram and record the peak height of the naphthalin and the short-term baseline noise in the chromatogram. Calculate the min detectable concentration according to the formula below.

$$C_{min} = 2 \times \frac{H_N}{H \times 20} \times c \times V$$

Wherein, the figure "20" is the standard sampling volume and its unit is μL;

C_{min}——min detectable concentration, the unit is g/ml;

H_N ——short-term baseline noise, the unit is mAU;

c ——standard solvent concentration, the unit is g/ml;

H ——the chromatogram peak height of the standard solvent, the unit is mAU;

V ——sampling volume, the unit is μL.

Criterion: the min detectable concentration should not be greater than 1×10^{-7} g/ml.

If the Qualification Fails

If the system or a component does not fulfill any of the system qualification criterions:

It might be some easy-to-be-ignored little problems (such as bubble) that make the system cannot reach the standard:

- Perform failure exclusion to search for similar problems and take corresponding measures to solve all the problems found. For detailed information about the failure exclusion procedures of a single system component, please refer to the corresponding instructions.
- Check if any vulnerable part has reached its service life.

It might be the failure of some vulnerable parts:

- Inspect the vulnerable parts and replace them if necessary.

If you cannot assign the cause of that failure or if you do not know how to exclude the failure or deal with it, please contact Rittun.

Chapter 7 Appendix

Appendix A: L-3320 Autosampler Accessories

Table 7-1 L-3320 Autosampler Accessories

Name	Specification	Quantity
Power Cord	National Standard/L＝1800±15 853 Grey	1
Fuse	250V/4A/Slow/5mm*20mm	3
Data cable	USB cable 1.5m	1
Silicon Drain Tubing	Inter diameter: 80mm, 1.5m	1
Vial Tray	Standard 48 vial tray	2
Sample Vial (include cap and septa)	1.5mL sample vial with cap and septa	100

Appendix B: Solvents Knowledge

Rating Requirements on Mobile Phase

A clean mobile phase is an important guarantee for good result repeatability and little maintenance. If the phase is not clean enough, it will cause big noise and migration as well as block the entrance of the mobile phase (In-line filter). In this case, a HPLC-level mobile phase is recommended and should be filtered and degassed. Note the pore size of the filtration membrane often is 0.45 μm.

Solvents Requirements

In order to prevent micelles carried with solvents from blocking the capillary, solvents should be filtered. Please do not use solvents listed below, because they may corrode the steel parts.

1. Solvents with hard complexing agents (such as EDTA ethylenediamine tetracetic acid).
2. Mixture of carbon tetrachloride and 2-propanol or tetrahydrofuran.
3. Organic acid solvents (acetic acid, formic acid, etc.). For example, the methyl alcohol with 1% acetic acid.
4. HPLC degree ether that may contain oxide (such as THF, dioxane, and dipropyl ether). Peroxide must be filtered from such ether by using dry alumina before using as the mobile phase.
5. Halogenated solvents or mixtures that can form free radicals or acid.
   $2CHCl_3 + O_2 \rightarrow 2COCl_2 + 2HCl$
   In the above chemical equation, the stainless steel can be used as catalyst. Once the dry course removes the stabilizing agent "**alcohol**", the above reaction will made quickly when it comes to dry chloroform.
6. High concentration of inorganic acid such as aqua fortis, sulfuric acid and especially for those sulfuric acids at elevated temperatures (if the chromatogram method requires, use the ortho-phosphoric acid that has poor causticity to stainless steel or phosphate buffer as the mobile phase).
7. Alkali halide and its acid solution (such as lithium iodide and potassium chloride).

Buffer Solution Requirements

Please use at least 10 mL HPLC-level water to wash the pump after you use buffer solution. If the pump stops for more than one day, purge the pump using methanol and water (10:90) to prevent microorganism growth.

Use of Buffer Solution and Tetrahydrofuran (THF)

When the buffer solution combined with water is in use for regulating the pH value of the solvents, indissoluble components should be filtered from the solvents and then mix the solvents with the organic solution. To reduce or prevent production of oxide because of longtime placement and avoid large baseline drift, please use the tetrahydrofuran with unopened package especially for those that are unstable.

WARNING

The tetrahydrofuran with high concentrations of peroxide may cause an explosion.

