cytiva

ÄKTA avant

Operating Instructions

Original instructions

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Table of Contents

1 Introduction

About this chapter

This chapter contains important user information, descriptions of safety notices, intended use of the ÄKTA™ avant instrument, and lists of associated documentation.

In this chapter

1.1 About this manual

Purpose of this manual

The *Operating Instructions* provide you with the information needed to install, operate and maintain the product in a safe way.

Scope of this manual

The *Operating Instructions* cover ÄKTA avant 25 and ÄKTA avant 150 instruments, using previously created methods in UNICORN software 6.3.2 and later.

Typographical conventions

Software items are identified in the text by ***bold italic*** text.

Hardware items are identified in the text by **bold** text.

In electronic format, references in *italics* are clickable hyperlinks.

1.2 Important user information

Read this before operating the product

All users must read the entire *Operating Instructions* before installing, operating or maintaining the product.

Always keep the *Operating Instructions* at hand when operating the product.

Do not operate the product in any other way than described in the user documentation. If you do, you may be exposed to hazards that can lead to personal injury and you may cause damage to the equipment.

Intended use of the product

ÄKTA avant is a liquid chromatography system intended for method and process development in purification of biomolecules. The system can be used to screen for optimal choice of columns, media and running parameters to purify selected proteins.

The ÄKTA avant system is intended for research use only, and shall not be used in any clinical procedures, or for diagnostic procedures.

Prerequisites

In order to follow this manual and use the system in the manner it is intended, it is important that:

- You have a general understanding of how the computer and Microsoft® Windows® work.
- You understand the concepts of liquid chromatography.
- You have read and understood the Safety instructions chapter in this manual.
- A user account has been created according to the *UNICORN™ Administration and Technical Manual*.

Definitions

This user documentation contains safety notices (WARNING, CAUTION, and NOTICE) concerning the safe use of the product. See definitions below.

WARNING

WARNING indicates a hazardous situation which, if not avoided, could result in death or serious injury. It is important not to proceed until all stated conditions are met and clearly understood.

CAUTION

CAUTION indicates a hazardous situation which, if not avoided, could result in minor or moderate injury. It is important not to proceed until all stated conditions are met and clearly understood.

NOTICE

NOTICE indicates instructions that must be followed to avoid damage to the product or other equipment.

Notes and tips

Note: *A note is used to indicate information that is important for trouble-free and optimal use of the product.*

Tip: *A tip contains useful information that can improve or optimize your procedures.*

1.3 Associated Documentation

Introduction

This section describes the user documentation that is delivered with ÄKTA avant.

User documentation on the CD

The user documentation listed in the table below is available in printed or PDF format. The complete documentation is also available on the User Documentation CD.

Document	Main contents
ÄKTA avant Unpacking Instruction (29101559)	Instructions for unpacking the instrument, and how to lift the instrument onto a bench.
ÄKTA avant Operating Instructions (29101556)	Instructions needed to install, operate and maintain the system in a safe way.
ÄKTA avant User Manual (29035184)	Instructions for handling the system. Descriptions of components. Information about how to run and maintain the system.
ÄKTA avant 25 Product Documentation (28991000) OR ÄKTA avant 150 Product Documentation (28984249)[1]	System specification and declaration of material conformity.

[1] The instrument is delivered with the relevant document.

UNICORN user documentation

The user documentation listed in the following table is available from the ***Help*** menu in UNICORN or from the ***UNICORN Online Help and Documentation*** software accessed by pressing the **F1** key in any UNICORN module.

Documentation	Main contents
UNICORN Help	Descriptions of UNICORN dialog boxes (available from the ***Help*** menu).
Getting started with Evaluation ***Note:*** *Available in UNICORN 7.0 and later.*	• Video clips showing common workflows in the Evaluation module. • Overview of features of the Evaluation module.

Documentation	Main contents
UNICORN Method Manual [1]	• Overview and detailed descriptions of the method creation features in UNICORN. • Workflow descriptions for common operations.
UNICORN Administration and Technical Manual [1]	• Overview and detailed description of network setup and complete software installation. • Administration of UNICORN and the UNICORN database.
UNICORN Evaluation Manual [1]	• Overview and detailed descriptions of the Evaluation Classic module in UNICORN. • Description of the evaluation algorithms used in UNICORN.
UNICORN System Control Manual [1]	• Overview and detailed description of the system control features in UNICORN. • Includes general operation, system settings and instructions on how to perform a run.

[1] Current UNICORN version is added to the title of the manual.

Data files, application notes and user documentation on the web

To order or download data files, application notes or user documentation, see the instruction below.

Step	Action
1	Go to cytiva.com/avant.
2	Click ***Product support***.
3	Click ***Related Documents***.
4	Select to download the chosen literature.

Additional literature

For practical tips on chromatography, refer to *Handbook 29010831*.

2 Safety Instructions

About this chapter

This chapter describes safety precautions, labels and symbols that are attached to the equipment. In addition, the chapter describes emergency and recovery procedures.

Important

WARNING

Before installing, operating or maintaining the product, all users must read and understand the entire contents of this chapter to become aware of the hazards involved.

In this chapter

2.1 Safety Precautions

Introduction

ÄKTA avant is powered by mains voltage and handles materials that may be hazardous. Before installing, operating or maintaining the system, you must be aware of the hazards described in this manual.

Follow the instructions provided to avoid personal injuries or damage to the product, or to other personnel and equipment in the area.

The safety precautions in this section are grouped into the following categories:

- *General precautions on page 11*
- *Personal protection on page 13*
- *Flammable liquids and explosive environment on page 14*
- *Installing and moving on page 15*
- *System operation on page 17*
- *Maintenance on page 20*

General precautions

WARNING

Risk assessment. Perform a risk assessment for any risks due to the process or process environment. Evaluate the effects the use of the product and the operational processes may have on the classification of the hazardous area. The process might cause the area to increase or the zone classification to change. Implement the risk reduction measures needed, including use of personal protective equipment.

WARNING

Always follow these General precautions to avoid injury when using the ÄKTA avant instrument.

- Do not operate the ÄKTA avant instrument in any other way than described in the ÄKTA avant and UNICORN manuals.
- Only properly trained personnel may operate and maintain the product.
- Before connecting a column, read the instructions for use of the column. To avoid exposing the column to excessive pressure, make sure that the pressure limit is set to the specified maximum pressure for the column.
- Do not use any accessories not supplied or recommended by Cytiva.
- Do not use ÄKTA avant if it is not working properly, or if it has suffered any damage, for example:
  - damage to the power cord or its plug
  - damage caused by dropping the equipment
  - damage caused by splashing liquid onto it

NOTICE

Avoid condensation. If ÄKTA avant is kept in a cold room, cold cabinet or similar, keep it switched on in order to avoid condensation.

Personal protection

WARNING

To avoid injury when working with the ÄKTA avant system, take the following measures for personal protection.

- Always use appropriate Personal Protective Equipment (PPE) during operation and maintenance of this product.
- **Hazardous substances and biological agents**. When using hazardous chemical and biological agents, take all suitable protective measures, such as wearing protective clothing, glasses and gloves resistant to the substances used. Follow local and/or national regulations for safe operation and maintenance of ÄKTA avant.
- **Spread of biological agents**. The operator must take all necessary actions to avoid spreading hazardous biological agents. The facility must comply with the national code of practice for biosafety.
- **High pressure**. The product operates under high pressure. Wear protective glasses and other required Personal Protective Equipment (PPE) at all times.

