Science Together

KNAUER

AZURA

Autosampler AS 6.1L

Instructions

Document No. V6821

HPLC

Note: For your own safety, read the instructions and observe the warnings and safety information on the device and in the instructions. Keep the instructions for future reference.

Manuel en français: Si jamais vous préfériez un manuel en français pour ce produit, veuillez vous contacter le support technique (Technische Kundenbetreuung) par email ou par fax avec le no. de série. Merci beaucoup.

Technical support

Telephone: +49 30 809727-111 (9-17h, Central European Time)
Fax: +49 30 8015010
E-Mail: support@knauer.net
Languages: German and English

Publisher:

KNAUER Wissenschaftliche Geräte GmbH
Hegauer Weg 38
14163 Berlin
Telephone: +49 30 809727-0
Fax: +49 30 8015010
Internet: www.knauer.net
E-Mail: info@knauer.net

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Product information

AZURA L features

The device is a member of the AZURA L product line and shares a number of common features.

- Removable front cover, for optional device and/or operator protection. The removal of the front cover of the AS 6.1 L leads to the decrease of the syringe speed.
- Instrument stability through a large base area and low center gravity.
- The LEDs indicate the instrument status. Therefore the user knows if the instrument is working properly or whether an error has occured.
- Power connection and control connectors on rear of device.

Identification

The device name can be found on the front panel, above the serial number. A silver sticker on the rear side displays the manufacturer name and address, the product number and power supply specifications.

Intended use

Note: Only use the device for applications that fall within the range of the intended use. Otherwise, the protective and safety equipment of the device could fail.

Operating ranges

The device can be used in the following areas:

- Biochemical analyses
- Chemical analyses
- Food analyses
- Pharmaceutical analyses
- Environmental analyses

Performance features

To make your HPLC/UHPLC separations as efficient as possible, pay close attention to the following:

- Use ultra-pure, filtered solvents – Gradient grade – for HPLC/UHPLC.
- Filtration of substances under analysis
- Use of inline filters

Device variants

Basic

Variant	Basic	Basic Cool/Heat
Order number	AAA00AA	AAA01AA
Pressur resistance	700 bar	700 bar
Sample needle	15 µl	15 µl
Syringe	250 µl	250 µl

Variant	Basic	Basic Cool/Heat
Buffer tubing	500 µl	500 µl
Sample loop	100 µl 0,4 mm ID	100 µl 0,4 mm ID

Standard

Variant	Standard	Cool/Heat
Order number	AAA10AA	AAA11AA
Pressure resistance	1240 bar	1240 bar
Sample needle	15 µl	15 µl
Syringe	250 µl	250 µl
Buffer tubing	500 µl	500 µl
Sample loop	10 µl 0,18 mm ID	10 µl 0,18 mm ID

Bio

Variant	Bio	Basic Cool/Heat
Order number	AAA20AA	AAA21AA
Pressure resistance	345 bar	345 bar
Sample needle	15 µl	15 µl
Syringe	250 µl	250 µl
Buffer tubing	500 µl	500 µl
Sample loop	100 µl 0,4 mm ID	100 µl 0,4 mm ID

Prep

Variant	Basic Cool/ Heat	Prep	Basic Cool/ Heat
Order number	AAA31AA	AAA40AA	AAA41AA
Pressure resistance	200 bar	200 bar	200 bar
Sample needle	60 µl	60 µl	60 µl
Syringe	2500 µl	2500 µl	2500 µl
Buffer tubing	2000 µl	2000 µl	2000 µl
Sample loop	10 ml	10 ml	10 ml

Views

Front view

Legend

① Opening for capillary feed
② Removable door
③ Removable side parts
④ Connection for drainage hose

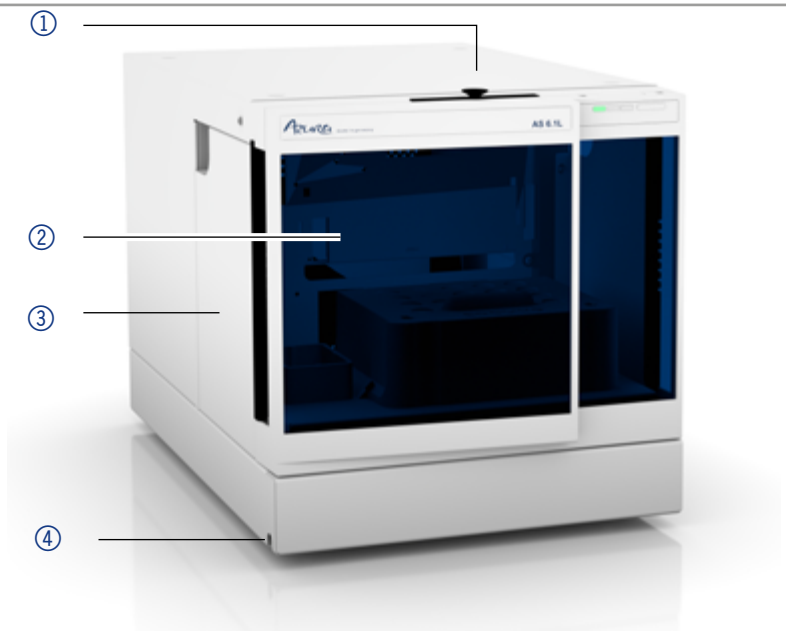


Fig. 1 Front view

Rear view

The following components, connections and warnings can be found at the rear panel:

Legend

① LAN connection
② I/O connection (9-pin)
③ Power switch
④ Fuse box
⑤ Socket for power connection
⑥ Optional ventilator for Cool/Heat-Version
⑦ Serial number and year of manufacture of device

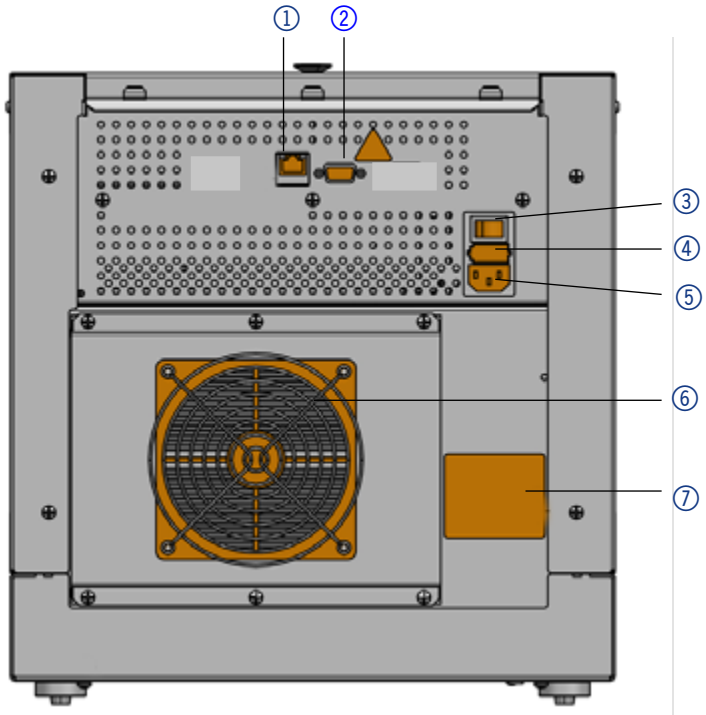


Fig. 2 Rear View

Sample compartment

Legend

① Syringe
② Needle guide
③ Flushing bottle
④ Connection for drainage hose (at the side)
⑤ Injection valve
⑥ Collection container
⑦ Temperature control with sample cooler

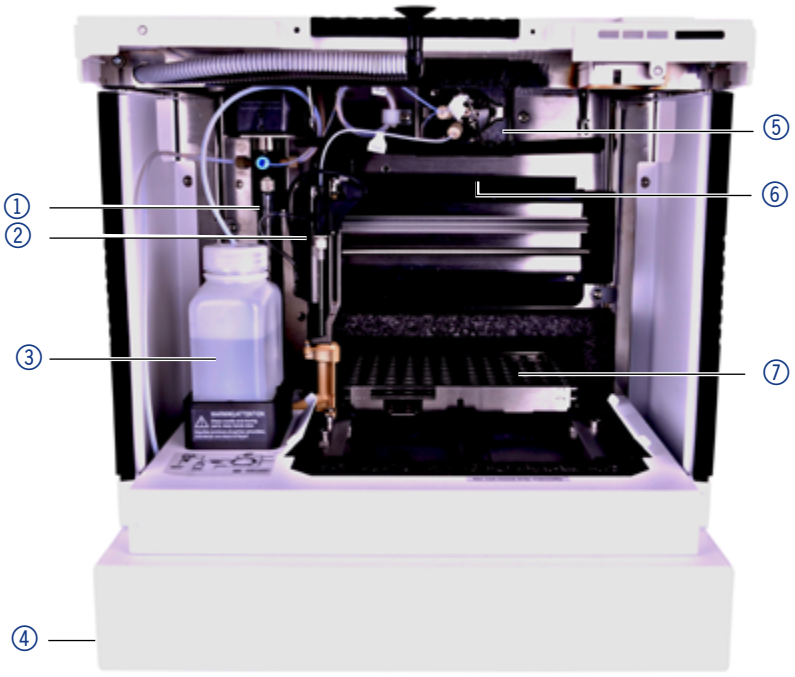


Fig. 3 Sample compartment view

Scope of delivery

Note: Only use spare parts and accessories made by KNAUER or a company authorized by KNAUER.

- Device Autosampler AZURA® Autosampler AS 6.1L
- Supply cable
- CD Autosampler
- Accessories kit AZURA® Autosampler AS 6.1L

Applicable documents:

- AZURA® Autosampler AS 6.1L Instructions (Document number V6821)
- Declaration of conformity

Basic safety instructions

Target group

This document address persons who are qualified as chemical laboratory-technicians or have completed comparable vocational training.

The following knowledge is required

- Fundamental knowledge of liquid chromatography
- Knowledge regarding substances that are suitable only to a limited extent for use in liquid chromatography
- Knowledge regarding the health risks of chemicals
- Participation during an installation of a device or a training by the company KNAUER or an authorized company.

If you do not belong to this or a comparable professional group, you may not perform the work described in these instructions under any circumstances.In this case, please contact your superior.

Safety equipment

When working with the device, take measures according to lab regulations and wear protective clothing:

- safety glasses with side protection
- protective gloves
- Lab coat

What must the user take into account?

- All safety instructions in this document
- The environmental, installation, and connection specifications in this document
- National and international regulations pertaining to laboratory work
- Original spare parts, tools, and solvents made or recommended by KNAUER
- Good Laboratory Practice (GLP)
- Accident prevention regulations published by the accident insurance companies for laboratory work
- Filtration of substances under analysis
- Use of inline filters
- Once the capillaries have been used, never re-use them in other areas of the HPLC system.
- Only use a given PEEK fitting for one specific port and never re-use it for other ports. Always install new PEEK fittings on each separate port.
- Follow KNAUER or manufacturer's instructions on caring for the colums.

More safety-relevant information is listed below:

- flammability: Organic solvents are highly flammable. Since capillaries can detach from their screw fittings and allow solvent to escape, it is prohibited to have any open flames near the analytical system.
- solvent tray: Risk of electrical shock or short circuit if liquids get into the device's interior. For this reason, place all bottles in a solvent tray.
- solvent lines: Install capillaries and tubing in such a way that liquids cannot get into the interior in case of a leak.
- leaks: Regularly check if any system components are leaking.
- power cable: Defective power cables are not to be used to connect the device and the power supply system.
- self-ignition point: Only use eluents that have a self-ignition point higher than 150 °C under normal ambient conditions.
- power strip: If several devices are connected to one power strip, always consider the maximum power consumption of each device.
- power supply: Only connect devices to voltage sources, whose voltage equals the device's voltage. Supply cable: Damaged supply cables must not be used to connect the devices to the power supply.
- toxicity: Organic eluents are toxic above a certain concentration.Ensure that work areas are always well-ventilated! Wear protective gloves and safety glasses when working on the device!

Where is use of the device prohibited?

Never use the system in potentially explosive atmospheres without appropriate protective equipment. For further information, contact the Technical Support of KNAUER.

Secure decommissioning

Take the device completely out of operation by either switching off the power switch or by pulling the power plug.

Opening the device

The device may be opened by the KNAUER Technical Support or any company authorized by KNAUER only.

Signal words

Possible dangers related to the device are divided into personal and material damage in these instructions.

⚠ DANGER — DANGER (red) indicates a hazardous situation which, if not avoided, will result in death or serious injury.

⚠ WARNING — WARNING (orange) indicates a hazardous situation which, if not avoided, could result in death or serious injury.

⚠ CAUTION — CAUTION (yellow) indicates a hazardous situation which, if not avoided, could result in minor or moderate injury.

NOTICE — NOTICE (blue) is used to address practices not related to physical injury.

Decontamination

Contamination of devices with toxic, infectious or radioactive substances poses a hazard for all persons during operation, repair, sale, and disposal of a device.

⚠ DANGER

Life-threatening injuries

Health danger if getting in contact with toxic, infectious or radio-active substances.

➔ Before disposing of the device or sending it away for repair, you are required to decontaminate the device in a technically correct manner.

All contaminated devices must be properly decontaminated by a specialist company or the operating company before they can be recommissioned, repaired, sold, or disposed of. All materials or fluids used for decontamination must be collected separately and disposed of properly.

Decontamination Report

Devices without a completed Decontamination report will not be repaired.If you would like to return a device to KNAUER, make sure to enclose a completed **Decontamination report** with the device: https://www.knauer.net/en/Support/contact

Symbols and signs

The following symbols and signs can be found on the device, in thechromatography software, or in the instructions:

Symbol	Meaning
Warning signs 	Electric shock hazard
	Electrostatic discharge hazard, damages to system, device, or components can occur.
CE Mark 	A device or system marked with CE fulfills the product specific requirements of European directives. This is confirmed in a Declaration of Conformity.
	Testing seals in Canada and the USA at nationally recognized testing centers (NRTL). The certified device or system has successfully passed the quality and security tests.
Notes 	Notes provide useful tips or information worth knowing.

Unpacking and setup

This chapter describes all preparatory steps prior to start-up.

Preparations

Work location

Note: The autosampler is exclusively designed for use in enclosed spaces. The intended use be ensured only if the requirements for ambient conditions of the operating environment are met. You will find the ambient conditions under Technical Data.

NOTICE

Device defect

The device overheats at exposure to sunlight and insufficient air circulation. Device failures are very likely.

➔ Set up the device in such a way that it is protected against exposure to direct sunlight.

➔ Leave room for air circulation: See space Requirements.

General requirements

- Position the device on a level surface.
- Protect the device against direct exposure to sunlight.
- Set up the device at a location not exposed to air drafts such as air conditioning systems.

- Do not set up the device near to other machines that cause floor vibrations.
- Keep the the devices away from high frequency sources. High frequencies may compromise measuring values.

Space requirements

- At least 5 cm, if there is another device on one side.
- At least 10 cm, if there are devices set up on both sides.
- At least 15 cm to the cooler fan on the rear.

Power supply

For power supply, use the supplied power cable and power adapter to meet the specifications which are described in the chapter Technical Data. Inspect the provided power cable beforehand to ensure that it is approved for your country. Replace defective power cables only with accessories from KNAUER. Detachable power cables are not allowed to be replaced with other cable types.

The maximum power input is 200 VA.

NOTICE

Electronic defect

Electronic hazard when using an identically constructed power adapter from another manufacturer.

➔ Only use spare parts and accessories from KNAUER or a company authorized by KNAUER.

Note: The nominal capacity of the connected devices must be maximum 50 % of the power supply to account for larger inrush currents when switching on the modules.

Conditions

- The electrical power supply at the installation site must be connected directly to the nearest main power line.
- The power must be free from ripple, residual current, voltage peaks and electromagnetic interference.
- The connectors for the mains voltage are grounded accordingly.
- The device receives sufficient power with reserve capacity

Power plug

- The device is intended for use with AC power networks of 100-240 V.
- Make sure that the power plug on the rear of the device is always accessible, so that the device can be disconnected from the power supply.

Unpacking the device

Prerequisites

- Check the carton for damage caused during transportation.

Tools

- Utility knife

⚠ CAUTION

Bruising danger

Damage to the device by carrying or lifting it on protruding housing parts. The device may fall and thus cause injuries.

➔ Lift the device only centrally on the side of the housing.

Process
1. Set-up the package in such a way that you can read the label. 2. Using the utility knife, cut the adhesive tape and open the packaging.
3. Lift the foam padding. Take out the accessories kit and the manual. 4. Open the accessories kit and check the scope of delivery. In case any parts are missing, contact the Technical Support.
5. Clasp the device from below, lift it out of the packaging and place it on its feet. Do not hold onto the front cover. 6. Check the device for signs of damage that occurred during transport. In case any parts are missing, contact the Technical support.
7. Set up the device at the operation site. 8. Remove the protective foil.

Next steps Store packaging and keep the included packing list for repeat orders.

Setting up the Device

⚠ DANGER

Life-threatening injuries

Electric shock possible due to incorrect positioning of devices on top of the autosampler. The solvent waste connection could dislocate and cause a leakage. This would cause a decrease of the electrical isolation of the autosampler.

➔ Position the devices carefully on top of the autosampler.

➔ Check the alignment of the solvent waste connection.

Connecting the device in a local area network (LAN) to a computer

The autosampler is operated by the chromatography software exclusively.

Remote control

Normally, the autosampler is controlled by the chromatography software through a local network (LAN).

Automatic configuration

The autosampler connected to the local area network (LAN) is automatically detected by the chromatography software.

Device status

When used in a local area network (LAN), the system status of the autosampler can be checked using the chromatography software.

Note: HPLC devices made by KNAUER work only with IP adresses which are assigned via IPv4. IPv6 is not supported.

This section describes how to set up an HPLC system in a local area network (LAN) and how a network administrator can integrate this LAN into your company network. The description applies to the operating system Windows and all conventional routers.