Table 7-2 Solubility Table of Common HPLC Solvents

Solvent	Cut-off wavelength (nm)	Viscosity (η, 20℃)	Boiling point (℃)	Polarity	Solubility coefficient (M)
n-Decane		0.92	174.1	-0.3	29
iso-Octane	197	0.50	99.2	-0.4	29
n-Hexane	190	0.313	68.7	0.0	29
Cyclohexane	200	0.98	80.7	0.0	28
n-Butyl Ether	220	0.70	142.2	1.7	26
Triethylamine		0.38	89.5	1.8	26
Isopropyl Ether	220	0.33	68.3	2.2	
Toluene	285	0.59	100.6	2.3	23
Paraxylene	290	0.70	138.0	2.4	24
Benzene	280	0.65	80.1	3.0	21
Benzyl Ether		5.33	288.3	3.3	
Dichloromethane	233	0.44	39.8	3.4	20
Ethylene Chloride		0.79	83.5	3.7	20
n-Butanol	210	3.00	99.5	3.9	
Tetrahydrofuran	212	0.55	66.0	4.2	17
Ethyl Acetate	256	0.47	77.1	4.3	19
n-Propanol	240	2.30	97.2	4.3	15
Isopropanol	205	2.35	117.7	4.3	15
Ethyl Formate		0.45	56.3	4.4	15，17
2-Butanone	329	0.43	80.0	4.5	17
Cyclohexanone		2.24	155.7	4.5	28

Nitrobenzene		2.03	210.8	4.5	14，20
Benzyl Cyanide		1.22	191.1	4.6	15，19
Dioxane		1.54	101.3	4.8	17
Ethanol	210	1.20	78.3	5.2	14
Pyridine		0.94	115.3	5.3	16
Nitroethane	380	0.68	114.0	5.3	
Acetone	330	0.32	56.3	5.4	15，17
Benzyl Alcohol		5.80	205.5	5.5	13
2-Methoxyethanol		1.72	124.6	5.7	13
Acetonitrile	190	0.37	81.6	6.2	11，17
Acetic Acid		1.26	117.9	6.2	14
N,N-Dimethyl Formamide	268	0.90	153.0	6.4	12
Dimethyl Sulfoxide	268	2.24	189.0	6.5	9
Methanol	205	0.60	64.7	6.6	12
Formamide		3.76	210.5	7.3	3
Water		1.00	100.0	9.0	

1. If the target solvent is intermiscible with the former one, directly replace it.
2. A mobile phase that can combine with the target and pervious solvents should be introduced if these two solvents are not intermiscible. Remove the pervious solvents thoroughly before new phase is injected.
3. Changing the temperature can affect the intersolubility of the mobile phase. Temperature increment enhances the intersolubility.
4. If you mix the organic solvent with water, the buffer solution dissolved in water may be precipitated. Rinse out the buffer solution using distilled water before you replace the mobile phase from buffer solution into organic solution.

See the following solvent miscibility:

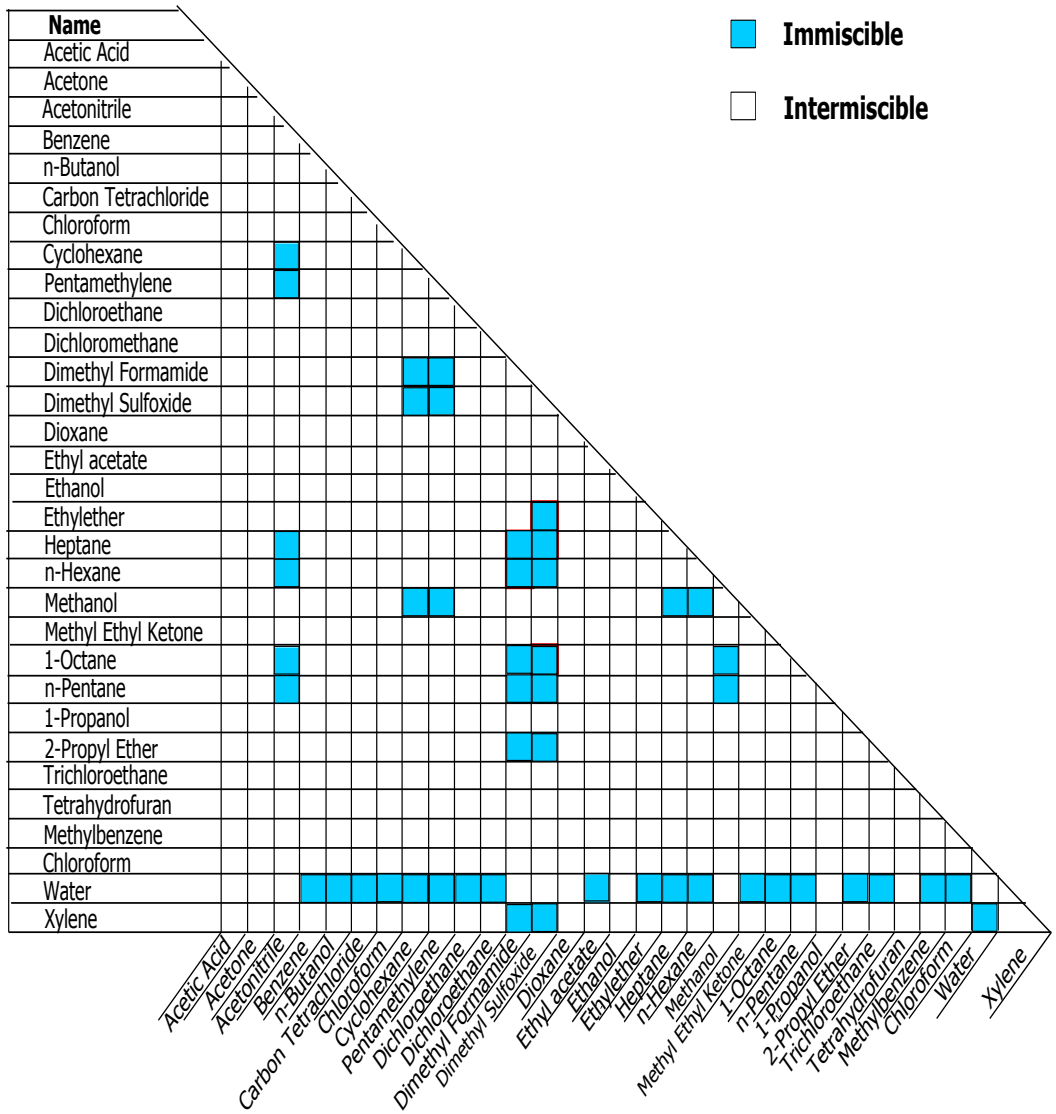


Figure 7-1 Solvent Miscibility Table

Table 7-3 Common HPLC Buffering Agents

Buffering agent	pKa	Buffering range[1]	UV cut-off wavelength[2]
Trifluoroacetic Acid (TFA)	＞2	1.5 - 2.5	210 nm (0.1%)
Acetic Acid/Potassium Acetate	4.8	3.8 - 5.8	210 nm (10 mM)
Tris Hcl/Tris	8.3	7.3 - 9.3	205 nm (10 mM)
Ammonium Chloride/Ammonia	9.2	8.2 - 10.2	200 nm (10 mM)
Phosphoric Acid/Monopotassium Phosphate	2.1	＜3.1	＜200 nm (0.1％)
Phosphoric Acid/Dipotassium Hydrogen Phosphate	7.2	6.2 - 8.2	＜200 nm (10 mM)
Phosphoric Acid/Dipotassium Hydrogen Phosphate	12.3	11.3 - 13.3	＜200 nm (10 mM)
Potassium Bicarbonate/Potassium Carbonate	6.4	5.4 - 7.4	＜200 nm (10 mM)
Potassium Bicarbonate/Potassium Carbonate	10.3	9.3 - 11.3	＜200 nm (10 mM)
Citric Acid/Tripotassium Dicitrate Bismuthate	3.1/4.7/5.4	2.1 - 6.4	＜230 nm (10 mM)
Formic Acid/Potassium Formate	3.8	2.8 - 4.8	＜210 nm (10 mM)
Triethylamine	11.0	10.0 - 12.0	＜200 nm (10 mM)

Remarks:

[1] Permissible pH value range.

[2] The cut-off wavelength is taken to be the wavelength obtained by changing the irradiating wavelength to make the absorbance A=1 when the solvents are contained in a 1 cm optical length absorption cell and the air is taken as a reference. When you

use a UV detector, the detection wavelength must be greater than the cut-off wavelength.

Table 7-4 Mixture Strength Methanol/Water, Acetonitrile/Water

%ACN	%MeOH	%ACN	%MeOH
5	7	55	65
10	14	60	70
15	21	65	74
20	28	70	78
25	34	75	82
30	40	80	86
35	45	85	90
40	50	90	95
45	55	95	98
50	60	100	100

Table 7-5 Pressure Conversion Table

	psi	atm	Kgf/cm^2	KPa	bar	MPa
1 psi=	1	0.068	0.07	6.9	0.07	0.007
1 atm=	14.7	1	1.03	101	1.01	0.101
1 Kgf/cm^2=	14.2	0.968	1	98	0.98	0.098
1 KPa=	0.145	0.01	0.1	1	0.01	0.001
1 bar=	14.5	0.987	1.02	100	1	0.1
1 MPa=	145	9.869	10.2	1000	10	1

Appendix C: Warranty

Rittun warrants that its products mainframe and accessories will be free from defects in materials and workmanship within the warranty period.

If a product is proven to be defective within the respective period, **R**ittun guarantees the free replacement or repair of products which are approved defective. To get repair service, please contact with your nearest **R**ittun sales and service office.

Rittun does not provide any other warranty items except the one being provided by this summary and the warranty statement. The warranty items include but not being subjected to the hint guarantee items related to tradable characteristic and any particular purpose. **R**ittun will not take any responsibility in cases regarding to indirect, particular and ensuing damage.

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