CAUTION

To avoid hazardous situations when working with the ÄKTA avant system, take the following measures for personal protection.

- Always use appropriate personal protective equipment when decommissioning the equipment.
- **Close doors.** To minimize the risk of exposure to hazardous chemicals and pressurized liquids, always close the foldable door and the pump cover before starting a run.
- **Cut injuries.** The tubing cutter is very sharp and must be handled with care to avoid injuries.

Flammable liquids and explosive environment

WARNING

When using flammable liquids with the ÄKTA avant system, follow these precautions to avoid any risk of fire or explosion.

- **Fire Hazard**. Before starting the system, make sure that there is no leakage.
- **Explosion hazard**. To avoid building up an explosive atmosphere when using flammable liquids, make sure that the room ventilation meets the local requirements.
- **Fraction collector**. Do **not** fractionate flammable liquids in the built-in fraction collector. When running RPC methods, collect fractions through the outlet valve or the optional external Fraction collector **F9-R**.
- **RPC runs with 100% acetonitrile and system pressure above 5 MPa (50 bar) in ÄKTA avant 25** . Always replace the green PEEK tubing between the used system pump and the pump pressure monitor with orange PEEK tubing, i.d. 0.5 mm, before running RPC with 100% acetonitrile. Set the system pressure alarm to 10 MPa (100 bar).
- **RPC runs with 100% acetonitrile in ÄKTA avant 150** . Always replace the beige PEEK tubing between the used system pump and the pump pressure monitor before running RPC with 100% acetonitrile. Replace it with green PEEK tubing, i.d. 0.75 mm.

Installing and moving

WARNING

To avoid injury when installing and moving the ÄKTA avant system, take the following measures for personal protection.

- **Move transport crates.** Make sure that the lifting equipment has the capacity to safely lift the crate weight. Make sure that the crate is properly balanced so that it will not accidentally tip when moved.
- **Heavy object**. The ÄKTA avant instrument weighs about 116 kg. Use proper lifting equipment, or use four or more persons when moving the instrument. All lifting and moving must be performed in accordance with local regulations.
- **Moving the product horizontally**. Three people are required to move the product horizontally.
- **Supply voltage.** Before connecting the power cord, make sure that the supply voltage at the wall outlet corresponds to the marking on the instrument.
- **Protective ground**. The product must always be connected to a grounded power outlet.
- **Power cord**. Only use power cords with approved plugs delivered or approved by Cytiva.
- **Access to power switch and power cord with plug**. Do not block access to the power switch and power cord. The power switch must always be easy to access. The power cord with plug must always be easy to disconnect.
- **Installing the computer**. The computer must be installed and used according to the instructions provided by the manufacturer of the computer.

NOTICE

To avoid damage to the instrument when installing and moving the ÄKTA avant system, take the following measures.

- Make sure that the waste vessels will hold all the produced volume of the run. For ÄKTA avant 25, a suitable waste vessel should typically have a volume of 2 to 10 liters. For ÄKTA avant 150, a waste vessel should have a volume of 40 liters.
- The maximum level of the waste vessel for the waste tubing from the valves must be lower than 30 cm above the lab bench.
- The maximum level of the waste vessel for the waste tubing from the fraction collector and the buffer tray must be lower than the bench height.
- **Vents on the product instrument**. To ensure adequate ventilation, keep papers and other objects away from the vents of the instrument.
- **Disconnect power.** To prevent equipment damage, always disconnect the power from the product before an instrument module is removed or installed, or a cable is connected or disconnected.
- **Misuse of UniNet-9 connectors.** The **UniNet-9** connectors at the rear panel should not be mistaken for Firewire connectors. Do not connect any external equipment to the **UniNet-9** connectors other than instrument modules designed for the product. See *ÄKTA avant User Manual* . Do not disconnect or move the **UniNet-9** bus cable.
- Any computer used with the equipment must comply with EN/IEC 60950-1, and be installed and used according to the manufacturer's instructions.

System operation

WARNING

To avoid personal injury when operating the ÄKTA avant system, follow these instructions.

- **Rotating the instrument.** Make sure that there is always at least 20 cm of free space around the ÄKTA avant instrument to allow for sufficient ventilation and rotation on the swivel foot. When rotating the instrument, take care not to stretch or squeeze tubing or cables. A disconnected cable may cause power interruption or network interruption. Stretched tubing may cause bottles to fall, resulting in liquid spillage and shattered glass. Squeezed tubing may cause increase in pressure, or block liquid flow. To avoid the risk of knocking over bottles, always place bottles on the buffer tray, and close the doors before rotating the instrument.
- **Fasten bottles and cassettes.** Always fasten bottles and cassettes to the rails at the front and side panel. Use appropriate holders for bottles. Shattered glass from falling bottles may cause injury. Spilled liquid may cause fire hazard and personal injury.
- **Electrical shock hazard after spillage.** If there is a risk that large volumes of spilled liquid may penetrate the casing of the instrument, immediately switch off the instrument, disconnect the power cord, and contact an authorized service engineer.
- **Moving parts in fraction collector**. Do not open the built-in fraction collector door when the instrument is running.
- **Using a Superloop** . After loading a Superloop, always plug the **Syr** port on the injection valve with a stop plug. With a Superloop connected to the valve, an over-pressure may be created during injection.
- **Overpressure.** Never block the outlet tubing with, for instance, stop plugs, since this will create overpressure and might result in injury.
- **Hazardous chemicals during run.** When using hazardous chemicals, run ***System CIP*** and ***Column CIP*** to flush the entire system tubing with distilled water, before service and maintenance.
- **Hazardous biological agents during run.** When using hazardous biological agents, run ***System CIP*** and ***Column CIP*** to flush the entire pump with bacteriostatic solution (e.g. 1M NaOH) followed by a neutral buffer and finally distilled water, before service and maintenance.

CAUTION

To avoid hazardous situations when operating the ÄKTA avant system, follow these instructions.

- **Risk of breaking test vials**. Do not use excessive force to press vials of wrong dimensions into the fraction collector cassettes. Glass vials may break and cause injuries.
- **Hazardous chemicals in UV flow cell**. Make sure that the entire flow cell has been flushed thoroughly with bacteriostatic solution, for example NaOH, and distilled water, before service and maintenance.
- **pH-electrode**. Handle the pH-electrode with care. The glass tip may break and cause injury.

- Do not fasten bottles with a volume of more than 1 liter in the front panel rails.
- **Max. weight on buffer tray**. Do not place containers with a volume of more than 10 liters each on the buffer tray. The total allowed weight on the buffer tray is 40 kg.

NOTICE

To avoid damage to the ÄKTA avant instrument or other equipment when operating the instrument, follow these instructions.

- **Keep UV flow cell clean**. Do not allow solutions containing dissolved salts, proteins or other solid solutes to dry out in the flow cell. Do not allow particles to enter the flow cell, as damage to the flow cell may occur.
- **Glass tube splinter**. Make sure to set the sample pressure below the maximum pressure of the Superloop before executing a flow in the ***Manual instructions*** dialog box when the Superloop is connected.
- **Avoid condensation**. If ÄKTA avant is kept in a cold room, cold cabinet or similar, keep it switched on in order to avoid condensation.
- **Avoid overheating.** If ÄKTA avant is kept in a cold cabinet and the cold cabinet is switched off, make sure to switch off ÄKTA avant and keep the cold cabinet open to avoid overheating.
- **Place the computer in room temperature**. If the ÄKTA avant instrument is placed in a cold room, use a cold room compatible computer or place the computer outside the cold room and use the Ethernet cable delivered with the instrument to connect to the computer.
- **UV and conductivity flow cells on the high pressure side**. When placing UV and/or conductivity flow cells on the high pressure side of the column, the UV flow cell has a maximum pressure limit of 2 MPa (20 bar) and the conductivity flow cell has a maximum pressure limit of 5 MPa (50 bar).