To set up a LAN, we recommend to use a router. That means the following steps are required:

Process

1. On the computer, go to the control panel and check the LAN properties.
2. Hook up the router to the devices and the computer.
3. On the computer, configure the router to set up the network.
4. Install the chromatography software from the data storage device.
5. Switch on the device and run the chromatography software.

Configuring the LAN settings

The LAN uses only one server (which is normally the router) from that the devices automatically receive their IP address.

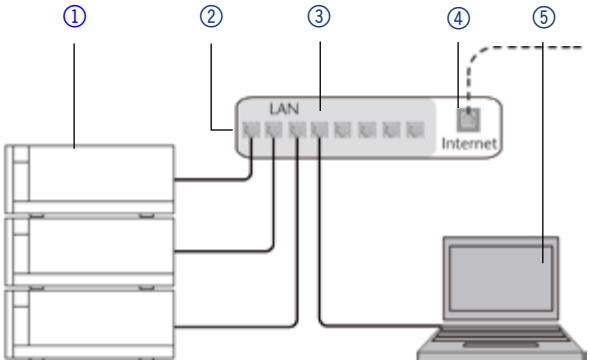
Prerequisites

- In Windows, power saving, hibernation, standby, and screen saver must be deactived.
- In case you use an USB-to-COM box, the option „Allow the computer to turn off ths device to save power“ in the devicemanager must be deactivated for all USB hosts.
- For all LAN devices: For the network adapter, the following option in the Device Manager must be deactivated: „Allow the computer to turn off this device to save power“.

1. In Windows open the **Network and Sharing Center.**
2. Double-click on **LAN Connection**.
3. Click on the button **Properties.**
4. Select **Internet Protocol version 4 (TCP/IPv4)**.
5. Click on the button **Properties**.
6. Check the settings in the tab **General**. The correct settings for the DHCP client are:
   a) Obtain IP address automatically
   b) Obtain DNS server address automatically
7. Click on the button **OK**.

Connecting the cables

A router ② has several LAN ports ③ and one WAN port ④ that can be used to integrate the LAN into a wide area network (WAN), e.g. a company network or the Internet. In contrast, the LAN ports serve to set up a network from devices ① and a computer ⑤. To avoid interference, we recommend operating the HPLC system separately from the company network.



You will find patch cables for each device and the router in the accessories kit. To connect the router to a WAN, an additional patch cable is required, which is not supplied within the scope of delivery.

Prerequisites

- The computer has been switched off.
- There is a patch cable for each device and the computer.

Process

1. Use the patch cable to connect the router and the computer. Repeat this step to connect all devices.
2. Use the power supply to connect the router to the mains power system.

Configuring the router

The router is preset at the factory. You find information about IP address, user name and password in the router instructions: https://goo.gl/ahGhmG.

Process

1. To open the router configuration, start your Internet browser and enter the IP address (not for all routers).
2. Enter user name and password.
3. Configure the router as DHCP server.
4. In the router configuration, check the IP address range and make changes if necessary.

Note: If the IP address range has been changed, it is necessary to note it down.

Result

Once the router has assigned IP addresses to all devices, the chromatography software can be used to remotely control the system.

Integrating the LAN into a company network

A network administrator can integrate the LAN into your company network. In this case you use the WAN port of the router.

Prerequisite

- There is a patch cable for the connection.

Process

1. Check that the IP address range of the router and of the company network do not overlap.
2. In case of an overlap, change the IP address range of the router.

3. Use the patch cable to connect the router WAN port to the company network.
4. Restart all devices, including the computer.

Controlling several systems separately in a LAN

Devices connected to a LAN communicate through ports, which are part of the IP address. If more than one HPLC system is connected to the same LAN and you plan on controlling them separately, you can use different ports to avoid interference. Therefore, the port number for each device must be changed and this same number must be entered into the device configuration of the chromatography software. We recommend to use the same port number for all devices in the same system.

Note: The port is set to 10001 at the factory. You must use the same numbers in the device configuration of the chromatography software as in the device, otherwise the connection fails.

Process

1. Find out port number and change it on the device.
2. Enter the port number in the chromatography software.

Result The connection is established.

Operation

Note: Before initial startup, wait approximately one hour until the temperature of the device has adapted to the ambient temperature.

Auto-injection system

The speed of the auto-injection system has been increased to fulfill the requirements of ultra-high performance liquid chromatography. However, be aware that the high speed of the auto-injection system can cause stab injuries when handled inappropriately. When the door of the autosampler is open, the syringe speed is reduced automatically.

Removing the front cover and the side parts

⚠ DANGER

Life-threatening injuries

Health danger if getting in contact with toxic or biohazardous substances as a result of a scratch or needle stick with the needle in the sample compartment .

➔ Operate the device only with the front panel closed.
➔ Stop operation before opening the front panel.

⚠ CAUTION

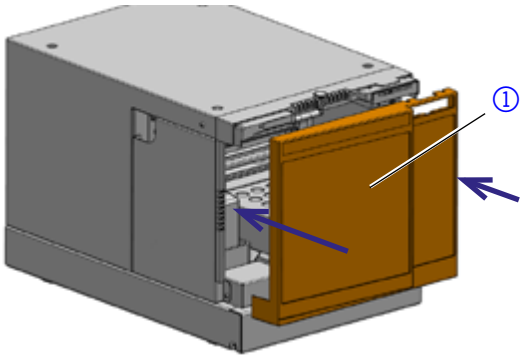
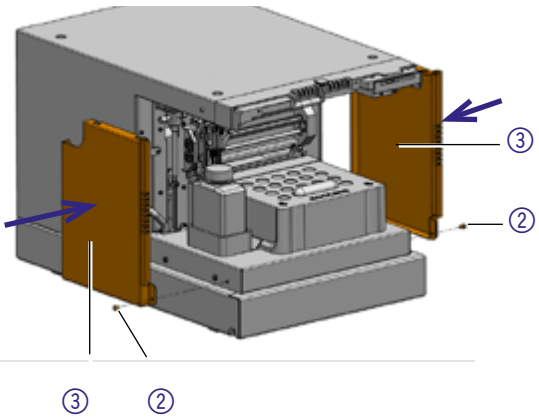
Stitching wounds

Behind the front panel is a chamber with a needle automatically transporting the sample during operation. Carelessness can lead to puncture injuries.

➔ Operate the device only with the front panel closed.
➔ Stop operation before opening the front panel.

Prerequisites The device is switched off.

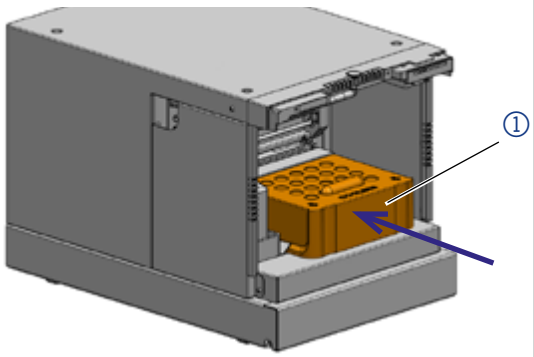
Tools Allen wrench

Process	Figure
1. Hold the front cover ① on both sides and remove it forwards.	 Fig. 4 Removing the front cover
2. Loosen the screws ② with an allen wrench. 3. Remove the side parts ③ .	 Fig. 5 Removing the side parts

Removing the cooler cover

Prerequisites

- The device is switched off.
- Front cover has been removed.

Process	Figure
1. Remove the cooler cover ① forwards.	 Fig. 6 Removing the Cooler Cover

Injection principles

The autosampler can be operated according to the following principles:

ILD™ for analytical autosamplers

For injections in the high pressure range up to 1000 or 700 bar, the autosampler has an ILD™ valve (Intermediate Loop Decompression by Spark Holland). This valve consists of a rotor-stator combination and a central port for pressure release. For applications in the high pressure range, pressure is released from the sample loop to avoid diluting the sample by eluent. Extremely fast switching valves reduce pressure surges further on. The results are more exact analyses and long-lasting columns.

Legend

① Sample loop
② Syringe
③ Sample vial
④ Column
⑤ Pump

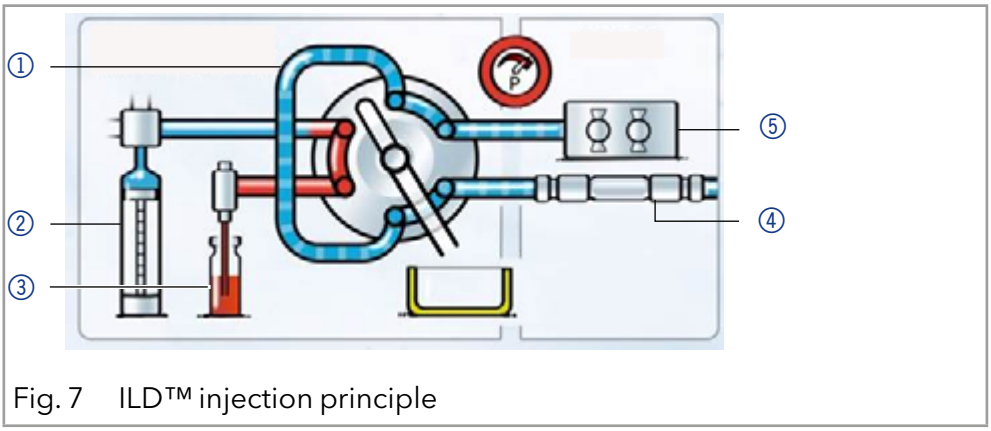


Fig. 7 ILD™ injection principle

PASA™ Loop injection

Loop injection with pressure assistance (Pressure Assisted Sample Aspiration PASA™) features the following:

- Samples do not have to be degassed.
- No air bubbles in sample loop.
- No clogging or contamination of sample needle.
- Precise control of syringe movement

Legend

① Buffer tube
② Syringe
③ Sample needle
④ Capillary to pump
⑤ Capillary to tube
⑥ Sample loop
⑦ Connector for compressed air
⑧ Air needle

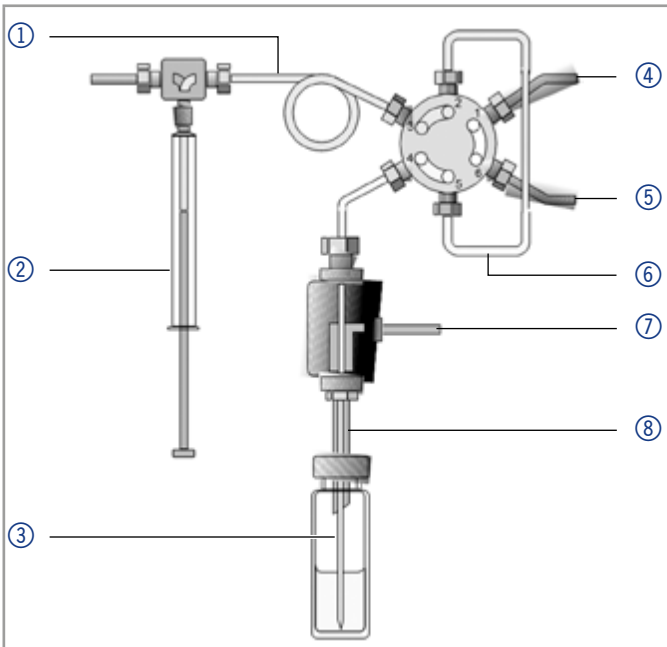


Fig. 8 PASA™ loop injection

Three different injection methods are available:

- Full loop filling
- Partial loop filling
- Microliter-pickup
- Microliter pick-up 84+3:

Full loop filling

In full loop filling mode, the sample loop is completely filled with the sample. The maximum reproducibility but not the maximum precision is achieved because the size of the sample loop may have a deviation of ±10 %. The maximum injection volume equals the loop volume. The sample loop is filled with a multiple of the loop volume:

- 3 x loop volume for loops up to 100 µl
- 2 x loop volume for loops from 100 µl to 500 µl
- 1.5 x loop volume for loops of more than 500 µl

The sample loss per injection is the sum of the overfilling of the sample injection times x and the flush volume set for the needle used.

Partial loop filling

In partial loop filling mode, the sample loop is filled with both sample and mobile solvent. This ensures the highest precision of the sample volume with minimal loss of sample. The maximum injection volume equals 50 % of the loop volume. The sample loss per injection equals the adjusted flush volume plus three times the sample volume for the needle used.

Microliter-pickup

In microliter pick-up mode, the sample loop is filled with a very small amount of sample and transport liquid or wash solution (mobile phase). This ensures very high precision with no loss of sample.

Microliter pick-up 84+3:

If the 84+3 sample plate is selected for the microliter pickup, the sample is transported by a separate transport liquid instead of the flushing fluid. The consumption of transport liquid depends on the needle volume and corresponds to the 2.5 needle volume for the segments before and after the sample.

The autosampler uses a system of two telescopic needles, one that pierces through the cap of the sample vial – the air needle – and one that extracts the sample – the sample needle.

Using a syringe, the sample is aspirated through the two needles out of the sample vial while under pressure and into the sample loop. To prevent the syringe from becoming contaminated, a buffer tube is situated between the syringe and the valve. Using washing solution, sample residue is removed from the sample needle and buffer tube.

Full loop filling

The sample loop is completely filled with sample. This kind of injection leads to outstanding reproducibility.

Reducing consumption

To reduce the flush volume, you can use an air segment of 5 µl. The air segment precedes the flush segment and is not injected.

The flush volume must be at 30 µl in case of a standard needle and an injection with air segment; in case of an injection without air segment, the flush volume has to be 35 µl. You may need to insert a higher flush volume for extremely viscous samples to reduce the syringe speed and improve the performance.

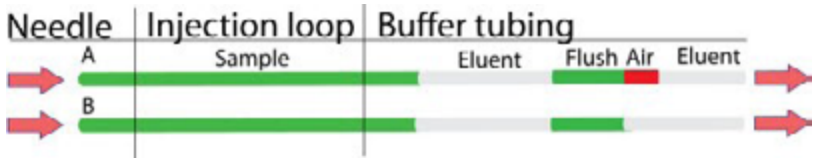


Fig. 9 Air segment with full loop filling.

Note: A flushing process takes places after every injection.

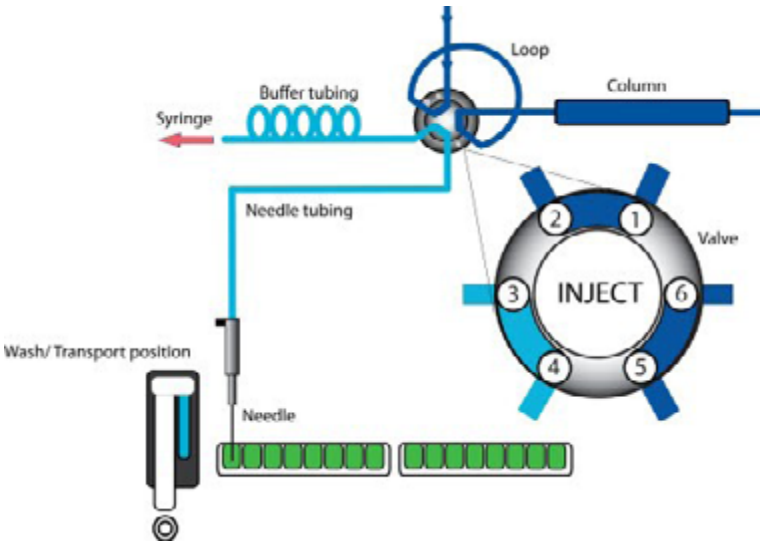
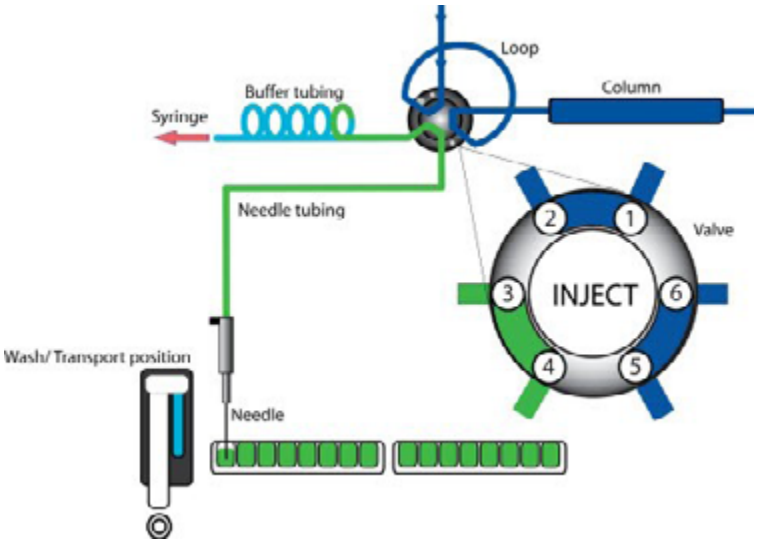
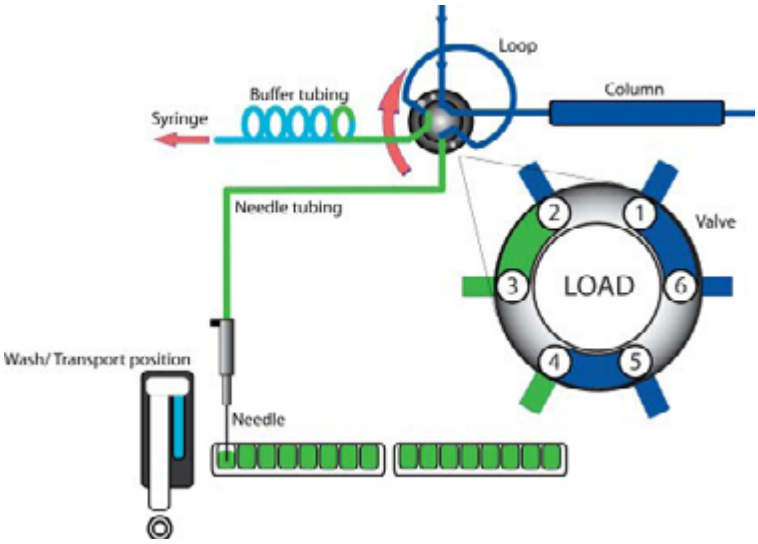
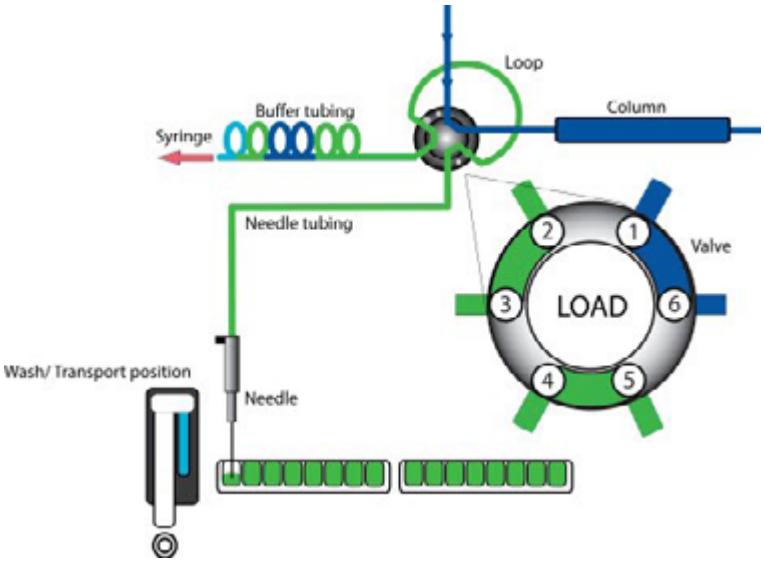
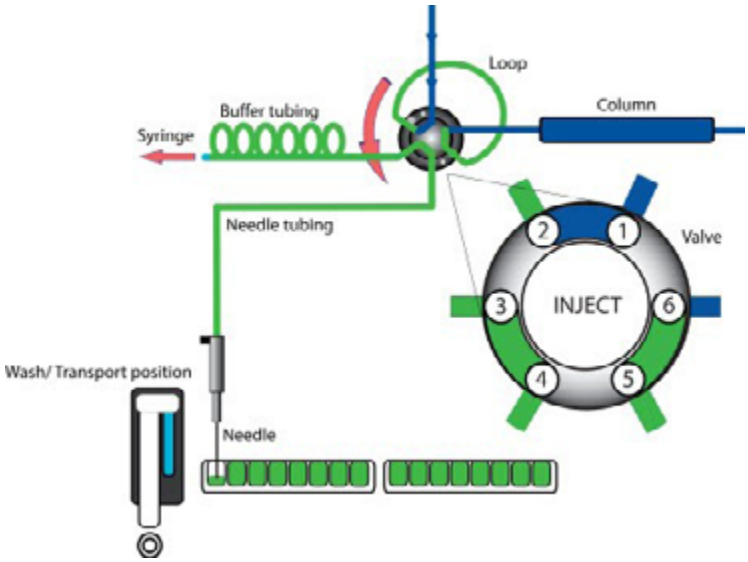
Explanations	Functional schematic
1. Initial conditions: The injection valve in INJECT position. Sample needle and air needle are inserted into the vial. The air needle creates pressure, which prevents air and steam bubbles from developing.	

Fig. 10 Full loop filling: Initial conditions

Explanations	Functional schematic
2. The syringe aspirates the flush volume from the sample vial into the sample line and removes any washing solution.	 Fig. 11 Full loop filling: The needle and sample lines are flushed
3. The valve switches to LOAD position in order to transport the sample material to the inlet of the sample loop.	 Fig. 12 Full loop filling: valve switches to LOAD position
4. The sample loop is filled by transporting a certain amount of the loop volume (depending on the volume of the loop) through the loop. 3 x loop volume for loops up to 100 µl	 Fig. 13 fFull loop filling: The sample loop is completely filled.

Explanations	Functional schematic
5. The valve switches to the INJECT position. From that moment, the sample loop is part of the HPLC flow of the mobile phase. The sample is transported to the column. The analysis starts.	 Fig. 14 Full loop filling: injection valve switches to INJECT position

Next steps Flush the needle after every injection.

Partial loop filling

This kind of injection results in maximum conciseness of sample volume.

Reducing consumption An air segment of 5 µl preceding the flushing volume, which is not injected, can reduce the amount of sample dilution caused by the dispersion during aspiration.

- Choose the following flush volumes for a standard needle:
  - At least 30 µl for injections with air segment
  - 35 µl for injections without air segment
- To achieve better results for highly viscous samples, increase the flush volume and reduce the syringe speed.

Fig. 15 Air segment at partial loop filling

Sample volume In the autosampler AS 6.1L, the syringe moves the sample into the sample loop. In partial loop filling mode, the sample volume is maximum 50 % of the loop volume.

Note: Partial loop fill is processed automatically.

Legend

① Flushing solution
② Tube connector for waste
③ Syringe
④ Drainage tube
⑤ Valve
⑥ Sample loop
⑦ Column
⑧ Needle
⑨ Microtiter plate with samples
⑩ Capillary

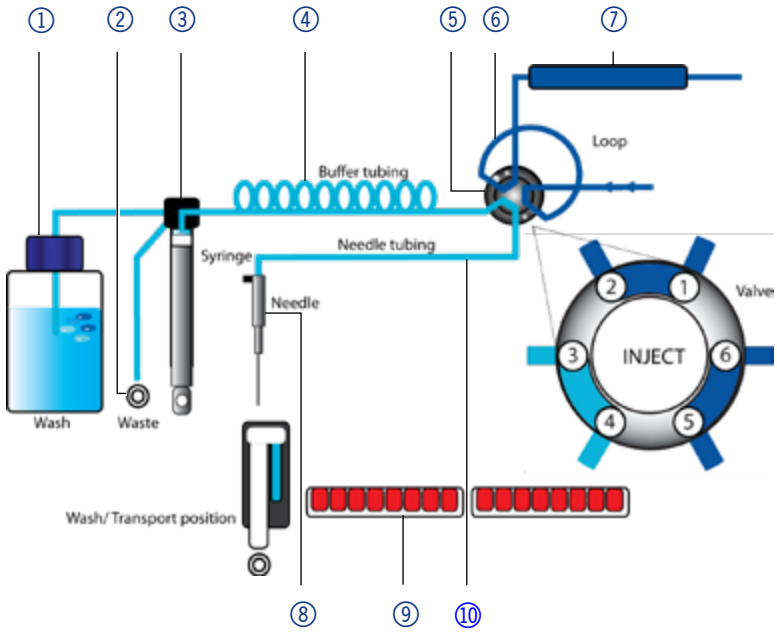


Fig. 16 Scheme of initial position for partial loop filling

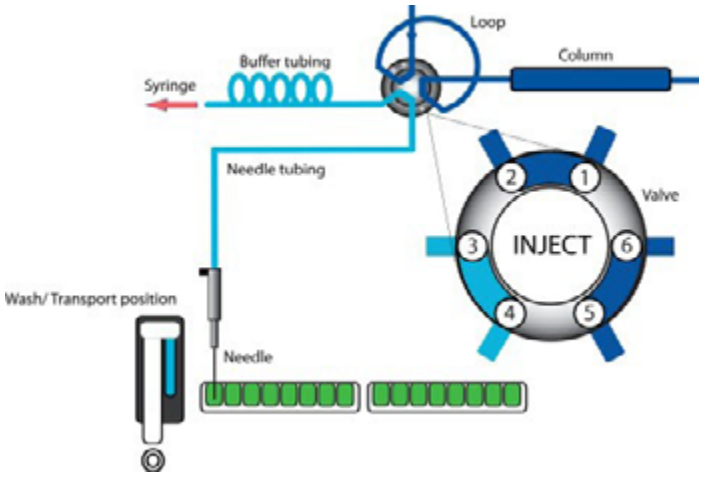
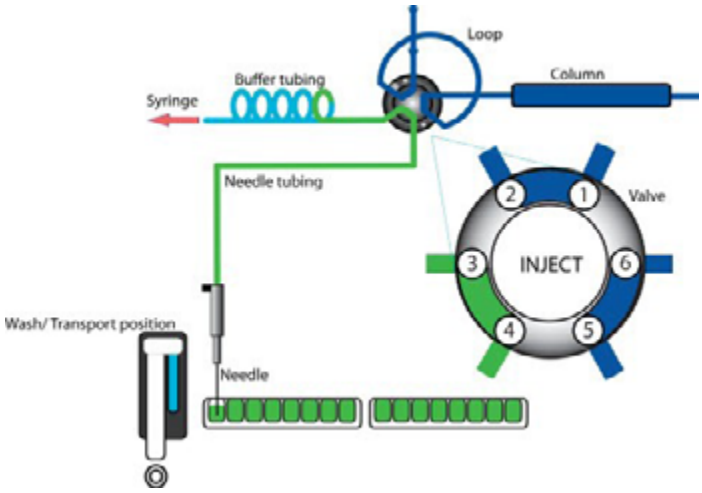
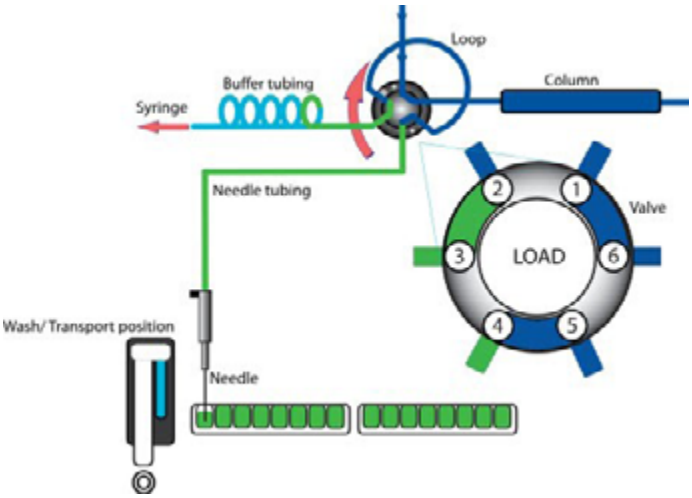
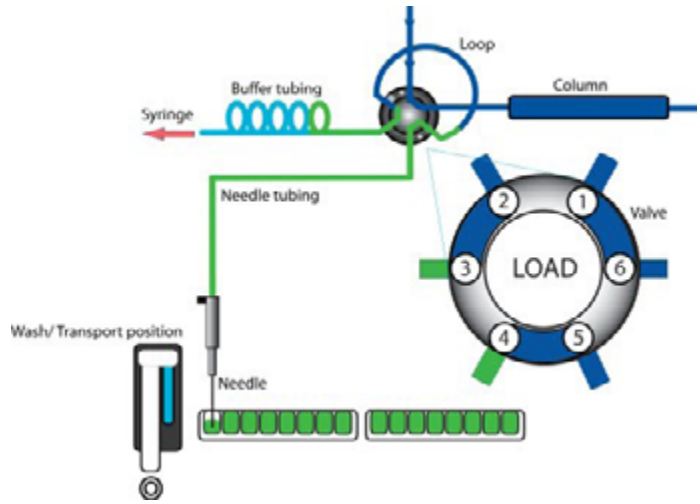
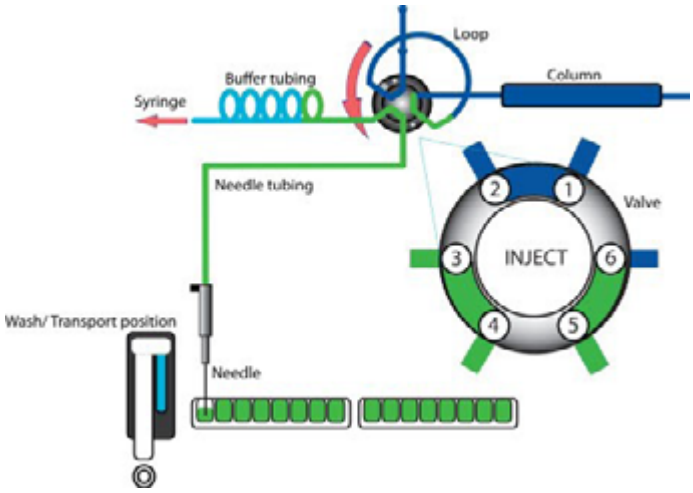
Process	Figure
1. The valve switches to the INJECT position. The tube and the capillary are filled with flushing solution.	

Fig. 17 Partial loop filling Loading the Valve Position

Process	Figure
2. The needle moves into the sample and the syringe aspirates the sample. The tube is partially and the capillary fully filled with sample.	 Fig. 18 Partial loop filling flushing sample and eluent
3. The valve switches to the LOAD position.	 Fig. 19 Partial loop filling The injection valve switches to LOAD.
4. In the AS-1, the syringe aspirates the sample into the sample loop. The tube is full of flushing solution and sample, the capillary of sample and eluent.	 Fig. 20 Partial loop filling Sample loop is partially filled

Process	Figure
5. The valve switches to the INJECT position and the pump transports the sample and the eluent to the column.	 Fig. 21 Partial loop filling Sample is injected

Microliter-pickup

For this kind of injection, the sample is delivered into the sample loop by flushing solution or transporting fluid. The process results in maximum precision of sample volume without sample loss.

The AS 6.1L provides two options to connect two different solvents onto the syringe port (see Fig.22). The port on the left to the syringe port ① is for the transport fluid, the port on the right ③ is for the wash solution. Using the software, you can choose for the microliter-pickup between the transport fluid OR the wash solution.

Legend

① Tubing connection for transport liquid (not available for all models)
② Connection buffer tube
③ Connection wash solution tube

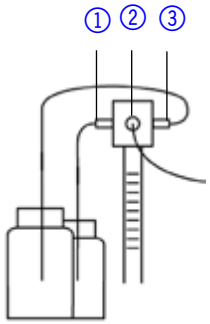


Fig. 22 Syringe connections

Reducing consumption

The following conditions apply:

- The air segment in front of the sample segment is injected into the system.
- In this mode, the needle does not create pressure because the sample volume could be distorted due to the air expansion during the movement from sample vial to wash position.

Note: Make sure that washing liquid and eluent are compatible. Use the software to flush the tubing extensively with transport liquid or wash solution. Depending on the chromatography software, there is a button "Transport Flush" to flush the fluidics with transport fluid.

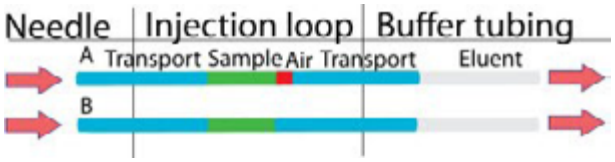
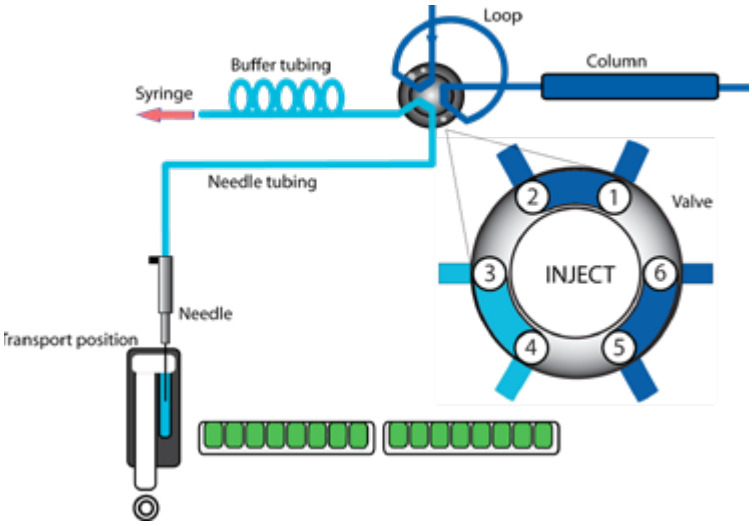
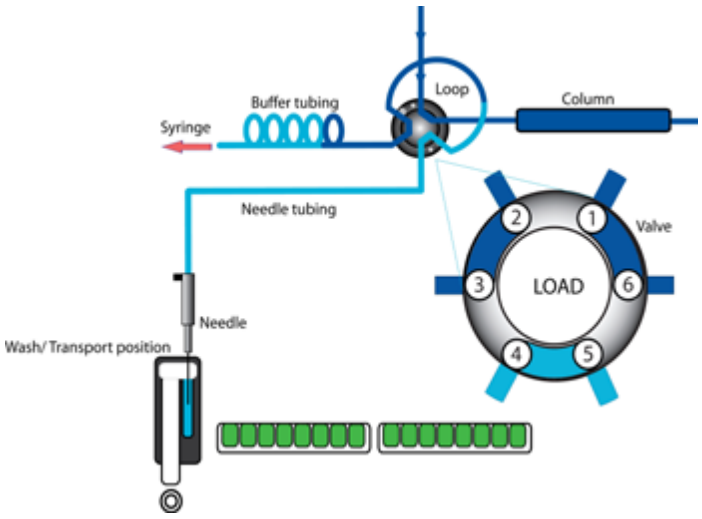
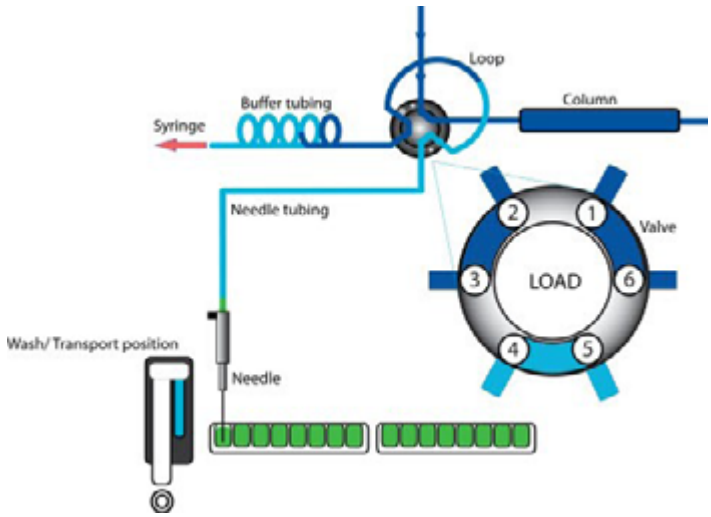
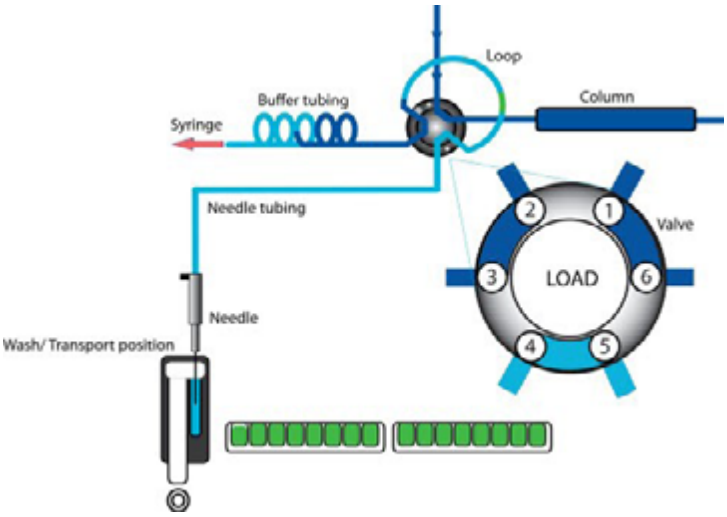
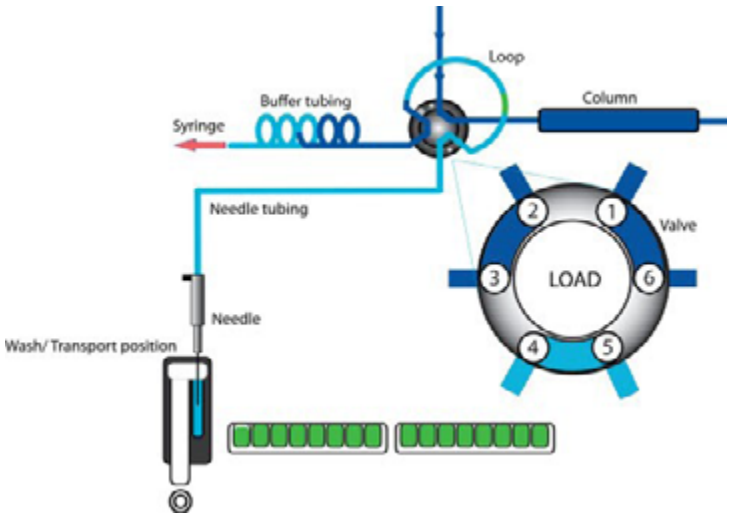