Maintenance

WARNING

To avoid personal injury when performing maintenance on the ÄKTA avant instrument, follow these instructions.

- **Electrical shock hazard**. All repairs should be done by service personnel authorized by Cytiva. Do not open any covers or replace parts unless specifically stated in the user documentation.
- **Disconnect power**. Always disconnect power from the instrument before replacing any component on the instrument, unless stated otherwise in the user documentation.
- **Corrosive chemicals during maintenance.** If the system or column is cleaned with a strong base or acid, flush with water afterwards and wash with a weak neutral buffer solution in the last step or phase.

NOTICE

To avoid damage to the ÄKTA avant instrument or other equipment when performing maintenance on the ÄKTA avant instrument, follow these instructions.

- **Cleaning**. Keep the exterior of the instrument dry and clean. Wipe regularly with a soft damp tissue and, if necessary, a mild cleaning agent. Let the instrument dry completely before use.
- **Advanced maintenance**. Read the instruction carefully before disassembly of the pump head.

2.2 Labels

Introduction

This section describes the information on the system label and other safety or regulatory labels that are attached to the product.

Labels on the ÄKTA avant instrument

The following illustration shows the labels that are attached to the ÄKTA avant instrument.

System label

The system label is located on the back of the equipment. The system label identifies the equipment and shows electrical data, regulatory compliance, and warning symbols.

Description of symbols on the labels

The following symbols and text may be present on the system label:

Label	Meaning
	**Warning!** **Fraction collector**. Do **not** fractionate flammable liquids in the built-in fraction collector. When running RPC methods, collect fractions through the outlet valve or the optional external Fraction collector **F9-R**.
	**Warning!** Read the Operating Instruction before using the system. **Electrical shock hazard**. All repairs should be done by service personnel authorized by Cytiva. Do not open any covers or replace parts unless specifically stated in the user documentation. **Supply voltage.** Before connecting the power cord, make sure that the supply voltage at the wall outlet corresponds to the marking on the instrument.
Voltage **Frequency** **Max. Power**	Electrical requirements: • Mains voltage (VAC) or other input voltage (AC or DC) • Frequency (Hz) • Max. power (VA)
Protection Class	Degree of protection provided by the enclosure.
Mfg. Year	Year (YYYY) and month (MM) of manufacture

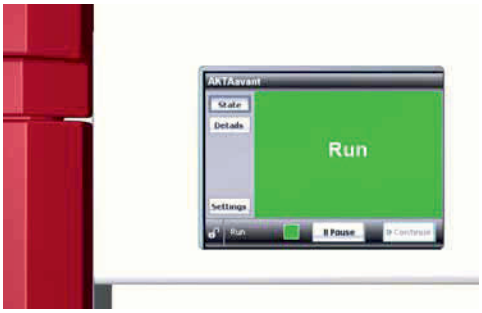
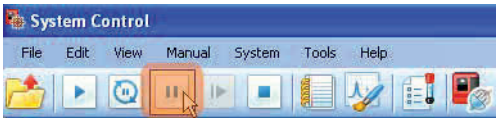
2.3 Emergency procedures

Introduction

This section describes how to perform an emergency shutdown of the ÄKTA avant instrument, including connected equipment. This section also describes the results in the event of power failure or network interruption.

Emergency shutdown

In an emergency situation, stop the run by either pausing the run or switching off the instrument as described in the following table:

If you want to...	then...
pause the run	• Press the ***Pause*** button on the instrument display. This will stop all pumps in the instrument.  or • To pause the run from UNICORN, click the ***Pause*** button in the ***System Control*** module:  *Result:* All pumps in the instrument are stopped.
switch off the instrument	• Push the **Power** switch to the **O** position, or • disconnect the power cord from the wall socket. *Result:* The run is interrupted immediately. ***Note:*** *The sample and data may be lost as a result of switching off the power.*

Power failure

The result of a power failure depends on which unit is affected.

Power failure to...	will result in...
ÄKTA avant instrument 	• The run is interrupted immediately • Data collected up to approx. 5 seconds before the power failure is available in UNICORN.
Computer 	• The UNICORN computer shuts down • The instrument display shows status ***Not connected*** • The run is interrupted immediately • Data generated up to 10 seconds before the power failure can be recovered ***Note:*** *The UNICORN client may lose connection to the instrument during a temporary overload of the processor and display an error message. This may appear as a computer failure. The run continues and you can restart the UNICORN client to regain control. No data will be lost.*

Uninterruptible power supply (UPS)

A UPS can prevent data loss during a power failure, and allow time for a controlled shut-down of ÄKTA avant.

For UPS power requirements, see the system specifications in this manual. Remember to also take into account the specifications for the computer and monitor. Refer to the manufacturers' documentation.

Note: *If using a UPS, the ÄKTA avant instrument, the computer and the monitor must be connected to the UPS.*

Restart the instrument after emergency shutdown or power failure

Follow the instructions to restart the instrument after an emergency shutdown or power failure.

Step	Action
1	Make sure that the condition that caused the emergency shutdown or power failure is corrected.
2	If the instrument was switched off, press the **Power** switch on the instrument.  *Result:* The instrument should start and the Instrument display should show **Not connected**.
3	Turn on the computer and monitor.
4	Start UNICORN and connect to the system. See instructions in *Section 4.4 Start UNICORN and connect to system, on page 67*.

3 System description

About this chapter

This chapter gives an overview of the ÄKTA avant instrument, software and accessories.

In this chapter

Illustration of the system

The following illustration shows the ÄKTA avant instrument with UNICORN software installed on a computer.

3.1 ÄKTA avant instrument overview

Introduction

This section shows an overview of the ÄKTA avant instrument. Technical details about the instrument and the individual modules are found in *ÄKTA avant User Manual*.

Exterior design

The ÄKTA avant instrument has a modular design, with all the liquid handling modules placed on the exterior of the instrument. Buffer vessels are placed on the buffer tray on top of the instrument. An instrument display is located on the front. From this side the built-in fraction collector is handled, as well as the sample. The remaining modules are placed on the right-hand side of the instrument. This side can be covered by a foldable door and a pump cover. By rotating the instrument using the swivel foot any side is easily accessed.