Fig. 23 Microliter pickup with air segment (A), without air segment (B)

Process	Figure
1. Initial conditions: The injection valve in INJECT position. The sample needle is in position wash/ transport. The transport container is filled with washing liquid. 2. The sample line is filled with wash solution until the fluid reaches the sample loop inlet. The valve remains in the INJECT position during filling/transportation.	 Fig. 24 Microliter-pickup Initial conditions
3. The injection valve switches to LOAD. A transport segment of wash fluid is sucked into the sample loop.	 Fig. 25 Microliter-pickup Sample line is filled with eluent

Process	Figure
4. The needle moves from the transport position to the sample vial.	 Fig. 26 Microliter-pickup Sample is aspirated
5. The sample is aspirated from the sample vial according to the amount of programmed injection volume.	 Fig. 27 Microliter-pickup The injection volume is aspirated.
6. The needle moves back into transport position. A second wash solution sample is sucked in. The sample is transported through the sample loop.	 Fig. 28 Microliter-pickup The sample is transported into the sample loop.

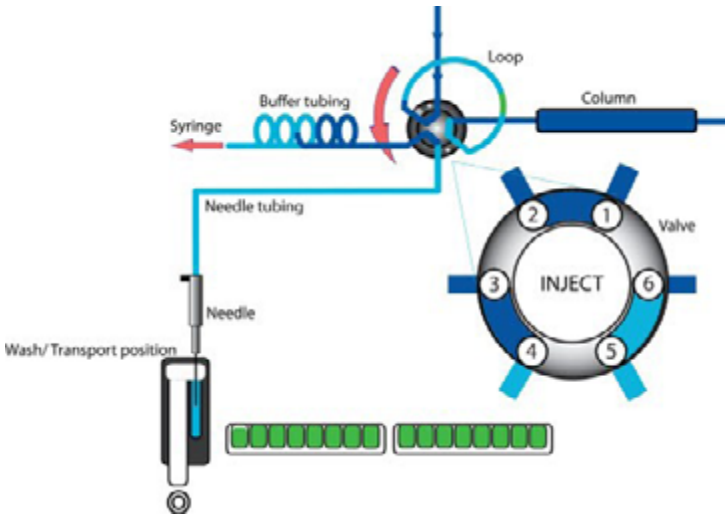
Process	Figure
7. The valve switches to INJECT. From that moment, the sample loop is part of the flow of the mobile phase. The sample is transported to the column. The analysis starts.	

Fig. 29 Microliter-pickup The sample is transported to the column.

Microliter pickup with 84+3 vial plate

If you choose to operate the microliter pickup with the 84+3 vial plate, three 10 ml vials are used automatically for transportation. In this case, the needle position wash/transport is only used to wash the needle.

Reducing consumption

If an air segment has been programmed, it is inserted in front of the transportation liquid segment and in front of each sample. In this injection mode, the air segment in front of the sample segment is injected into the system.

In this mode, the needle does not create pressure because the sample volume could be distorted due to the air expansion during the movement from sample vial to wash position

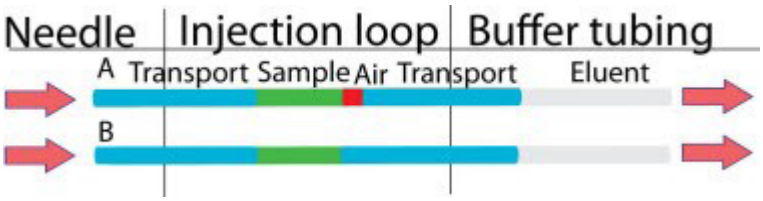
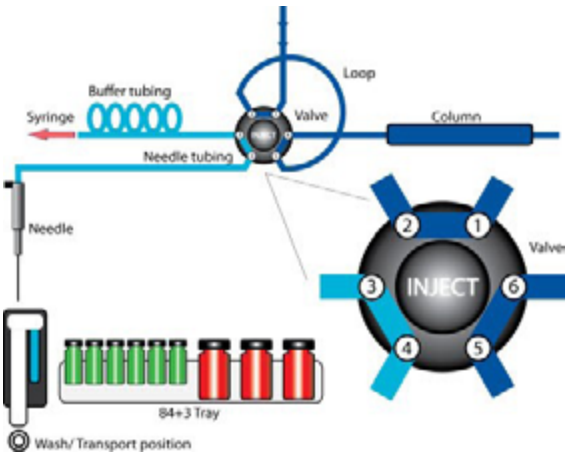
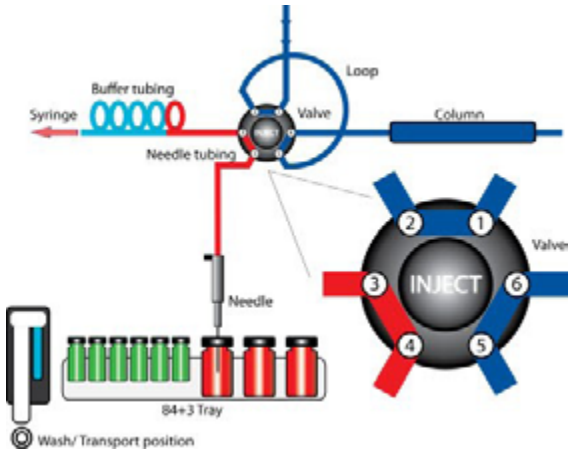
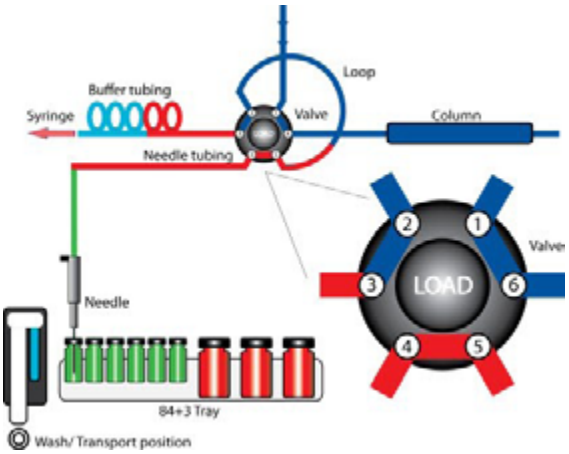
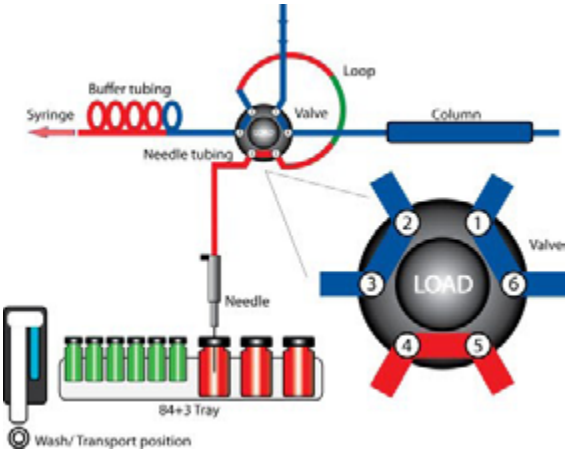
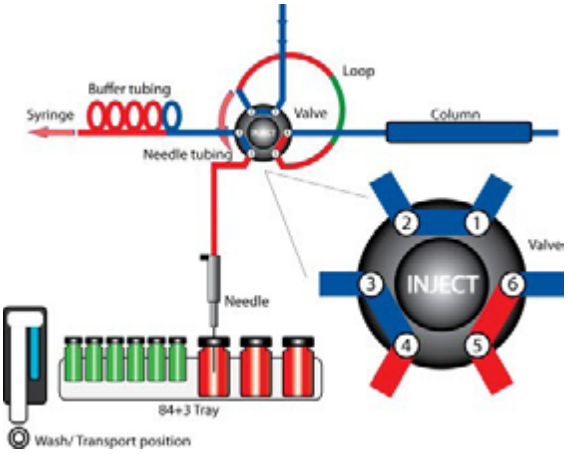


Fig. 30 microliter pickup 84+3 with air segment (A), without air segment (B)

Process	Figure
1. The sample needle begins with the wash/transport position. The valve starts with the INJECT position.	 Fig. 31 Initial conditions
2. The first injection starts in the transport position with the syringe aspirating transport liquid from the vial to fill the sample line with transport liquid and to remove the flushing solution.	 Fig. 32 Sample line is filled with transport liquid.
3. The injection valve switches to LOAD. The programmed injection volume is aspirated out of the sample vial.	 Fig. 33 Injection valve switches to LOAD position

Process	Figure
4. The needle moves back into the transport position. A second segment of transport liquid is aspirated. The sample is transported through the sample loop.	 Fig. 34 The sample is transported into the sample loop.
5. The valve switches to INJECT. From that moment, the sample loop is part of the flow of the mobile phase. The sample is transported to the column. The analysis starts.	 Fig. 35 The injection valve switches to position INJECT.

84+3 vial plate

The 84+3 vial plate is to be ordered separately. Positionthe plate on top of the plate holders inside the autosampler.

Note: Position 87 of the vial plate must always be located at the rear right corner of the sample compartment.

The vial plate offers space for maximum 84 vials with 1.5 ml and 3 vials with 10 ml. It was designed with the caps of all vials being level,

independently from their unique heights, which means that one needle can be used for all vial sizes.

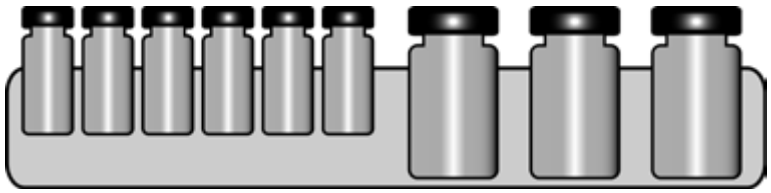


Fig. 36 Height of 84+3 vial plate

The length of the sample needle must be programmed at 1.5 ml. Relating to the liquid level in 10 ml vials, it is possible to program the needle at two different lengths.

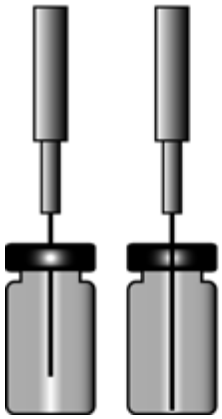


Fig. 37 Needle length and liquid level

Follow the numeric order if you are to program a sample sequence for a vial plate.

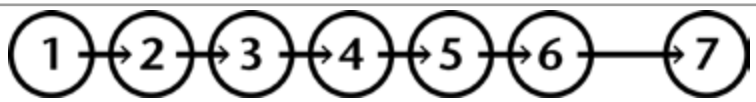


Fig. 38 Sequence

In case you are creating a sequence chart with only one sample per row (1 sample in each run), programming the sequence is free to you.

Microliter-pickup parameters

Before using the 84+3 vial plate, it is necessary to change the software settings.

Note: If you choose to operate the microliter pickup with the 84+3 vial plate, three 10 ml vials are used automatically for transportation. In this case, the needle position wash/transport is only used to wash the needle.

The vial positions for the 84+3 vial plate are as follows:

Position of the first sample:	Vial positions 1-84
Position of the final sample:	Vial positions 1-84
First destination position:	Vial positions 1-84
Transport position:	Vial positions 85-87

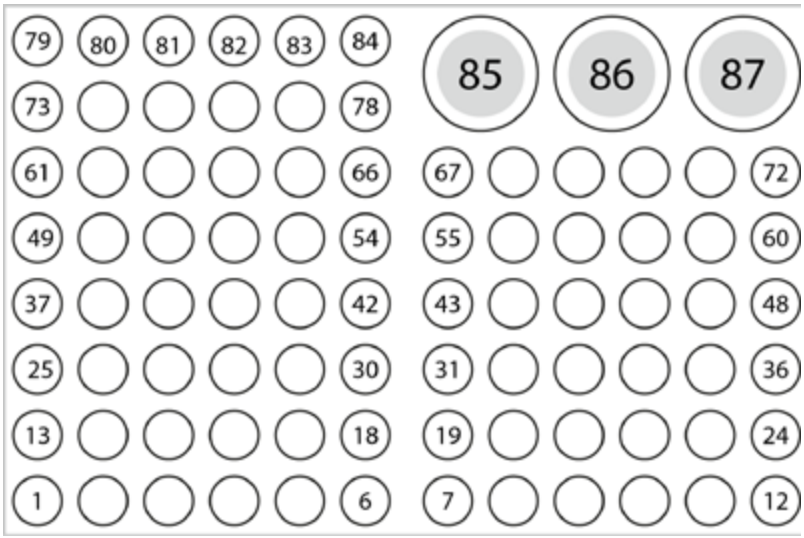


Fig. 39 Reagent/Transport position on 84+3 vial plate

You need to define the first and the final sample to enter a sample range. The transport positions are always positions 85, 86, 87.

Details on programming

- The position of the transport vial can be programmed. Some possible positions are 85, 86, and 87. Position 85 is the standard position for transport vials. The system calculates the necessary transport volume. Make sure before system start that the vial is filled with at least 8000 µl of liquid.
- The liquid levels of the transportation liquids are not updated at system start. To avoid contamination of the air needle, the needle stops inside the transport vial in the up most position.
- If conducting a sample sequence or one sample per row, the autosampler takes the residual volume of the transport liquid into account. If the volume falls below 4000 µl, the needle moves deeper into the transport vial. If the amount of transport liquid reaches 0 µl, the autosampler sends out error message 369 (not enough transport liquid available).
- The liquid levels of the transportation liquid will reset after reprogramming the mode.
- The needle does not move automatically to the next transport vial. If transport liquid is to be taken from another position, you have to change the program settings.

Air needles

Six different lengths of air needles from 50 to 80 mm are available for the autosampler. The needle holder allows you to further adjust the needle height by 6 mm.

Standard air needle

The standard air needle is 62 mm long and can be used for a wide range of deep and shallow vial plates.

When 10 ml sample vials are used, the needle deeply penetrates the sample vial. If this is not filled to more than 60%, the needle can be used in the typical manner. The same applies to deep microtiter plates.

For non-standard settings, use the corresponding needle types.

Legend
① 10 ml sample vials
② 2 ml sample vials

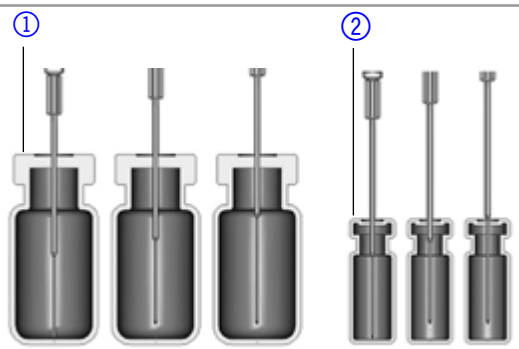


Fig. 40 Standard air needle with 10 and 2 ml sample vial

Note: The PASA™ loop injection principle is not suitable for shallow microtiter plates. The function of the air needle is only ensured when it pierces the closure to a sufficient degree.