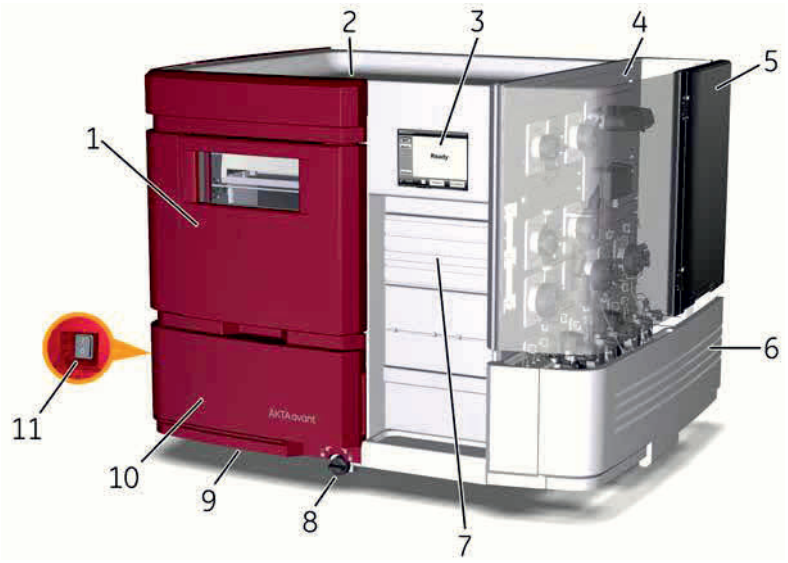
Operating ranges

The following table shows some of the operational limits of ÄKTA avant 25 and ÄKTA avant 150

Parameter	Limits	
	ÄKTA avant 25	**ÄKTA avant 150**
Flow rate	0.001 to 25 ml/min ***Note:*** *When running the **Column packing flow** instruction, the maximum flow rate is 50 ml/min.*	0.01 to 150 ml/min ***Note:*** *When running the **Column packing flow** instruction, the maximum flow rate is 300 ml/min.*
Max. operating pressure	20 MPa (200 bar)	5 MPa (50 bar)
UV monitor wavelength	190 to 700 nm	190 to 700 nm

Illustration of the main parts of the instrument

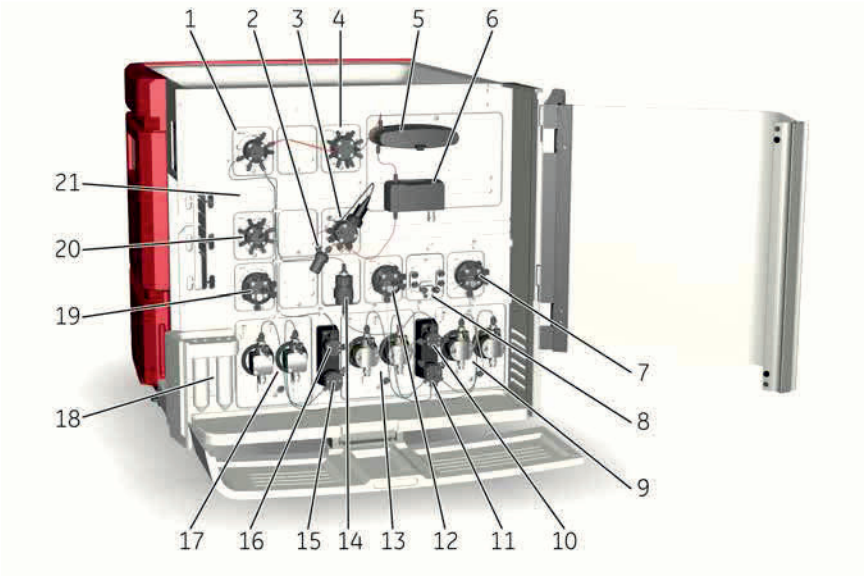
The following illustration shows the location of the main parts of the instrument.



Part	Function	Part	Function
1	Fraction collector	2	Buffer tray
3	Instrument display	4	Wet side
5	Foldable door	6	Pump cover
7	Holder rails	8	Swivel foot lock/unlock knob
9	Swivel foot	10	Swing out toolbox
11	Power switch		

Illustration of the wet side modules of the instrument

The following illustration shows the modules of the wet side of the instrument.



Part	Function	Part	Function
1	Injection valve	2	Flow restrictor
3	pH valve	4	Column valve
5	UV monitor	6	Conductivity monitor
7	Inlet valve B	8	Quaternary valve
9	System pump B	10	Pressure monitor of system pumps
11	System pump flow restrictor	12	Inlet valve A
13	System pump A	14	Mixer
15	Sample pump flow restrictor	16	Pressure monitor of sample pump
17	Sample pump	18	Pump rinsing solution tube
19	Sample inlet valve	20	Outlet valve
21	Holder rails		

Available modules

The ÄKTA avant instrument is always delivered with the standard modules installed, but one or more optional modules may be added to the flow path.

The following tables contains information on the standard modules and the optional modules of the ÄKTA avant 25 and ÄKTA avant 150 instruments. The sections that follows contain descriptions of the modules.

Note: *The valves for ÄKTA avant 25 and ÄKTA avant 150 are compatible with both systems but for the best performance the specific valve type should be used. The narrow channels in the valves for ÄKTA avant 25 will give too high back pressure if used above 50 ml/min. The larger volumes in the "H" valves for ÄKTA avant 150 may decrease resolution and increase peak broadening if used in ÄKTA avant 25.*

Standard modules

Module	Label in	
	ÄKTA avant 25	**ÄKTA avant 150**
System pump A	**P9 A**	**P9H A**
System pump B	**P9 B**	**P9H B**
Sample pump	**P9-S**	**P9H S**
Pressure monitor	**R9**	**R9**
Mixer	**M9**	**M9**
Injection valve	**V9-Inj**	**V9H-Inj**
Quarternary valve	**Q9**	**Q9**
Inlet valve A	**V9-IA**	**V9H-IA**
Inlet valve B	**V9-IB**	**V9H-IB**
Sample inlet valve	**V9-IS**	**V9H-IS**
Column valve	**V9-C**	**V9H-C**
pH valve	**V9-pH**	**V9H-pH**
Outlet valve	**V9-O**	**V9H-O**
UV monitor	**U9-M**	**U9-M**
Conductivity monitor	**C9**	**C9**
Built-in fraction collector	NA	NA

Optional modules

Module	Label in	
	ÄKTA avant 25	**ÄKTA avant 150**
Second Inlet valve A	**V9-A2**	**V9H-A2**
Second Inlet valve B	**V9-B2**	**V9H-B2**

Module	Label in	
	ÄKTA avant 25	**ÄKTA avant 150**
Extra Inlet valve X1	**V9-IX**	**V9H-IX**
Extra Inlet valve X2	**V9-IX**	**V9H-IX**
Second Sample inlet valve	**V9-S2**	**V9H-S2**
Versatile valve	**V9-V**	**V9H-V**
Loop valve	**V9-L**	**V9H-L**
Second Column valve	**V9-C2**	**V9H-C2**
Second Outlet valve	**V9-O2**	**V9H-O2**
Third Outlet valve	**V9-O3**	**V9H-O3**
External air sensor L9-1.5	**L9-1.5**	**L9-1.5**
External air sensor L9-1.2	**L9-1.2**	**L9-1.2**
I/O-box	**E9**	**E9**
Second UV monitor	**U9-L**	**U9-L**
Second Conductivity monitor	**C9**	**C9**
Second Fraction collector	**F9-R**	**F9-R**

Description of standard modules

The following modules are installed in the instrument when delivered.

Module	Description
System pump A **P9 A** or **P9H A**	A high precision pump, which delivers buffer in purification runs.
System pump B **P9 B** or **P9H B**	A high precision pump, which delivers buffer in purification runs.
Sample pump **P9-S** or **P9H S**	A high precision pump which delivers sample or buffer in purification runs.
Pressure monitor **R9**	Pressure monitor which reads the system pressure after System Pump A and System Pump B.

Module	Description
Pump flow restrictor	Prevents the system from siphoning when the flow path after the pump is open. Gives a small back pressure to the pump in extreme low pressure applications.
Mixer **M9**	Mixes the buffers delivered from the system pumps to a homogeneous buffer composition. Three mixer chambers are available for ÄKTA avant 25. Available volumes are: 0.6 ml, 1.4 ml (mounted at delivery) and 5 ml. Three mixer chambers are available for ÄKTA avant 150. Available volumes are: 1.4 ml, 5 ml (mounted at delivery), and 15 ml.  **CAUTION** **Risk of explosion.** Do not use Mixer chamber 15 ml with an ÄKTA avant 25 system configuration. The maximum pressure for Mixer chamber 15 ml is 5 MPa (50 bar).
Inlet valve A **V9-IA** or **V9H-IA**	Inlet valve for System Pump A with seven inlet ports and integrated air sensor.
Inlet valve B **V9-IB** or **V9H-IB**	Inlet valve for System Pump B with seven inlet ports and integrated air sensor.
Quaternary valve **Q9**	Valve which allows automatic mixing of four different solutions.
Sample inlet valve **V9-IS** or **V9H-IS**	Inlet valve for sample solution, with eight inlet ports (seven sample inlets and one buffer inlet) and integrated air sensor.
Injection valve **V9-Inj** or **V9H-Inj**	Valve which directs sample onto the column.
Column valve **V9-C** or **V9H-C**	Column valve which connects up to five columns to the instrument, and directs the flow to one column at a time. The column valve features two integrated pressure sensors. Allows the user to choose flow direction through the column, or to bypass the column.

Module	Description
pH valve **V9-pH** or **V9H-pH**	Valve which enables the pH electrode to be included in the flow path or by-passed during a run. The pH electrode may be calibrated when installed in the pH Valve. It also enables the flow restrictor to be included in the flow path (default position) or by-passed during a run.
Outlet valve **V9-O** or **V9H-O**	Valve which directs the flow to the fraction collector, any of the ten outlet ports or waste.
UV monitor **U9-M**	Monitor which measures the UV/Vis absorbance at up to three wavelengths simultaneously in the range 190 to 700 nm.
Conductivity monitor **C9**	Monitor which continuously measures the conductivity of buffers and sample solutions.
Built-in fraction collector	Built-in fraction collector. A cooling function protects the fractions from heat degradation.

Core modules

Core modules need to be installed for the system to run. They are mandatory in the software.

All standard modules except the built-in fraction collector are considered core modules.

Description of optional modules

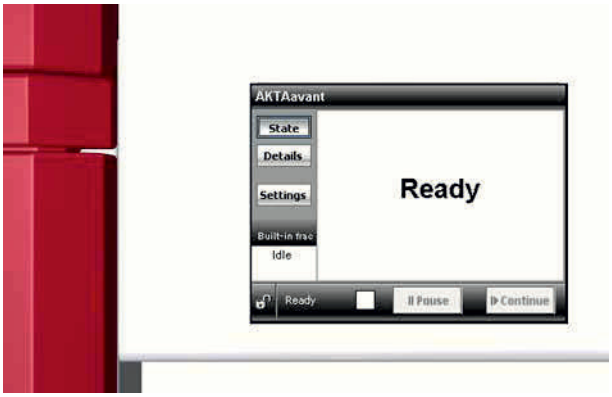
The following modules may be added to the flow path.

Module	Description
Second Inlet valve A **V9-A2** or **V9H-A2**	Second inlet valves for System pump A, to extend the number of inlets up to 14.
Second Inlet valve B **V9-B2** or **V9H-B2**	Second inlet valves for System pump B, to extend the number of inlets up to 14.
Inlet valve X1 and Inlet valve X2 **V9-IX** or **V9H-IX**	Inlet valve with eight inlet ports. No integrated air sensor.
Second Sample inlet valve **V9-S2** or **V9H-S2**	Second inlet valve for Sample pump to extend the number of sample inlets up to 14.
Versatile valve **V9-V** or **V9H-V**	A 4-port, 4-position valve, which can be used to customize the flow path.

Module	Description
Loop valve **V9-L** or **V9H-L**	Valve which enables automatic sample application from up to five sample loops, or to collect intermediate fractions in automated two-step purification.
Second Column valve **V9-C2** or **V9H-C2**	Valve which connects five additional columns to the instrument, extending the number of columns up to 10. The valve allows the user to choose flow direction through the column, or to by-pass the column.
Second Outlet valve **V9-O2** or **V9H-O2**	Valve which adds 12 outlet ports to the system, giving a total of 21 outlets.
Third Outlet valve **V9-O3** or **V9H-O3**	Valve which adds 12 outlet ports to the system, giving a total of 32 outlets
External air sensor **L9-1.5** or **L9-1.2**	Sensor which prevents air from being introduced into the flow path.
I/O-box **E9**	Module which receives analog or digital signals from, or transfers analog or digital signals to, external equipment that has been incorporated in the system.
Second UV monitor **U9-L**	Monitor which measures the UV absorbance at a fixed wavelength of 280 nm.
Second Conductivity monitor **C9**	Monitor which measures the conductivity of buffers and sample solutions.
Second Fraction collector **F9-R**	Round fraction collector that can collect up to 175 fractions.

Illustration of the instrument display

The following illustration shows the instrument display with the system state ***Ready*** showing.



Instrument display indicators and buttons

The instrument display is a touchscreen that shows the current system status. The instrument display includes the following indicators and buttons

Indicator/Button	Description
	Indicates if the Instrument display buttons are unlocked or locked. The buttons can be locked from UNICORN ***System Control***.
	Pauses the run and stops all pumps.
	Resumes instrument operation from the following states: • ***Wash*** • ***Pause*** • ***Hold***

3.2 UNICORN software

Introduction

This section gives an overview of the UNICORN software. It also describes the ***System Control*** module.

To learn more about ***System Control*** and the other three modules ***Administration***, ***Method Editor*** and ***Evaluation***, see the UNICORN documentation package.

In this section

3.2.1 UNICORN software overview

Introduction

This section gives a brief overview of the UNICORN software: a complete package for control, supervision and evaluation of chromatography instruments and purification runs.

From hereon, UNICORN refers to compatible versions of the software. The examples given in this manual are from UNICORN 6.4.

UNICORN modules overview

UNICORN consists of four modules: ***Administration***, ***Method Editor***, ***System Control*** and ***Evaluation***. The main functions of each module are described in the following table.

Module	Main functions
Administration	Perform user and system setup, system log and database administration.
Method Editor	Create and edit methods using one or a combination of: • Predefined methods with built-in application support • Drag-and-drop function to build methods with relevant steps • Line-by-line text editing The interface provides easy viewing and editing of run properties.
System Control	Start, monitor and control runs. The current flow path is illustrated in the ***Process Picture***, which allows manual interactions with the system and provides feedback on run parameters.
Evaluation	Open results, evaluate runs and create reports. • The default ***Evaluation*** module includes a user interface optimized for workflows like quick evaluation, compare results and work with peaks and fractions. • To perform operations like Design of Experiments, users can easily switch to ***Evaluation Classic***.

When working with the modules ***Administration***, ***Method Editor***, ***System Control*** and ***Evaluation Classic*** it is possible to access descriptions of the active window by pressing the **F1** key. This can be especially helpful when editing methods

3.2.2 The System Control module

Introduction

The ***System Control*** module is used to start, view, and control a manual or method run.