Legend
① Deep microtiter plate with closure
② Shallow microtiter plate

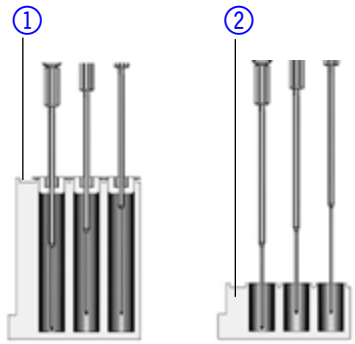


Fig. 41 Standard air needle with microtiter plates

Choosing the correct air needle

To choose the correct air needle, take the following dimensions into consideration.

Fig. 43 H_t = height of sample plate
Fig. 44 D_w = hole depth
Fig. 45 C_d = thickness of closure
Fig. 46 N_h = set needle height
Fig. 47 A_c = distance from air needle tip to closure (min. 2 mm)
Fig. 48 ? = Excess length

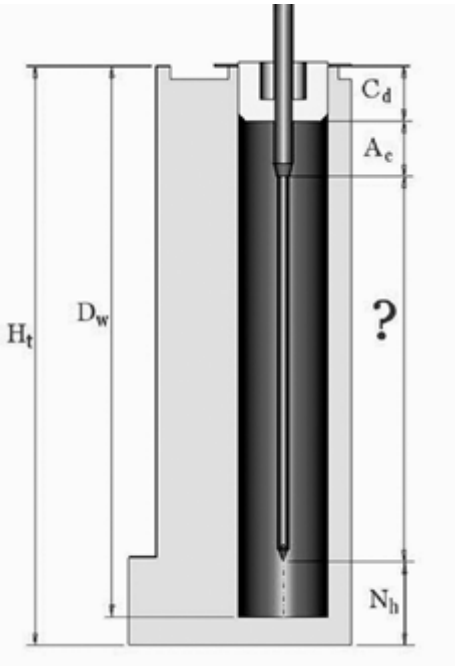


Fig. 42 Calculating the correct air needle

Condition: $H_t - D_w$ = 2 to 6 mm

Excess length of the sample needle:
$H_t - C_d - N_h - A_c = ?$

Choose the correct needle type on the basis of the excess length.

Air needle type	Protrusion length
50 mm, yellow	34-40 mm
56 mm, red	28-34 mm
62 mm, nature (standard needle)	22-28 mm
68 mm, blue	16-22 mm
74 mm, green	10-16 mm
80 mm, black	4-10 mm

Legend

① 10 ml sample vial,50 mm air needle
② 2 ml sample vial,62 mm air needle

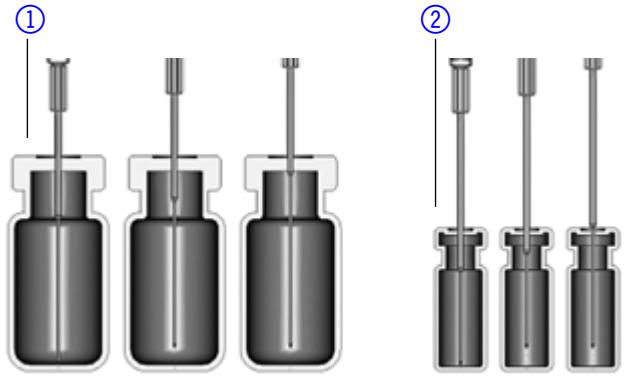


Fig. 49 Selected air needles with sample vials

Legend

① Deep microtiter plate with closure, 56 mm air needle

② Shallow microtiter plate, 80 mm air needle

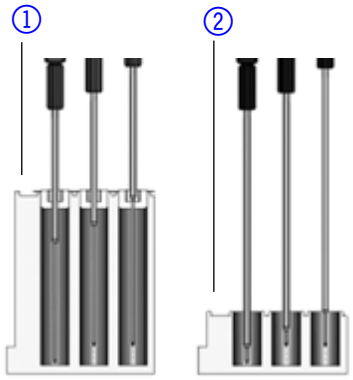


Fig. 50 Selected air needles with microtiter plates

Calculation example for air needle

Starting point:

- autosampler with standard setting for needle height.
- deep microtiter plate with closure

Dimensions:

H_t = 41.4 mm

D_w = 37.8 mm

C_d = 3.8 mm

N_h = 6.0 mm (standard)

A_c = 2.0 mm (minimum)

Condition:

$H_t - D_w$ = between 2 and 6 mm

$H_t - D_w$ = 41.4 mm - 37.8 mm = 3.6 mm. Condition has been met.

$H_t - C_d - N_h - A_c$ = excess length

41,4 mm - 3,8 mm - 6,0 mm - 2,0 mm = 29.6 mm

Air needle type	Protrusion length
56 mm, red	28-34 mm

An air needle length of 56 mm is required.

Handling the sample vials

When handling the sample vials, observe the following:

- Fill the sample vials using a pipette to allow air to escape.
- To prevent the sample from contaminating the air needle, do not fill the sample vials to the very top.
- Do not use sample vials that are unclosed.
- Only use air-tight closure seals to prevent air bubbles from forming and volatile components from evaporating.

- Do not use sample vials with hard closures that the sample needle cannot pierce.

Mixing and thinning

A mix method can be programmed for the autosampler to mix or dilute the sample fluid.

- Configure the mixing routine and syringe speed using the chromatography software.
- A maximum of 15 steps can be programmed for a mix method.

Three types of actions are possible:

1. Add
2. Mix
3. Wait

Add When adding, the defined volume is aspirated from either the sample vial, the vial with Reagent A or Reagent B or flushing fluid and then dispensed into the destination vial.

Note: To prevent carryover, the autosampler removes 125% of the given volume from the corresponding sample vial and uses the additional 25% to flush the tube and needle.

Mix When mixing, the contents of a specific sample vial is mixed by aspirating and dispensing the defined volume n times. If a destination vial has not been defined, mixing is performed in the current sample vial.

Note: When entering the Sample Vials, the Destination Vial is used automatically.

Wait With the Wait command, the system waits until the programmed delay time has elapsed before executing the next line of the program.

Example: Add

The command ADD 100 µl from Reagent A to Destination (100 µl Reagent A to the target vial) triggers the following steps:

1. An air segment of 5 µl is aspirated to separate the flushing solution in the buffer tube from Reagent A.
2. 50 µl of Reagent A are aspirated to flush the tube and needle.
3. Syringe is emptied into the waste container through the drainage tube.
4. 100 µl of Reagent A are aspirated and then dispensed into the destination vial.
5. Tube and needle are flushed with flushing solution.

Example: Mix

In a previous command ADD ... to Destination mixing is performed in the destination vial If this is preceded by an ADD to Sample command, mixing is performed in the sample vial.

The MIX 3 times with 100 µl command triggers the following steps:

1. An air segment of 50 µl is aspirated to separate the flushing solution in the buffer tube from the sample solution to be mixed.
2. Syringe is emptied into the waste container through the drainage tube.
3. 100 µl solution are aspirated and dispensed into the same sample vial.
4. Step 3 is repeated twice. Step.
5. Tube and needle are flushed with flushing solution.

Sample positions in mixing routine

When configuring a mix method, the positions of the sample vials depend on whether the vial plates are to be processed in rows or columns.

Processing in Columns

When column processing is used, the following positions are possible for the Sample, Destination, Reagent A and Reagent B:

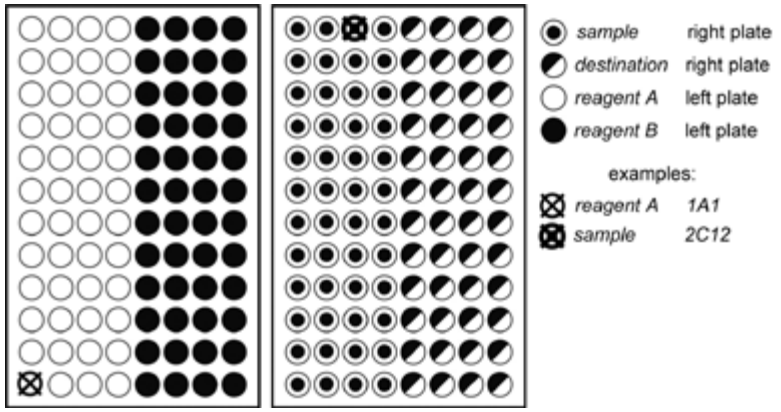


Fig. 51 Sample positions in columns

Processing in rows

When row processing is used,the following positionsfor the Sample, Destination Reagent A and Reagent B are possible:

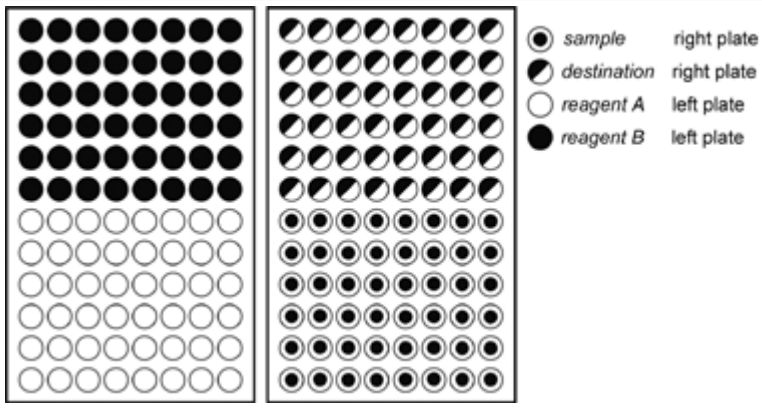


Fig. 52 Sample positions in rows

Parameters for mixing method with 84+3 vial plate

Before using the 84+3 vial plate, it is necessary to change the software settings.

The vial positions for the 84+3 vial plate are as follows:

Position of the first sample:	Vial positions 1-84
Position of the final sample:	Vial positions 1-84
First destination position:	Vial positions 1-84
Transport position:	Vial positions 85-87

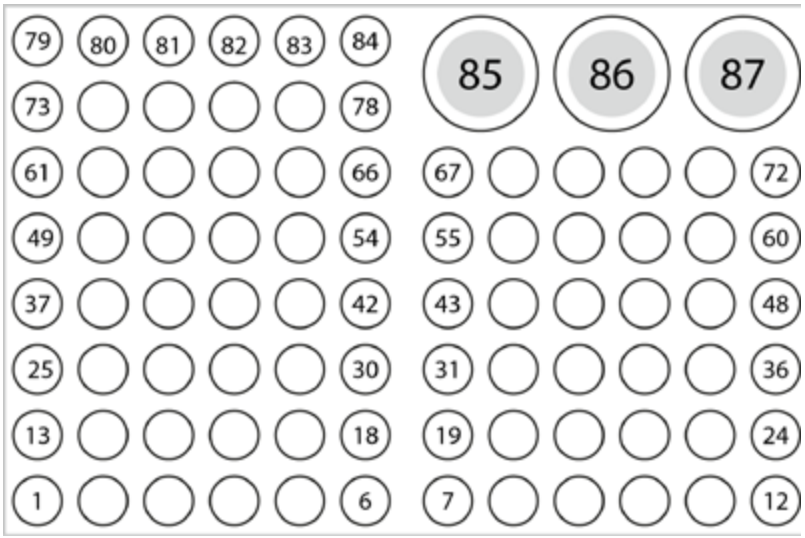


Fig. 53 Reagent/Transport position on 84+3 vial plate

You need to define the first and the final sample to enter a sample range. The same range can be used for the destination vials. The reagent positions are always positions 85, 86, 87.

Details on programming the 84+3 mixing method

- The position of the reagent vial can be programmed. Some possible positions are 85, 86, and 87. The standard transport positions are 86 for Reagent A and 87 for Reagent B. The system calculates the necessary reagent volume. Before system start, make sure that the vials are filled with at least 8000 µl of liquid.
- The liquid levels of the reagent liquids are not updated at system start. To avoid contamination of the air needle, the needle stops inside the reagent vial in the up most position.
- If conducting a sample sequence or one sample per row, the autosampler takes the residual volume of the reagent liquid into account. If the volume falls below 4000 µl, the needle moves deeper into the reagent vial. If the amount of reagent liquid reaches 0 µl, the autosampler sends out error message 370 (not enough reagent liquid available).
- The liquid levels of the reagent liquid will reset after reprogramming the mode.
- Reagent will only be taken from preprogrammed positions. The needle does not move automatically to the next reagent vial. You have to change the settings when reagent is to be taken from one of the other vials.

Connecting capillary and tubing

Connecting the valve

Legend

① Connection tube buffer to syringe
② Sample loop
③ Stainless steel capillary to column
④ Stainless steel capillary to pump
⑤ Connection plastic capillary to injection needle
⑥ Cleanout ILD™ valve (analytical Autosampler)

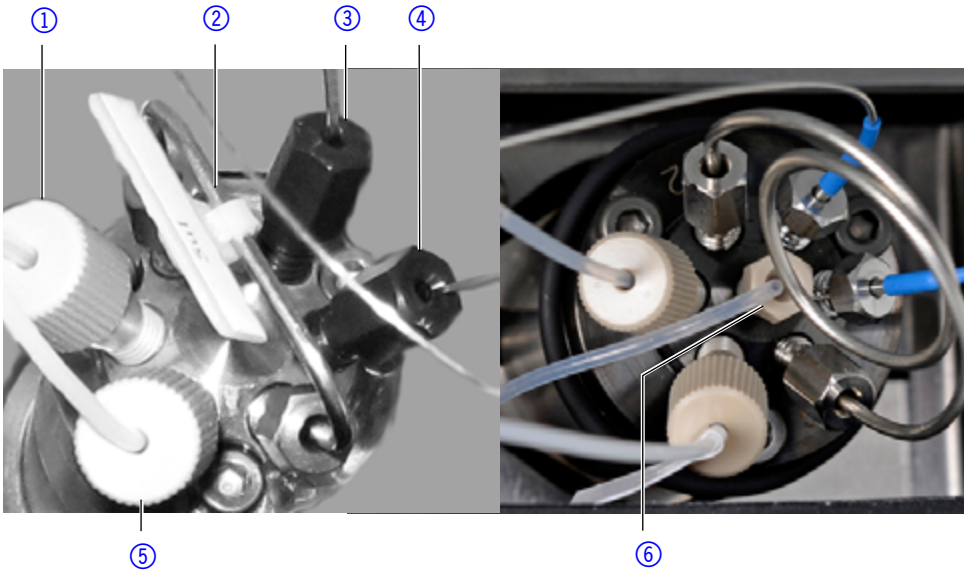


Fig. 54 Left preparative injection valve, right analytical injection valve with central port

Connecting the syringe

Legend

① Tubing connection for transport liquid (not available for all models)
② Connection buffer tube
③ Connection flushing solution

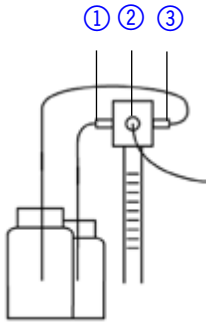


Fig. 55 Syringe connections

Tubing guide for flushing solution

Note: Use the tubing guide in the collecting container for the flushing solution in order to not hinder horizontal movement of the needle.

Legend

① Tubing guide for flushing solution
② Collecting container with hole for tube guide (flushing fluid)

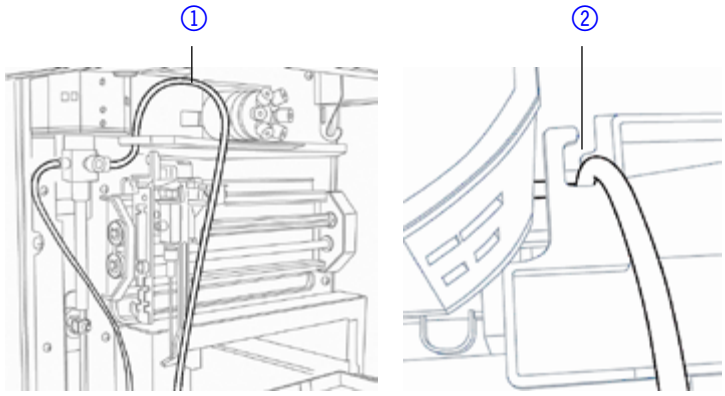


Fig. 56 Tubing guide for flushing solution tube

Connecting the drainage tubing

The waste drainage removes all flushing fluids and non-injected sample solutions.

Connect the drainage hose at the left side of the device and insert the end of the hose into a waste bottle on the floor. Make sure that the drainage tubing is not kinked so that the liquid can drain.

⚠ DANGER

Life-threatening injuries

Health danger if contacting with toxic vapors.

➔ Attach the waste tube according to the manual.

➔ Ensure that the room is always well ventilated.

⚠ DANGER

Life-threatening injuries

Health danger if getting in contact with toxic vapors, toxic material or biological hazardous substances, which can escape through the overflow hole.

➔ Avoid overflowing the waste container.

➔ Clean the overflow hole after overflowing. Observe the safety regulations applicable to the materials.

Prerequisites

- The front cover has been removed.

Process	Figure
1. Mount the drainage hose at the front of the ① device. 2. Place the waste bottle below the device. 3. Connect the drainage hose with the waste bottle.	 Fig. 57 Leakagetray withdrainage tube

Next Steps Attach the front cover.

Connecting the autosampler with other devices

Controlling the autosampler with chromatography software

The autosampler is controlled directly with software, e. g. OpenLAB® and ClarityChrom® by KNAUER or Thermo Scientific™ Dionex™ Chromeleon™.

Connect the autosampler to the network, using the LAN connection at the rear panel of the device.

Checking and configuring the parameters of the autosampler

You can configure the autosampler parameters using the chromatography software, e.g. ClarityChrom®:

1. Select the autosampler in LAN.
2. Set the syringe volume to either 250 µl (default) or 500 µl.
3. Enter the serial number of the autosampler.
4. Choose the cooling option if your autosampler offers temperature control.