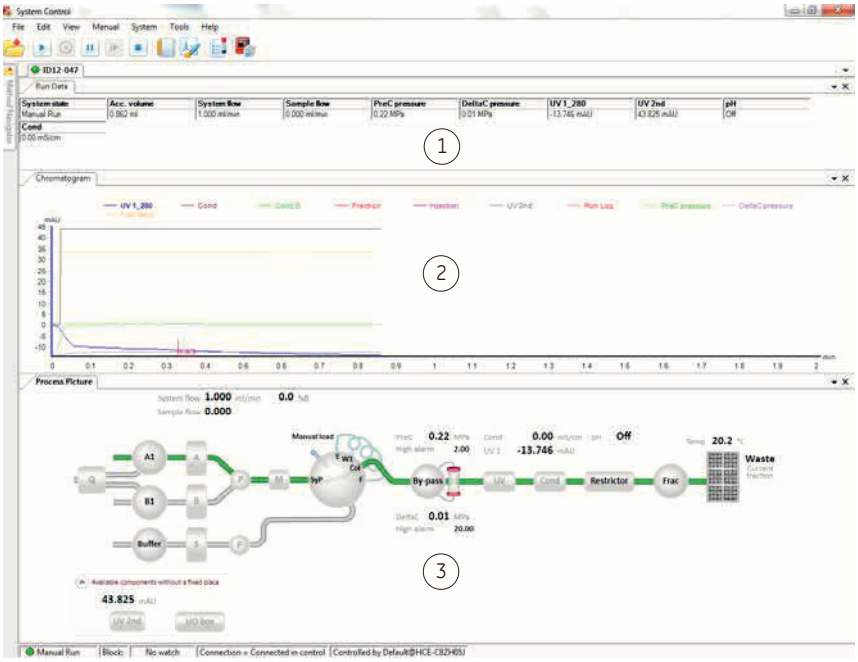
System Control panes

As seen in the following illustration, three panes are shown in the ***System Control*** module by default.

The ***Run Data*** pane (1) presents current data in numerical values.

The ***Chromatogram*** pane (2) illustrates data as curves during the entire run.

The current flow path is illustrated in the ***Process Picture*** (3), which allows manual interactions with the system and provides feedback on run parameters.



Note: *On the **View** menu, click **Run Log** to open the **Run Log** pane which presents all registered actions.*

System Control toolbar buttons

The following table shows the System Control toolbar buttons that are referred to in this manual.

Button	Function	Button	Function
	***Open Method Navigator***. Opens the ***Method Navigator*** where available methods are listed.		***Run***. Starts a method run.
	***Hold***. Suspends the method run, while current flow rate and valve positions are sustained.		***Pause***. Suspends the method run and stops all pumps.
	***Continue***. Resumes for example a held or paused method run.		***End***. Permanently ends the method run.
	***Customize***. Opens the ***Customize*** dialog box where curve settings, run data groups and run log contents can be set.		***Connect to Systems***. Opens the ***Connect to Systems*** dialog box where systems can be connected, and currently connected users are displayed.

4 Installation

About this chapter

This chapter provides the instructions necessary to enable users and service personnel to install the instrument and the software.

Read the entire Installation chapter before starting to install the ÄKTA avant instrument.

Note: *For information on how to how to unpack the ÄKTA avant instrument and how to lift the instrument onto a laboratory bench see ÄKTA avant Unpacking Instructions .*

In this chapter

4.1 Site preparation

Introduction

This section describes the site planning and the preparations necessary to perform before installation of an ÄKTA avant system. The purpose is to provide planners and technical staff with the data needed to prepare the laboratory for the installation.

The laboratory site must be planned and prepared before installing the ÄKTA avant system. The performance specifications of the system can be met only if the laboratory environment fulfills the requirements stated in this chapter.

In this section

4.1.1 Delivery and storage

Introduction

This section describes the requirements for receiving the delivery box and storing the instrument before installation.

Heavy object. The ÄKTA avant instrument weighs about 119 kg. Use proper lifting equipment, or be four or more people when moving the instrument. All lifting and moving must be performed in accordance with local regulations.

When you receive the delivery

- Record on the receiving documents if there is any apparent damage on the delivery box. Inform your Cytiva representative of such damage.
- Move the delivery box to a protected location indoors.

Delivery box

ÄKTA avant instruments are shipped in a delivery box with the following dimensions and weight:

Contents	Dimensions (mm)	Weight
ÄKTA avant instrument with accessories	1000 × 900 × 800 (width × height × depth)	155 kg

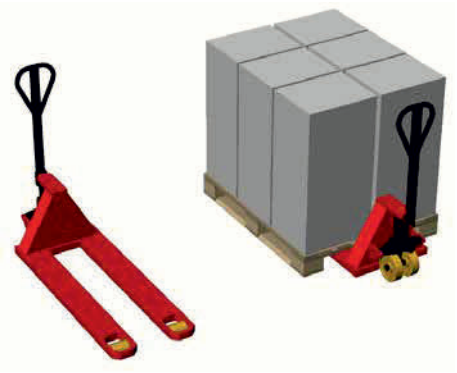
Storage requirements

The delivery boxes should be stored at a protected place indoors. The following storage requirements must be fulfilled for the unopened boxes:

Parameter	Allowed range
Ambient temperature, storage	-25°C to 60°C
Relative humidity	20% to 95%, noncondensing

Equipment for transportation

The following equipment is recommended for handling the delivery boxes:

Equipment	Specifications
Pallet mover	Suitable for a lightweight pallet 80 × 100 cm 
Cart for transporting the instrument to the lab	Dimensioned to accommodate the size and weight of the instrument

Unpacking the ÄKTA avant instrument

For information on how to how to unpack the ÄKTA avant instrument and how to lift the instrument onto a laboratory bench see *ÄKTA avant Unpacking Instructions*.

4.1.2 Room requirements

Introduction

This section describes the requirements for the transportation route and the room where the ÄKTA avant instrument is placed.

WARNING

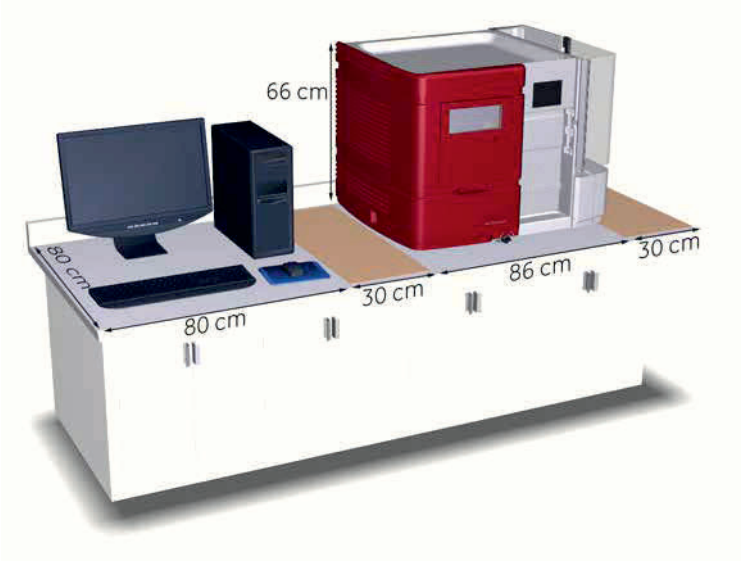
Access to power switch and power cord with plug. Do not block access to the power switch and power cord. The power switch must always be easy to access. The power cord with plug must always be easy to disconnect.

Transportation route

Doors, corridors and elevators must have a minimum width of 75 cm to allow for transporting the instrument. Allow additional space for moving around corners.

Space requirements

The following illustration shows the space recommended for the ÄKTA avant system.



Allow space on the laboratory bench for:

- handling of samples and buffers (2 × 30 cm)
- computer and monitor (80 cm)
- access for service (see the following topic)

Service access

To access the rear panel, the instrument can be rotated on a swivel foot. There must be at least 20 cm additional space on the bench to allow for free rotation.