Configuration window of ClarityChrom®

Legend

① Serial number
② Device detection in local network
③ Manual search for device in network
④ Volume of sample loop
⑤ Volume of syringe

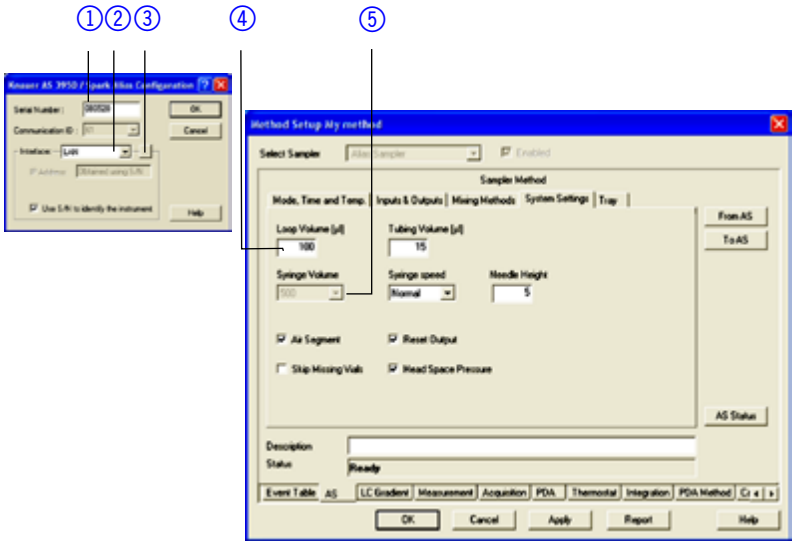


Fig. 58 Autosampler configuration with ClarityChrom®

Autosampler device software

A software CD with Autosampler AS 6.1L Service Manager is included with the autosampler. Using this software, you can check and control the device. These settings can also be made with the chromatography software, so that it is not absolutely necessary to install the Service Manager.

Flushing the system

The system should be flushed before the column is connected. The flushing of the system can be controlled using the chromatography software or Autosampler 6.1L Service Manager.

Note: KNAUER recommends using a mixture of water and isopropanol (80 %/20 %) or the mobile phase as the flushing solution.
The following steps are explained for when the Service Manager is used.

System flushing with Autosampler 6.1L Service manager

1. Install Autosampler 6.1L Service manager.
2. Fill the flushing solution into a solvent bottle and degas it using helium or an ultrasonic bath.
3. Slide the tube for the washing liquid into the solvent bottle.
4. Select the Alias → **Direct Control** menu.

5. In the Syringe field, click **End**. One syringe volume is sucked into the syringe through the wash solution tube.
6. In the Syringe field, click **Home**. The syringe content is emptied into the drainage tube.
7. Repeat step 5 and 6 until the syringe and the wash solution tube have been filled completely.
8. In the Initial wash field, click **Start**. All tubes that are connected to the syringe are flushed.
9. In the Initial wash field, click **Stop**.
10. Click **Close** to exit the Direct Control window.

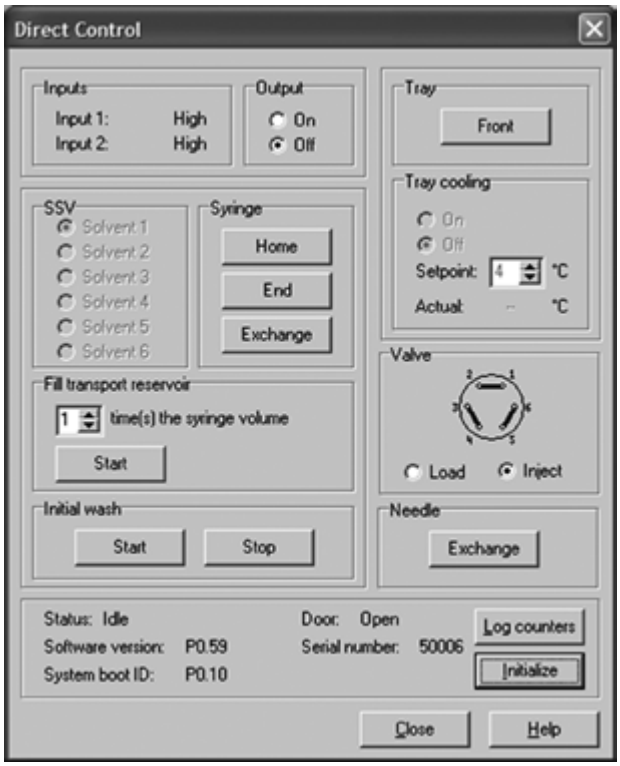


Fig. 59 System flushing with the Service manager software

I/O connection

By default, the autosampler has an I/O connection that supports TTL inputs (low-active) and a closed-contact output. Devices without LAN connection that require a trigger signal for the injection can be connected by means of the I/O connection.

The TTL inputs allow you to control the autosampler using other devices and are defined using the instrument methods of the chromatography software.

Note: Only connect the autosampler to devices that fulfill the required safety standards!

Defining the TTL inputs

- Next Injection Input: The injection sequence is started. After ending the injection sequence, the autosampler waits for the next start signal.
- Freeze Input: The analysis time is paused. The autosampler runs the configured program up to the filling of the sample loop. The injection is not performed until the input is deactivated.

- Stop Input: Immediately halts the analysis.

Defining the closed-contact output

- Inject Marker: The closed-contact output is activated when the injection valve switches from LOAD to INJECT.
- Alarm: The closed-contact output is activated when there is an autosampler fault.
- Auxiliary: -

Configuration of I/O connection (9 pins)

Explanation	Cable color
1. Output, start injection	Red in three-conductor cable
2. Output, start injection	Black in three-conductor cable
3. Input 1, programmable input for stopping injections (low-active)	Red in four-conductor cable
4. Input 2, programmable input for stopping injections (low-active)	Black in four-conductor cable
5. Not occupied.	-
6. Output	Brown in three-conductor cable.
7. Not occupied.	-
8. Ground, for inputs 1 and 2	Orange in four-conductor cable
9. Ground, for inputs 1 and 2	Brown in four-conductor cable

Device test

The reproducibility of the sample volume is a critical factor for maintaining high-quality analysis results.

- Test the autosampler features with the chromatography software on a regular basis.
- If the device test determines that the autosampler does not fulfill the requirements, mark the device as defective and do not continue using it.
- Do not re-use the autosampler until it has been repaired and/or serviced.

Test intervals

Run the device test at the following time intervals:

- Average use of 1-5 days/week: Device test every 6 months

- Average use of more than 5 days/week or 24 hours/day: Device test every 3 months
- Operation with buffer solutions or other salt solutions: Device test every 3 months

Devices and components for the test

- Autosampler AS 6.1L, 250 µl syringe, 1240 bar version with 10 µl sample loop or 700 bar version with 100 µl sample loop
- pump, 1 ml/min flow rate
- UV detector (with a data rate of 50 Hz if possible, otherwise 10 Hz, flow cell: 10 mm path length)
- Chromatography software
- Eluent: 90 % water, 10 % methanol (HPLC quality)
- Test solution (sample):
  - a: 50 ppm uracil dissolved in water (HPLC quality)
  - b: 250 ppm uracil dissolved in water (HPLC quality)
- Flushing solution:
  - 80 % water, 20 % isopropanol (HPLC quality)
  - Alternative: 80 % water, 20 % methanol (HPLC quality)
- Restriction capillary:
  - Inner diameter 0.25 mm
  - Length 200 cm

Note: Degas the eluent to prevent malfunctions caused by the presence of air bubbles.

1. Reproducibility of sample volume

The variation coefficient must not exceed 0.5 %.

- Configure the pump, UV detector and autosampler using the chromatography software
- Inject 10 µl of test solution a (50 ppm uracil, dissolved in water)

Standard Setting of Autosampler

- Loop volume: 10 µl for the 1240 bar version, 100 µl for the 700 bar version
- Tubing volume: 15 µl
- Syringe volume: 250 µl

Method parameters of pump

- Flow: 1 ml/min
- Time: 1 min

Method parameters of autosampler

- Injection method: Partial loopfill
- Syringe speed: normal
- Flush volume: 30 µl, (40 µl for microtiter plates)
- Needle wash: active 2 times
- Air segment: yes
- Headspace pressure: yes
- Injections/vial: 9 (7 for microtiter plates)
- Inj. volume: 2 µl
- Vial position: 1A1. Start the single run with

Method parameters of the UV detector

- Wavelength: 254 nm
- Sampling rate: 50 Hz if possible, else 10 Hz
- Time: 0.5 min

Configuring repeat runs autosampler

- Injections/vial: 9x (7x for microtiter plates)
- Inj. volume: 1 µl

Starting repeat runs

1. Put a vial with at least 500 µl test solution on position 1A1 of the sample plate.
2. Start the repeat runs.

Analyzing the individual chromatograms

1. Calculate the average of the measuring values of the Peak areas.

2. Calculate the variation coefficient VK_1 .
3. Enter the results into the Test Report form.

Formula for determining the arithmetic mean

$$\overline{Peakareas} = \frac{\sum_{i=1}^{n} Peakareas_i}{n}$$

- Formula for determining the standard deviation (i = 1-9):

$$\sigma_{n-1} = \sqrt{\frac{\sum (Peakarea_i - \overline{Peakareas})^2}{n-1}}$$

- Formula for determining the variation coefficient:

$$VK_1 [\%] = \frac{\sigma_{n-1}}{peakarea} \times 100$$

2. sample carryover

The percentage of sample carryover must not exceed 0.3 %.

- Alternately inject 10 µl of test solution b (250 ppm uracil, dissolved in water) followed by eluent.
- Position of sample vial: 1A1
- Position of eluent: 1A2
- Injection volume: 1 µl

Creating a sequence with 6 lines

- Test solution: Position 1A1
- Eluent: Position 1A2
- Injection volume: 1 µl
- Repeats: 1

Note: For microtiter plates, select 6 consecutive positions that are alternately to be filled with test solution and eluent.

Analyzing the individual chromatograms

1. Calculate the average of the measuring values of the peak areas.
2. Put the average of the eluent injection in relation to the average of the test solution injection.
3. Enter the results into the Test report form.

Formula for calculating sample carryover

$$PV\,[\%] = \frac{\sum_i \frac{Peakarea_{i\,Fließmittel}}{3}}{\sum_i \frac{Peakarea_{i\,Testlösung}}{3}} \times 100$$

3. Linearity

To determine the linearity, the correlation coefficient of the regression lines is determined from the measured values for the peak areas and injection volume.

Note: The procedure is only possible if a 100 µl sample loop is installed. The correlation coefficient must not exceed 0.998 %.

- Inject 10, 20, 30, 40 and 50 µl of test solution b (250 ppm uracil, dissolved in water) respectively.
- Position of sample vial: 1A1
- Injection volume: 10 µl, 20 µl, 30 µl, 40 µl, 50 µl
- Repetition: 3

Note: Fill consecutive positions on the microtiter plates with test solution.

Analyzing the individual chromatograms

1. Calculate the correlation coefficient r of the regression lines from the measured values for the Peak areas and the injection volume.
2. Enter the results into the Test Report form.

Formula for determining the correlation coefficient

$$r = \frac{\sum x_i y_i - n\bar{x}\bar{y}}{\sqrt{\left(\sum x_i^2 - n\bar{x}^2\right)\left(\sum y_i^2 - n\bar{y}^2\right)}}$$

y_i Y-value of measured value i (injection volume)

x_i X-value of measured value i (peak surface)

$\bar{y}$ y = arithmetic mean of Y across all n measured values

$\bar{x}$ x = arithmetic mean of X across all n measured values

n = number of measured value pairs

4. Mixture test

To create a mixing method, follow the instructions in the manual of the chromatography software.

- The test solution variation coefficient VK_2 must not exceed 0.5 %.
- The dilution variation coefficient VK_2 must not exceed ≤ 0.5 %.
- The dilution factor F10 has to be within the range of $9.85 < x < 10.25$.

- Test solution: 10 µl (50 ppm uracil, dissolved in water)
- For injecting the dilution, go to Mix methods and create a mixing method in which 40 µl of the test solution is mixed with 360 µl eluent.
  - Dilution: 10 µl, 5 ppm uracil, dissolved in deionized water
- Inject 10 µl test solution and und 10 µl dilution three times each.

Creating a sequence with 2 lines

- Injection volume: 1 µl
- Repeats: 3

Positioning vials for dilution

- To position the vials on the sample plates, select the Columns option in the chromatography software.
- Put a vial with test solution (Sample) at position 2A1 of the vial plate.
- Put an empty vial (Destination) at position 2A5 of the vial plate.
- Put a vial with eluent (Reagent A) at position 1A1 of the vial plate.

Note: Note the plate assignments for the vials (Sample, Reagent A, Reagent B, Destination).

Analyzing the individual chromatograms

1. Calculate the mean of the measuring values of the Peak areas for the test solution and the dilution.
2. Calculate the variation coefficient VK_2 .
3. Calculate the variation coefficient VK_3 .
4. Calculate the dilution factor F10 from the ratio of the mean of the Peak areas of the test solution and the dilution.
5. Enter the results into the Test Report form.

Archiving

- Enter all test results into the Test Report form.
- Enter the serial number, date of the test, date of the next test and name of the tester.
- File the Test Report form in the device logbook.

Test report

Module	Autosampler	Version
Autosampler AS 6.1L	Please enter serial number:	Cool/heat version? Yes ☐ No ☐ Prep version? Yes ☐ No ☐ Bio version? Yes ☐ No ☐

Nr.	Test	Setting	Specification	Result
1	Reproducibility	▪ Inject 10 µl test solution nine times. ▪ Inject 10 µl test solution seven times.	▪ VK1 ≤ 0,5 %	
2	Carryover	Inject 10 µl test solution and 10 µl eluent three times.	PV ≤ 0,3 %	
3	linearity	Inject 10, 20, 30, 40 and 50 µl test solution three times each.	r ≥ 0,998	
4	Mixture test	Inject 10 µl test solution and 10 µl of the dilution created by the autosampler, three times each.	VK_2 ≤ 0,5 % VK_3 ≤ 0,5 % F10: 9,85 < x < 10,25	

Date:

Date of the next device test:

Functionality tests

Note: Standard procedure for IQ and OQ can be handled differently in individual cases for devices.

Operation Qualification (OQ)

The OQ is a detailed operating test based on the standardized KNAUER OQ documents. The Operation Qualification is a standardized KNAUER document and is free of charge. It is not included with the instrument. If necessary, contact technical customer service.

The OQ protocol includes the following:

- definition of customer requirements and acceptance terms
- documentation on device specifications
- device functionality check at installation site

Test Intervals

To make sure that the device operates within the specified range, you should test the device using the Operation Qualification at following intervals: The test intervals are determined by the use of the device.

Execution

The OQ can be carried out either by the Technical Support of KNAUER or from a provider authorized by KNAUER.

Maintenance and care

Maintenance contract

The following maintenance work on the device may only be performed by KNAUER or a company authorized by KNAUER and is covered by a separate maintenance contract:

- Opening the module
- Removing the hood or the side panels.

⚠ DANGER

Electric shock

Danger of electric shock from voltage-carrying parts inside the device. The housing serves as a protective cover against voltages inside the device.

➔ Switch the device off before opening the device.

➔ Pull the power plug.

Which type of maintenance tasks may users perform on the device?

Users may perform the following maintenance tasks themselves:

- Regularly check for clogged capillaries – test back pressure without column.
- Exchanging the fuses

- Exchanging the air and sample needle
- Exchanging the injection valve
- Exchanging the sample loop
- Exchanging the rotor seals
- Exchanging the capillary and tubing

NOTICE

Electronic defect

Performing maintenance tasks on a switched on device can cause damage to the device.

➔ Switch off the device

➔ Pull the power plug.

Note: If leaks occur on the capillary screw fittings after maintenance and proper assembly, do not tighten them further, but instead replace them with new connection capillaries.

System flushing

Process
1. Connect the autosampler to the power supply. 2. Establish a connection to the PC. 3. Start Autosampler 6.1L Service Manager. 4. Select the Alias →**Direct Control** menu. 5. Click **Initialize** to check whether the valve is correctly positioned at the Inject position. 6. In the Initial Wash field, click **Start** to flush the system. 7. In the Initial Wash field, click **Stop** to stop flushing the system.

Exchanging the fuses

Note: If the fuses blow repeatedly, consult with KNAUER Technical Support for replacements and help in identifying the cause.

Process
1. Switch off the autosampler and remove the power plug to completely disconnect the device from the power supply. 2. Remove the fuses from the fuse box at the rear of the device. 3. Insert new fuses (2 x 2.5 A). 4. Plug in the power plug.

Exchanging the injection valve and rotor seal

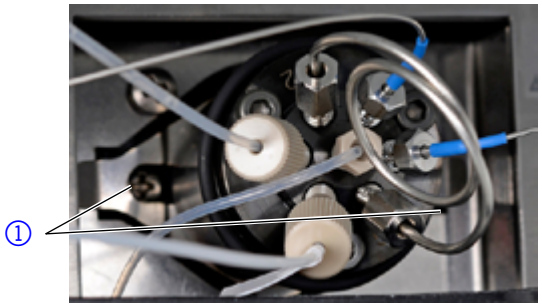
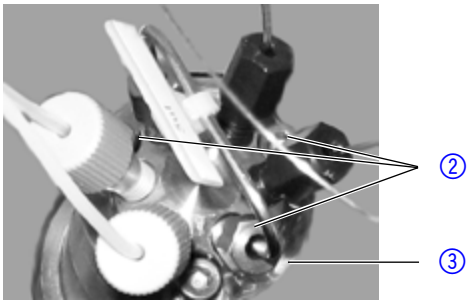
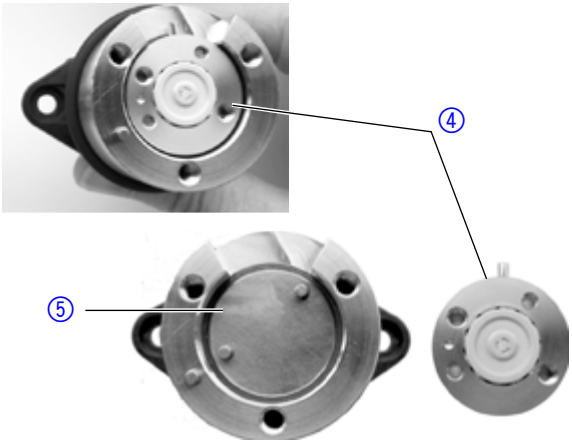
- Regularly clean the rotor seal of the injection valve.
- Regularly replace the rotor seal (approx. every three years).