WARNING

Rotating the instrument. Make sure that there is always at least 20 cm of free space around the ÄKTA avant instrument to allow for sufficient ventilation and rotation on the swivel foot. When rotating the instrument, take care not to stretch or squeeze tubing or cables. A disconnected cable may cause power interruption or network interruption. Stretched tubing may cause bottles to fall, resulting in liquid spillage and shattered glass. Squeezed tubing may cause increase in pressure, or block liquid flow. To avoid the risk of knocking over bottles, always place bottles on the buffer tray, and close the doors before rotating the instrument.

Laboratory bench

The bench must be clean, flat and stable to support the weight of the ÄKTA avant system, see the following table *Equipment weight on page 45*.

Equipment dimensions

The outer dimensions of the ÄKTA avant instrument are shown in the following illustration.

Equipment weight

Item	Weight
ÄKTA avant instrument	116 kg

Item	Weight
Computer	approximately 9 kg
Monitor	approximately 3 kg
Total	approximately 130 kg

4.1.3 Site environment

Introduction

This section describes the environmental requirements for installation of the ÄKTA avant instrument.

Environmental conditions

The following general requirements must be fulfilled:

- The room must have exhaust ventilation
- The instrument should not be exposed to sources of heat, such as direct sunlight
- Dust in the atmosphere should be kept to a minimum

The installation site must comply with the following specifications.

Environmental requirements

Parameter	Requirement
Allowed location	Indoor use only
Ambient temperature, operation	4°C to 35°C
Ambient temperature, storage	-25°C to 60°C
Relative humidity, operating	20% to 95%, noncondensing
Relative humidity, non-operating	20% to 95%, noncondensing
Altitude, operation	Up to 2000 m
Pollution degree of the intended environment	Pollution degree 2

Heat output

The heat output data is listed in the following table.

Component	Heat output
ÄKTA avant instrument	Typically 400 W Maximum 800 W
Computer, incl. monitor and printer	Typically 300 W Refer to manufacturer's instructions for more information.
Total heat output	*Typically 700 W* *Maximum 1100 W*

4.1.4 Power requirements

Introduction

This section describes the electrical power requirements for the ÄKTA avant instrument.

Electrical power requirements

The following table specifies the power requirements.

Parameter	Requirement
Supply voltage	100 to 240 V AC ±10%
Frequency	50/60 Hz
Transient overvoltages	Overvoltage category II
Max power consumption	800 VA
Number of sockets	1 socket per instrument, up to 3 sockets for computer equipment
Type of sockets	EU or US plugs. Grounded mains sockets, fused or protected by equivalent circuit breaker.
Location of sockets	Maximum 2 m from the instrument (due to length of mains cable). Extension cables can be used if required.

WARNING

Protective ground. The product must always be connected to a grounded power outlet.

Quality of power

The mains power supply must be stable and conform to specifications at all times to ensure reliable operation of the ÄKTA avant instrument. There should be no transient or slow changes in average voltage outside the limits specified above.

4.1.5 Computer requirements

Introduction

ÄKTA avant systems are controlled by UNICORN software running on a PC. The PC can be part of the delivery or be supplied locally.

The PC used must fulfill the recommendations stated in this section.

General computer specifications

For information about compatibility between UNICORN versions and the supported operating systems and database versions see the UNICORN compatibility matrix at *cytiva.com/UNICORNcompatibility*.

	UNICORN Client	Database Server	Workstation installation	E-License Server
Min. free disk space	6 GB	6 GB	12 GB	500 MB
Min. available RAM	3 GB	3 GB	3 GB	2 GB
Disc format	NTFS	NTFS	NTFS	NTFS
Architecture	Intel Dual Core (or faster)	Intel Dual Core (or faster)	Intel Dual Core (or faster)	Intel Dual Core (or faster)

Note:

- *UNICORN is tested using the English (U.S.) Code 1033 operating system language version. Using other language versions of the operating system may cause errors.*
- *A screen resolution of 1280x1024 or higher is recommended. Parts of the UNICORN user interface may not be displayed properly using a lower resolution.*
- *Changing the default font and changing the font size from 100% in Windows may cause problems in the UNICORN user interface.*
- *The Windows basic color scheme is recommended[1].*
- *Using the Windows 7 Aero color scheme is not recommended.*
- *Windows power save features should be turned off to avoid conflicts with system operations.*
- *UNICORN is not compatible with the Windows 7 feature High DPI Awareness, which allows the graphic user interface to be scaled. The interface scale must remain at 100% to avoid issues with clipping and misaligning of parts of the UNICORN user interface. Normally, the scale is set at 100% by default.*

[1] *UNICORN must be closed when the color scheme is changed.*

4.1.6 Required materials

Introduction

This section describes the accessories required for the installation and operation of the ÄKTA avant instrument.

Buffers and solutions

The buffers and solutions listed in the following table are required during the installation procedure and should be provided at the installation site.

Buffer/solution	Required volume	Scope of use
Distilled water	1 liter	Air sensor test, fraction collector test, Quaternary Valve test and system test
1% acetone in distilled water	0.5 liter	Quaternary Valve test
1% acetone and 1 M NaCl in distilled water	0.5 liter	System test
20% ethanol	200 ml	Priming of the pump piston rinsing system

Laboratory equipment

The equipment listed in the following table is required during the installation procedure and should be provided at the installation site.

Equipment	Specification
Flasks, liquid containers	For buffers and waste
Gloves	For protection
Protective glasses	For protection

Fraction collector tubes and bottles

The tubes and bottles used in the built-in fraction collector must fulfill the requirements listed in the following table. Examples of manufacturers are also listed in the table.

Tube or bottle size (ml)	Diameter (mm)		Height (mm)		Examples of manufacturers
	Min.	Max.	Min.	Max.	
3	10.5	11.5	50	56	Nunc™
5	10.5	12	70	76	VWR™
8	12	13.3	96	102	BD™ Biosciences, VWR
15	16	17	114	120	BD Biosciences
50	28	30	110	116	BD Biosciences
250 mL bottle	L: 55 W: 55[1]	L: 64.5 W: 64[1]	-	121	Nalgene™, Kautex™

[1] Length and width of the rectangular bottle base

Deep well plates, requirements

The deep well plates used in the built-in fraction collector must fulfill the requirements listed in the table below.

Property	Specification
No. of wells	24, 48, or 96
Shape of wells	Square, not cylindrical
Well volume	10, 5, or 2 ml

Approved deep well plates

The deep well plates listed in the table below are tested and approved by Cytiva to be used with the built-in fraction collector.

Plate type	Manufacturer	Part no.
96 deep well plate	Cytiva	7701-5200
	BD Biosciences	353966
	Greiner Bio-One	780270
	Porvair Sciences	219009
	Seahorse Bioscience	S30009
	Eppendorf™	951033405/ 0030 501.306
48 deep well plate	Cytiva	7701-5500

Plate type	Manufacturer	Part no.
	Seahorse Bioscience	S30004
24 deep well plate	Cytiva	7701-5102
	Seahorse Bioscience	S30024

4.2 Hardware installation

About this section

This section describes the installation procedure of an ÄKTA avant system.

Note: *For information on how to how to unpack the ÄKTA avant instrument and how to lift the instrument onto a laboratory bench see ÄKTA avant Unpacking Instructions .*

WARNING

Protective ground. The product must always be connected to a grounded power outlet.