Removing the injection valve and rotor seal

Prerequisites

- Remove the front panel of the autosampler.
- Remove the capillary connections, except the sample loop, from the valve.
- During removal, consecutively loosen all screws by half a turn respectively, until they can be removed.

Note: Do not remove the screw in the cover plate hole diagonal to the valve.

Process	Figure
1. With a screwdriver, remove screw ① on both sides of the analytical/preparative injection valve-housing. 2. Unscrewing the injection valve.	 Fig. 60 Unscrewing the injection valve (figure shows analytical valve)
3. With a hexagon wrench key, remove the screws ② from the valve stator block 2. 4. Carefully remove the stator ③ .	 Fig. 61 Unscrewing the stator (figure shows preparative valve)
5. Remove the rotor seal ④ from the rotor ⑤ . 6. Clean or exchange the rotor seal.	 Fig. 62 Remove the rotor seal (size and model of the rotor seal can vary, depending on the equipment of the autosampler)

Installing the injection valve

- During installation, hold the injection valve so that the bore hole for connecting the capillary to the pump (port 6) is facing upward.
- Alternately tighten all screws by half a turn, until all screws have been fully tightened.

Legend

① Connector port 1
② Connection from capillary to pump

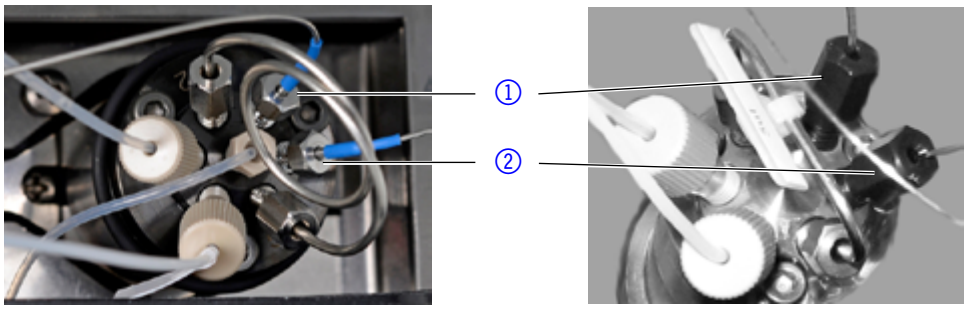


Fig. 63 Installing the injection valve

Process
1. Insert the rotor seal. 2. Place the stator onto the rotor and use a hexagon wrench key to tighten the screws. 3. Insert the injection valve and use a screwdriver to tighten the screw on both sides of the valve housing. 4. Connect the capillary again.

Replacing the sample loop

The 1240 bar version is equipped with a 10 ml sample loop, the 700 bar version with a 100 ml sample loop.

- When assembling a sample loop with a different injection volume, make sure to use the correct combination of syringe and capillaries and configure the controller software appropriately.
- Always connect the sample loop to ports 2 and 5 of the injection valve.
- Calculate the maximum injection volume according to the following formula:
  - full loop filling:
    Maximum injection volume = 3 times sample volume for loops with 100 µl, 2 times loop volume for loops with 100-500 µl, 1.5 times loop volume for loops above 500 µl
  - partial loop filling:
    Maximum injection volume = 50 % loop volume
  - microliter-pickup:
    No maximum injection volume

Exchanging the sample needle

- When using sample plates with 12, 48 or 108 sample vials, make sure that the needle height setting is >2 mm to prevent the needle from contacting the bottom of the sample vial.
- Tighten the screw fitting until it is finger-tight to prevent the plastic capillary from becoming blocked.

Legend
① Screw fitting
② Plastic capillary
③ Nut
④ Sample needle

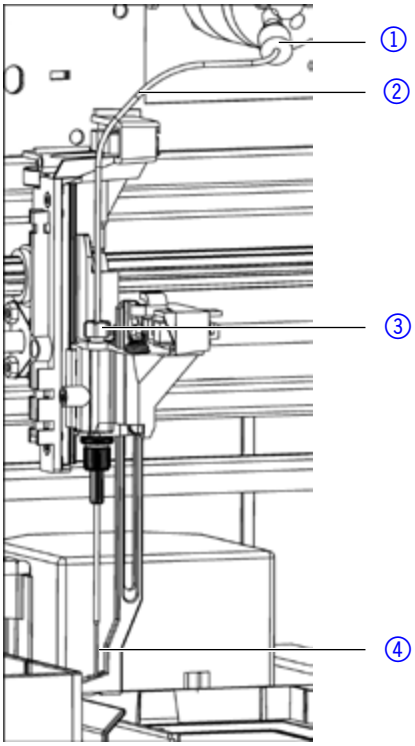


Fig. 64 Exchanging the sample needle

Procedure

1. Start Autosampler 6.1L Service Manager.
2. Select the Alias →**Direct Control** menu.
3. In the Needle field, click **Exchange**. The needle moves to the replacement position.
4. Loosen the union nut ③ .
5. Loosen the screw fitting ① of the plastic capillary ② on the injection valve.
6. Remove the sample needle ④ with the plastic capillary.
7. Install a new sample needle unit. Make sure that the air seal fully surrounds the sample needle.
8. Fasten the sample needle with the union nut.
9. Fasten the plastic capillary using the screw fitting on the injection valve.
10. In the Direct Control window, click **Initialize**. The needle moves to the initial position.
11. In the Initial Wash field, click **Start** to flush the system.
12. In the Initial Wash field, click **Stop** to stop flushing the system.
13. Click **Close** to exit the Direct Control window.
14. Select the Alias →**Adjustments** menu.
15. On the Needle-Tray tab, update the settings for the sample plates.

Exchanging the air needle

- When exchanging the air needle, make sure that the thread of the new height adjustment screw is flush with the lower edge of the retaining nut.
- Make sure that the sealing ring is located in the retaining nut.

Legend

① Nut
② Retaining nut
③ Height adjustment screw
④ Air needle
⑤ Sample needle

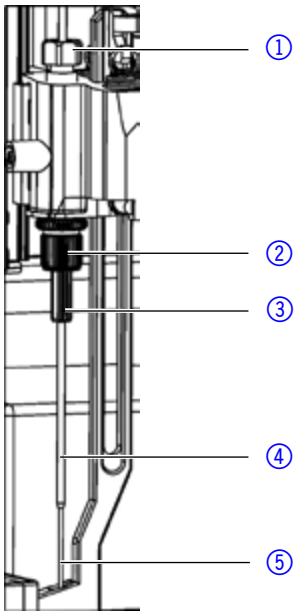


Fig. 65 Exhanging the air needle

Procedure

1. Start Autosampler 6.1L Service Manager.
2. Select the Alias →**Direct Control** menu.
3. In the Needle field, click **Exchange**. The needle moves to the replacement position.
4. Loosen the union nut ① .
5. Loosen the screw fitting of the plastic capillary on the injection valve.
6. Remove the sample needle ⑤ with the plastic capillary.
7. Loosen the retaining nut ② and pull it downwards together with the air needle ④ .
8. Unscrew the retaining nut from the height adjustment screw ③ .
9. Screw a new air needle with a new height adjustment screw into the retaining nut.
10. Screw in the retaining nut.
11. Insert the sample needle and fasten with the union nut.
12. Fasten the plastic capillary using the screw fitting on the injection valve.
13. In the Direct Control window, click I**nitialize**. The needle moves to the initial position.
14. In the Initial Wash field, click **Start** to flush the system.
15. In the Initial Wash field, click **Stop** to stop flushing the system.
16. Click Close to exit the Direct Control window.
17. Select the Alias → **Adjustments** menu.
18. On the Needle-Tray tab, update the settings for the sample plates.

Change of the syringe

By standard, the autosampler is equipped with a 250 µl syringe.

Use isopropanol as flushing solution to remove air bubbles from the new syringe.

Legend

① Syringe valve
② Syringe
③ Syringe drive
④ Syringe plunger

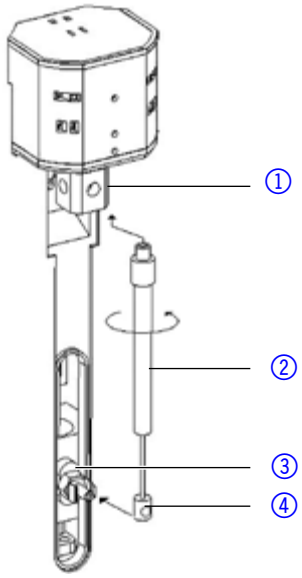


Fig. 66 Procedure

Procedure

1. Start Autosampler 6.1L Service manager.
2. Select the Alias →**Direct Control** menu.
3. In the Syringe field, click **Exchange**. The syringe plunger is lowered.
4. Unscrew the syringe ② by rotating it counterclockwise; leave the adapter in the syringe valve ① .
5. Remove the syringe plunger ④ from the syringe drive ③ .
6. Fill new syringe with flushing solution.
7. Insert the syringe plunger into the syringe drive.
8. Tighten the syringe in the syringe valve by rotating it clockwise.
9. In the Syringe field, click **Home**. The syringe content is emptied into the drainage tube.
10. If there is still air in the syringe, click **End** in the Syringe field. One syringe volume is aspirated into the syringe through the flushing solution tube.
11. In the Syringe field, click **Home**. The syringe content is emptied into the drainage tube.
12. Slightly tap the body of the syringe if it still contains air. If necessary, repeat step 10 and 11. Repeat the step.
13. In the Initial Wash field, click **Start** to flush the system.
14. In the Initial Wash field, click **Stop** to stop flushing the system.
15. Click **Close** to exit the Direct Control window.

Exchanging the syringe plunger or plunger tip

1. Start Autosampler 6.1L Service Manager.
2. Select the Alias →**Direct Control** menu.
3. In the Syringe field, click **Exchange**. The syringe plunger is lowered.
4. Remove the syringe (see above).
5. Pull the syringe plunger out of the glass cylinder of the syringe.
6. Use a pair of tweezers to remove the plunger tip.

7. Wet the new plunger tip with isopropanol.
8. Mount the new plunger tip onto the syringe plunger.
9. Push the syringe plunger into the glass cylinder of the syringe.
10. Install the syringe (see above.)
11. In the Syringe field, click **Home**. The syringe content is emptied into the drainage tube.

Exchanging the syringe valve

The syringe valve has four connections, one of them remains unused.

- Hand-tighten all fittings on the connections to the syringe valve.
- To exchange the valve, set it to the Waste position, because the hexagon socket screws are only accessible if the valve is in this position.

Legend

① Upper hexagon socket screw
② Lower hexagon socket screw
③ Connection flushing solution tube (hidden)
④ Connection buffer tube
⑤ Connection syringe
⑥ Unused connection

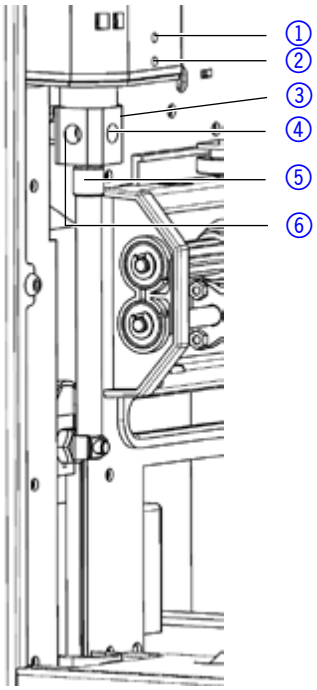


Fig. 67 Exchanging the syringe valve

Procedure

1. Start Autosampler 6.1L Service Manager.
2. Select the Alias →**Direct Control** menu.
3. In the Syringe field, click **Exchange**. The syringe plunger is lowered.
4. Loosen the lower hexagon socket screw by 2 turns.
5. Loosen the upper hexagon socket screw by 2 turns.
6. Pull out the upper part of the syringe.
7. Remove the syringe.
8. Exchange the syringe valve.
9. Insert a new syringe.
10. Tighten the hexagon socket screws.

Cleaning and caring for the device

All smooth surfaces of the device can be cleaned with a mild, commercially available cleaning solution, or with isopropanol.

- Clean collecting container and vial plates with a soft cloth.

- To remove deposits, flush the drainage tube regularly using solvent.

Putting the instrument out of operation

The device is designed for the usage of different solvents. Solvent residue can damage the device or irritate the skin. That is why we recommend to flush components of the flow path in the autosampler before maintenance.

Prerequisites The device has been flushed.

Process
1. Switch off the autosampler. 2. Pull the plug from the socket. 3. Pull the plug from the device.

Next Steps Conduct any allowed maintenance tasks. For storage, select a location according to the requirements, which are listed in the according chapter of this user manual.

Troubleshooting

Device errors

One possible cause of device errors is a malfunctioning valve.

Checking the valve

Remove the valve and check all parts for wear and contamination. After the problem has been eliminated and the valve reinstalled, perform the following steps:

Procedure

1. Select the Alias →**Direct Control** menu.
2. In the Direct Control window, click **Initialize**. The needle moves to the initial position.
3. In the Initial Wash field, click **Start** to flush the system.
4. In the Initial Wash field, click **Stop** to stop flushing the system.
5. Click **Close** to exit the Direct Control window.

LAN

Software faults can occur due to flawed communications between the devices or incorrect installation of the software.

Procedure

1. Check the cable connections.
2. Start Autosampler 6.1L Service Manager.
3. Select the Alias →**Direct Control menu**.
4. In the Direct Control window, click **Initialize**.

Go through the following steps, in case no connection between the computer and the devices can be established. Check after each step if the problem is solved. If the problem cannot be located, call the Technical Support.

Check the status of the LAN connection in the Windows taskbar:

-  Connected
-  Connection not established

If no connection was established, test the following:

- Is the router switched on?
- Is the patch cable connected correctly to the router and the computer?

5. Check the router settings:

- Is the router set to DCHP server?

NOTICE

Device defect

Intruding liquids can cause damage to the device.

➔ Place solvent bottles next to the device or in a solvent tray.

➔ Moisten the cleaning cloth only slightly.

- Is the IP address range sufficient for all the connected devices?

6. Check all connections:

- Are the patch cable connected to the LAN ports and not the WAN port?
- Are all cable connections between devices and router correct?
- Are the cables plugged in tightly?

7. If the router is integrated into a company network, pull out the patch cable from the WAN port.

- Can the devices communicate with the computer, even though the router is disconnected from the company network?

8. Turn off all devices, router, and computer. Erst den Router anschalten und warten bis dieser seinen Selbsttest erfolgreich durchgeführt hat. Firstly, turn on the router and secondly turn on the devices and the computer.

- Has this been successful?

9. Replace the patch cable to the device with that no connection could be established.

- Has this been successful?

10. Make sure that the IP port of the device matches the port in the chromatography software.

Analytical errors

Possible causes:

- Wear due to errors in the injection and method settings.
- Unsuitable combination of sample loop, buffer tube and syringe.
- External effects such as temperature, and light-sensitive samples being exposed to light.

Solutions:

- Check whether the application has run previously without errors and that no changes have been made to the analytical system.
- Determine whether the fault is caused by the autosampler or other devices in the system.

If the required degree of reproducibility is not achieved, check the following possible sources of error and instigate steps to eliminate them:

Cause of fault	Elimination
Air in liquid path	Initialize the autosampler AS 6.1L.
Leaking syringe	If the syringe is leaking at the top, check whether it has been installed correctly. If the syringe is leaking at the bottom, exchange the syringe plunger.
Leaking syringe valve	Check valve and exchange if required.
Rotor seal worn	Exchange the rotor seal and check the stator block of the valve.
Dead volume in capillary connections	Install new fittings onto capillary connections.

If an empty sample run returns an excessively large peak, check the following possible causes of error and instigate steps to eliminate them:

Cause of fault	Elimination
Solubility problems	Either modify sample or accept carryover.
Interaction between the empty sample and the hardware	Check hardware: ▪ Flush needle (inside and outside) or install a different needle type (steel, PEEK or with glass coating). ▪ Valve: Exchange rotor seal (other material). ▪ Capillaries and tubing: Use other connections between the autosampler and the columns (steel, PEEK) or other flushing solutions.
Empty sample contaminated	Use new empty sample.
Cause unknown.	Attempt to solve problem by using different solvents and eluents.

If no injection is performed:

Cause of fault	Elimination
Liquid path blocked	1. Disconnect the plastic capillary of the needle from the injection valve. 2. Start system flushing. 3. If solvent escapes at the injection valve connection to the needle, check the needle. 4. If no solvent escapes at the injection valve connection to the needle, disconnect the buffer tube from the injection valve. 5. Start system flushing. 6. If solvent flows out at the open end of the buffer tube, check the rotor seal. 7. If no solvent flows out of the open end of the buffer tube, disconnect the buffer tube from the syringe valve. 8. Start system flushing. 9. If solvent flows out of syringe valve, check the buffer tube. 10. If no solvent flows out of the syringe valve, check whether the connections of the liquid path have been tightened too much.
Leaking valve	1. Unscrew the plastic capillary leading to the needle from the injection valve. 2. Disconnect the plastic capillary leading to the syringe from the injection valve. 3. Connect the pump to the injection valve. 4. Close the connection to the column at the injection valve. 5. Start the pump at a low flow rate. 6. Check the connections to the syringe and to the needle at the injection valve to ensure that they are tight. 7. If liquid escapes there, check the rotor seal. 8. If no liquid escapes there, check the system with a manual valve.

System messages in OpenLAB®

The various system messages of the chromatography software OpenLAB®from KNAUER are explained below. The system messages are sorted alphabetically.

System message	Explanation
Autosampler is in run mode.	▪ Quit the control software and restart. ▪ Switch the device off and on.
Autosampler is not responding. Please check communication settings and ensure the device is online.	▪ Switch the device off and on. Check the network settings. ▪ Inform the Technical Support of the manufacturer in case the system message repeats itself.
Cannot run autosampler.	▪ Switch the device off and on. Check the network settings. ▪ Inform the Technical Support of the manufacturer in case the system message repeats itself.
Cannot set destination vial to (number).	Check parameters in control software and correct entry.
Cannot set first transport vial to to (number).	Check parameters in control software and correct entry.
Cannot set last transport vial to (number).	Check parameters in control software and correct entry.
Cannot stop autosampler.	▪ Check the network settings. ▪ Inform the Technical Support of the manufacturer in case the system message repeats itself.
Communication port for autosampler was not initialized. Please check the configuration settings.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Configuration settings do not match with the device. Run cannot start.	Check configuration and settings.
Destination position not reached.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Deviation of more than+/- 2 mm towards home.	▪ Look for visible obstructions in area of vial plate. ▪ Check the belt tension of the vial plate.
Dispenser error.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.

System message	Explanation
Electronics error.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
EEPROM error in adjustments.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
EEPROM error in log counter.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
EEPROM error in settings.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
EEPROM write error.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error 369	Not enough transport liquid in store. Refill transport liquid.
Error 370	Not enough reagent in store. Refill reagent.
Error by setting Mix&Dilute vials.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error occurred during initialization, the Autosampler AS6 cannot start.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error resetting output.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error running user defines program.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error setting injection mode.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error setting needle height.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.

System message	Explanation
Error setting injection mode.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error setting syringe speed.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error setting the analysis time.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error setting the auxiliaries.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error setting the flush volume.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error setting the injection volume.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error setting the loop volume.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error setting the prep. mode.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error setting the syringe volume.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error setting timed events.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error setting the tray configuration.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error setting the tray temperature.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.

System message	Explanation
Error setting the vial number.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error setting tubing volume.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Error setting wash volume.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Flush volume error.	Check parameters in control software and correct entry.
Home sensor activated when not expected.	▪ Check parameters in control software and correct entry. ▪ Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Home sensor not de-activated.	▪ Check whether there are visible obstructions impairing the vial plate. ▪ Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Home sensor not reached.	▪ Check whether there are visible obstructions impairing the vial plate. ▪ Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Horizontal: home sensor activated when not expected.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Horizontal: home sensor not de-activated.	▪ Check whether there are visible obstructions impairing the needle unit. ▪ Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.

System message	Explanation
Horizontal: home sensor not reached.	▪ Check whether there are visible obstructions impairing the needle unit. ▪ Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Horizontal: needle position is unknown.	Initialize the needle unit using the control software.
Illegal sensor readout.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Incorrect first destination vial.	Check parameters in control software and correct entry.
Injection needle unit error.	▪ Check whether there are visible obstructions impairing the needle unit. ▪ Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Injection valve or ISS unit error.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Injection volume (number) is invalid. For specified injection method, volume should be within the range %.2f µl-%.2f µl, with (number). µl increments.	Check parameters in control software and correct entry.
Injection volume error.	Check parameters in control software and correct entry.
Invalid (number) vial position (number). The vial position must be between 01 and (number).	Check parameters in control software and correct entry.
Invalid combination of the trays. The combination of different trays for the Mix&Dilute mode is not allowed.	▪ Insert the correct vial plate. ▪ Check parameters in control software and correct entry.
Invalid combination of the trays. The combination of plates 384 low and 96 high is not allowed.	▪ Insert the correct vial plate. ▪ Check parameters in control software and correct entry.

System message	Explanation
Invalid configuration. ISS option not installed on autosampler. Please switch off this option in configuration dialog.	Check parameters in control software and correct entry.
Invalid configuration. SSV option not installed on autosampler. Please switch off this option in configuration dialog.	Check parameters in control software and correct entry.
Invalid flush volume (number) µl. The flush volume should be between 0 and (number) µl.	Check parameters in control software and correct entry.
Invalid instrument is detected.	Check parameters in control software and correct entry.
Invalid loop volume (number) µl. The loop volume should be between 0 and (number) µl.	Check parameters in control software and correct entry.
Invalid mix program: no Destination vial is specified in the configuration dialog.	Check parameters in control software and correct entry.
Invalid mix program: no Reagent A vial is specified in the configuration dialog.	Check parameters in control software and correct entry.
Invalid mix program: no Reagent B vial is specified in the configuration dialog.	Check parameters in control software and correct entry.
Invalid mix times. The time should be between 1 and 9.	Check parameters in control software and correct entry.
Invalid needle height (number) mm. The needle height should be between (number) and (number) mm.	Check parameters in control software and correct entry.
Invalid time-based method. Several AUX events have the same time.	Check parameters in control software and correct entry.

System message	Explanation
Invalid time-based method. Several SSV events have the same time.	Check parameters in control software and correct entry.
Invalid tray temperature (number) ˚C. The temperature should be between 4 and 22 ˚C.	Check parameters in control software and correct entry.
Invalid loop volume (number) µl. The loop volume should be between 0 and (number) µl.	Check parameters in control software and correct entry.
Invalid loop volume (number) µl. The loop volume should be between 0 and (number) µl.	Check parameters in control software and correct entry.
Invalid wait time. The time should be between 0 and 9 h 50 min 59 sec.Invalis wash volume (number) µl. The loop volume should be between 0 and (number) µl.	Check parameters in control software and correct entry.
Invalid loop volume (number) µl. The volume should be between the 0 and the syringe volume (%d µl).	Check parameters in control software and correct entry.
ISS valve error.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Missing destination vial.	▪ Check position of sample vial. ▪ Check parameters in control software and correct entry.
Missing reagent vial.	▪ Check position of sample vial. ▪ Check parameters in control software and correct entry.
Missing transport vial.	▪ Check position of sample vial. ▪ Check parameters in control software and correct entry.
Needle movement error.	▪ Check position of needle unit. ▪ Switch the device off and on.

System message	Explanation
Missing vial.	▪ Check position of needle unit. ▪ Switch the device off and on.
No destination vial is specified in the configuration.	Check parameters in control software and correct entry.
No reagent A vial is specified in the configuration.	Check parameters in control software and correct entry.
No reagent B vial is specified in the configuration.	Check parameters in control software and correct entry.
No user defined or mix program is running.	Check parameters in control software and correct entry.
Not enough reagent liquid.	Check volume of liquid and change as required.
Not enough transport liquid available due to missing transport vials.	Check volume of liquid and change as required.
Please specify inject marker or AUX event to be able to trigger the run.	Check parameters in control software and correct entry.
Selecting transport position failed.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Serial number is not valid. Please check the configuration.	Check parameters in control software and correct entry.
Setting mix program error.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Setting service mode failed.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Syringe dispenser unit error.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Syringe home sensor not de-activated.	▪ Needle flushing with control software. ▪ Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.

System message	Explanation
Syringe home sensor not reached.	▪ Needle flushing with control software. ▪ Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Syringe position is unknown.	Initialize the syringe unit using the control software.
Syringe rotation error.	▪ Needle flushing with control software. ▪ Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Syringe valve did not find destination position.	▪ Needle flushing with control software. ▪ Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Temperature above 48 °C at cooling ON.	▪ Switch off the cooling and check whether ambient temperature sensor is properly functioning. ▪ Inform the Technical Support of the manufacturer in case the system message repeats itself.
ISS option not installed on autosampler. Please switch off ISS-B option in configuration dialog.	Check control software configuration and correct entry.
The autosampler is not ready. Please try later.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
The injection volume of (number) µl is invalid. For the specified injection method, volume should equal (number) µl.	Check parameters in control software and correct entry.
Tray error.	Check parameters in control software and correct entry.
Valve error.	Check parameters in control software and correct entry.
Vertical: home sensor not de-activated.	▪ Check whether there are visible obstructions impairing the needle unit. ▪ Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.

System message	Explanation
Vertical: home sensor not reached.	▪ Check whether there are visible obstructions impairing the needle unit. ▪ Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Vertical: needle position is unknown.	Initialize the instrument in the control software.
Vertical: stripper did not detect plate (or wash/ waste). Missing vial.	▪ Check sample vial and plate. ▪ Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Vertical: stripper stuck.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Vertical: The sample needle arm is at an invalid position.	Switch the device off and on. Inform the Technical Support of the manufacturer in case the system message repeats itself.
Wear-out limit reached.	Switch the device off and on. If the system message appears again, notify KNAUER Technical Support. The valve must be replaced.
Wrong tubing volume. The largest tubing volume for standard injections is 200 µl.	Check parameters in control software and correct entry.

Technical data

Main features

Sample injection

Max. plate/vial height	47 mm (incl. septa or capmat)
Injection volume range	see "Analytical versions" or "Preparative versions" on page 69.
Sample loop	see "Analytical versions" or "Preparative versions" on page 69.
Dispenser syringe	see "Analytical versions" or "Preparative versions" on page 69.

Headspace pressure	built-in compressor, only for sample vials with septum
Switching time inj. valve	< 100 ms
Piercing needle precision	± 0.6 mm
Sample carrier Cooling / -heating	with cooling/heating function 4-40 °C
Vial detection	missing vial/well plate detection by sensor
Needle flushing	programmable: wash between injections and wash between vials
Wetted materials	Tefzel®, Vespel®, glass, Teflon® (PTFE) standard: Stainless steel, PEEK bio: PEEK
Injection modes	see "Analytical versions" or "Preparative versions" on page 69.
Injection precision	RSD (Relative Standard Deviation): ▪ full loop filling < 0.3 % ▪ partial loop filling at injection volumes > 5 µl: < 0.5 % ▪ microliter pickup at injection volumes > 5 µl: < 1.0 %
Sample carryover	see "Analytical versions" or "Preparative Versions" on page 69.
Injections per vial	max. 9 injections
Injection cycle time	see "Analytical Versions" or "Preparative Versions" on page 69.
Analysis time	max. 9 h, 59 min, 59 s

Communication

Interfaces	LAN, ANALOG, RS-232
Control	Ethernet (LAN)
Inputs	2 programmable TTL inputs (next injection, freeze, stop)
Outputs	1 programmable relay output (inject marker, auxiliary, alarm)

General

Energy demand	95 – 240 V AC +/- 10%, 50 - 60 Hz
Energy consumption	200 VA
Dimensions (width × height × depth)	360 × 370 × 540 mm 360 × 370 × 605 mm
weight	30 kg Cool/Heat 32 kg
Stackable weight (Maximum resting weight)	65 kg
Leak sensor	none
Ambient conditions	Ttemperature range: 10 – 40 ˚C; 50 – 104 ˚F Air humidity: 20-80 %

Device variants

Analytical Versions

Sample capacity	max. 768 samples (microtiter plates) or 108 standard autosampler vials
Injection volume range	0.1–10 000 µl programmable
Sample loop	10 µl/100 µl
Dispenser syringe	250 µl
Injection mode	Full loop filling, partial loop filling and microliter pickup, PASA™ (pressure-assisted sample aspiration)
Injection volumes	▪ full loop filling: max. 10 000 µl ▪ partial loop filling: 5000 µl (50 % of loop volume) ▪ microliter pickup: max. 4 625 µl (50 % loop volume - 1.5× needle volume) ▪ 0.1 µl increment for all injection modes
Sample carryover	< 0.01 % under typical conditions using needle wash < 0.005 % under special conditions with extended needle wash
Injection cycle time	min. 7 s from the same vial, 14 s from different vials; <60 s for =100 µl sample injection in all injection modes, incl. 300 µl needle wash

Preparative Versions

Sample capacity	24 vials, 10 ml each (LSV)
Injection volume range	1 – 10 000 µ µl programmable
Sample loop	10 ml
Dispenser syringe	500 µl/500 µl
Injection modes	Partial loop filling
Sample carryover	< 0.1 % under typical conditions using needle wash
Injection cycle time	min. 7 s from the same vial, 14 s from different vials; <60 s for =100 µl sample injection in all injection modes, incl. 300 µl needle wash

Repeat orders

The list of repeat orders is current at the time of publication. Deviations are possible at later date.

Use the included packing list for repeat orders of spare parts. If there are any questions concerning repeat orders, contact the Technical Support.

Further information on spare parts and accessories can be found online: www.knauer.net.

	Name	Order number
Device	AZURA® Autosampler 6.1L, Basic, 700 bar incl. accessories	AAA00AA
	AZURA® Autosampler 6.1L, Basic cool/heat, 700 bar incl. accessories	AAA01AA
	AZURA® Autosampler 6.1L, Basic, 1240 bar incl. accessories	AAA10AA
	AZURA® Autosampler 6.1L, Cool/Heat, 1240 bar incl. accessories	AAA11AA
	AZURA® Autosampler AS 6.1L, Bio	AAA20AA
	AZURA® Autosampler AS 6.1L, Bio Cool/Heat	AAA21AA
	AZURA® Autosampler AS 6.1L, Prep Cool/Heat	AAA31AA
	AZURA® Autosampler AS 6.1L, Prep	AAA40AA
	AZURA® Autosampler AS 6.1L, Bio Cool/Heat	AAA41AA
Vial plate	Vial plates for 48 1,5 ml Vials, 2 pieces	A50050

	Name	Order number
	Vial plates for 84 × 1.5 ml und 3 × 10 ml vials	A500501
	Prep. vial adapter plate for 12×10 ml	A500502
	96 well plate, U sanitized, 0.35 ml	A1823
	96 well plate, U sanitized, 1.2 ml	A1823V1
Tubing	PTFE tubing, 3.2 mm (1/8") OD, 1.5 mm ID, 300 cm length	A0732
	Silicone tubing 8.0 mm ID, 2 m	A0991-69
Syringe	Syringe 100 µl	M2866
	Syringe 250 µl	M0361
	Syringe 500 µl	M2070
	Syringe 2500 µl	M20701
Sample needle	Sample needle kit for SPARK valve 1/16"	A64700
	Bio inert silicone sample needle including tubing, nut and ferrule	A15086
Air needle	Air needle, nature, 62 mm	A50058
	Set with air needles (includes one piece of M20401, M20402, M20403, M20404, M20405)	A50059
	Air needle, yellow, 50 mm	M20401
	Air needle, red, 56 mm	M20402
	Air needle, blue, 68 mm	M20403
	Air needle, green, 74 mm	M20404
	Air needle, black, 80 mm	M20405
Optional accessories	2 x 2.5 A fuse	M2067
	Network cable	A5255
	Wash bottle 250 ml rectangular	M2054
	Accessory kit with vials caps, septum, and pliers for opening and closing	A0664
	125 Vials 10 ml, 500 crimp caps and 500 septa, ø 22 mm	A1662

Legal information

Transport damage

The packaging of our devices provides the best possible protection against transport damage. Check the devices for signs of transport damage. In case you notice damages, contact the Technical Support and the forwarder company within three workdays.

Warranty conditions

The factory warranty for the device is stipulated by contract. During the warranty period, any components with material or design-related defects will be replaced or repaired by the manufacturer free of charge. Please connect to our website for further information on terms and conditions.

All warranty claims shall expire in the event that any unauthorized changes are made to the device. This warranty also excludes the following:

- accidental or willful damage
- damage or errors caused by third parties that are not contractually related to the manufacturer at the time the damage occurs
- wear parts, fuses, glass parts, columns, light sources, cuvettes and other optical components
- damage caused by negligence or improper operation of the device and damage caused by clogged capillary
- packaging and transport damage

In the event of device malfunctions, directly contact the manufacturer:

KNAUER Wissenschaftliche Geräte GmbH
Hegauer Weg 38
14163 Berlin, Germany
Phone: +49 30 809727-111
Fax: +49 30 8015010
E-Mail: support@knauer.net
Internet: www.knauer.net

Warranty seal

A warranty seal is attached on some devices. The warranty seal is color-coded. A blue seal is used by the assembly or technical support of KNAUER for devices to be sold. After repair, service technicians stick an orange seal in identical position. If unauthorized persons interfere with the device or the seal is damaged, the warranty claim becomes void.

Declaration of Conformity

The Declaration of Conformity accompanies the product as a separate document and is available online: https://www.knauer.net/de/Support/Declarations-of-conformity

Disposal

Hand in old devices or disassembled old components at a certified waste facility, where they will be disposed of properly.

AVV marking in Germany

According to the German „Abfallverzeichnisverordnung“ (AVV) (January, 2001), old devices manufactured by KNAUER are marked as waste electrical and electronic equipment: 160214.

WEEE registration

KNAUER as a company is registered by the WEEE number DE 34642789 in the German „Elektroaltgeräteregister“ (EAR). The number belongs to category 8 and 9, which, among others, comprise laboratory equipment.

All distributors and importers are responsible for the disposal of old devices, as defined by the WEEE directive. End-users can send their old devices manufactured by KNAUER back to the distributor, the importer, or the company free of charge, but would be charged for the disposal.

Solvents and other operating materials

All solvents and other operating materials must be collected separately and disposed of properly.

All wetted components of a device, e. g. flow cells of detectors or pump heads and pressure sensors for pumps, have to be flushed first with isopropanol and then with water before being maintained, disassembled or disposed.

HPLC glossary

Term	Definition
Analytical	analysis and determination in terms of volume for HPLC samples (see: preparative).
Chromatogram	Record of a detector signal, depending on output volume of mobile phase and time
detector	device measuring the composition or the quantity of a substance.
Eluent	mobile phase transporting substances to be separated or isolated through the column
GLP	Good Laboratory Practice, quality assurance system for laboratories
HPLC	High-Pressure Liquid Chromatography (HPLC) liquids containing particles
Capillary	thin metal or PEEK pipe that connects components and devices within the chromatography system
Solvent	mobile solvent transporting substances to be separated or isolated through the column
Peak	deflection of an analyte by the detector in a differential chromatogram
Sample	Mixture of different components, which are to be separated using chromatography. They are transported by the mobile phase and dissolved from the column.
Sample loop	A loop, which is separated from the system by the valve, that contains sample first. After switching the valve, the eluent flows through the loop and is flushed to the column.
Pump	Device that pumps the mobile phase at a controlled volume flow into the chromatography system
Column	Pipe with final closures, which allow the mobile phase to pass. The pipe contains the packing materials.
Valve	mechanism to insert the sample into the eluent flow

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