WARNING

Power cord. Only use power cords with approved plugs delivered or approved by Cytiva.

WARNING

Access to power switch and power cord with plug. Do not block access to the power switch and power cord. The power switch must always be easy to access. The power cord with plug must always be easy to disconnect.

In this section

4.2.1 Install the computer equipment

Introduction

The computer is supplied as a part of the ÄKTA avant delivery, or supplied locally.

Unpacking and installing

Unpack and install the computer according to the manufacturer's instructions.

NOTICE

Any computer used with the equipment must comply with IEC 60950 and be installed and used according to the manufacturer's instructions.

4.2.2 Connect the system units

Introduction

The following interconnections must be made:

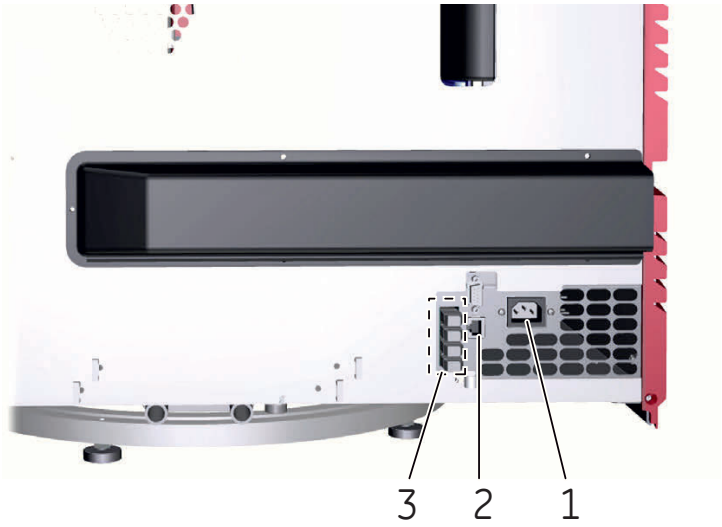
- power supply to the ÄKTA avant instrument
- power supply to the computer equipment
- network connection between the computer and the ÄKTA avant instrument

WARNING

- **Power cord**. Only use power cords with approved plugs delivered or approved by Cytiva.
- **Supply voltage.** Before connecting the power cord, make sure that the supply voltage at the wall outlet corresponds to the marking on the instrument.

Connector illustration

The following illustration shows the location of the connectors.



Part	Function
1	**Power** input connector
2	**Network** connector (Ethernet)

Part	Function
3	**UniNet-9** connectors ***Note:*** *Termination plugs must be connected to the connectors that are not in use.*

Other connectors are for use by authorized service engineers only.

NOTICE

Misuse of UniNet-9 connectors. The **UniNet-9** connectors at the rear panel should not be mistaken for Firewire connectors. Do not connect any external equipment to the **UniNet-9** connectors. Do not disconnect or move the **UniNet-9** bus cable.

Connect power to the ÄKTA avant instrument

Follow the instructions to connect power to the ÄKTA avant instrument.

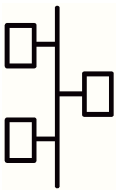
Step	Action
1	Select the correct power cord to be used. Each instrument is delivered with 2 alternative power cords: • Power cord with US-plug, 2 m • Power cord with EU-plug, 2 m Discard the unused power cord.
2	Connect the power cord to the **Power** input connector on the back of the instrument and to a grounded wall outlet 100 to 240 VAC , 50 to 60 Hz.
3	Attach the power cord to the rear of the instrument using the cable clip. 

Connect power to computer equipment

Follow the manufacturer's instructions to connect power to the computer, monitor and local printer (if used).

Connect to network

Follow the instructions to make network connections.

Step	Action
1	Connect a network cable between the network connector (Ethernet) on the back of the instrument and the computer network card dedicated to ÄKTA. The illustration shows the symbol of the Ethernet connector. 
2	If the computer is to be connected to an external network, connect a network cable between the main network card of the computer and a network wall outlet. ***Note:*** *If the computer has not been supplied by Cytiva and if network configuration is to be used, see UNICORN Administration and Technical Manual for further information on network settings.*

4.2.3 Prepare waste tubing

Location of waste tubing

All waste tubing is found on the rear of the instrument, see the following illustration.

Part	Description
1	Waste tubing from the injection valve, the pH valve and the outlet valve (pieces of tubing marked **W**, **W1**, **W2** and **W3**).
2	Waste tubing from the fraction collector and the buffer tray.

Prepare the waste tubing

Follow the instructions to prepare the waste tubing.

Step	Action
1	Place the four pieces of waste tubing from the injection valve, the pH valve and the outlet valve (pieces of tubing marked **W**, **W1**, **W2** and **W3**) in a vessel placed below the bench.

NOTICE

The maximum level of the waste vessel for the waste tubing from the valves must be lower than 30 cm above the lab bench.

2	Place the three pieces of waste tubing from the fraction collector and the buffer tray in a waste vessel placed below the bench.

NOTICE

The maximum level of the waste vessel for the waste tubing from the fraction collector and the buffer tray must be lower than the bench height.

3	Cut the waste tubing from the fraction collector and the buffer tray to appropriate length. It is important that the tubing is not bent and will not be submerged in liquid during the run.

Note: *If the tubing is too short, replace it with new tubing. Do not lengthen the tubing as this might cause obstruction of the tubing and flooding in the fraction collector chamber.*

CAUTION

Make sure that the waste vessels will hold all the produced volume of the run. For ÄKTA avant 25, a suitable waste vessel should typically have a volume of 2 to 10 liters. For ÄKTA avant 150, a waste vessel should have a volume of 40 liters.

4.2.4 Install the Barcode Scanner 2-D and the pH electrode

Introduction

This section describes how to install the Barcode Scanner 2-D and the pH electrode.

Install the barcode scanner

Connect the cable of the Barcode Scanner 2-D to the scanner head and to a USB port on the computer.

Install the pH electrode

If pH monitoring is to be used, you need to replace the dummy electrode mounted at delivery with a pH electrode.

CAUTION

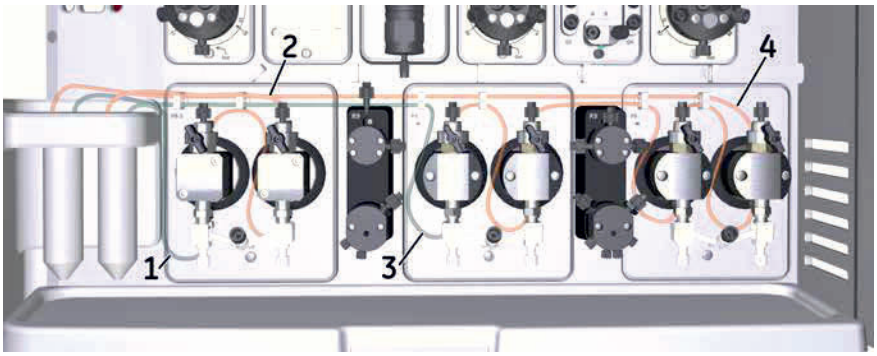
pH electrode. Handle the pH electrode with care. The glass tip may break and cause injury.

Follow the instructions to install the pH electrode.

Step	Action
1	Unpack the pH electrode. Make sure that the electrode is not broken or dry.
2	Unscrew the dummy electrode from the flow cell.
3	Pull off the plug from the connector on the front of the pH valve, and store the plug together with the dummy electrode.
4	Remove the cover from the tip of the pH electrode.
5	Carefully insert the electrode in the flow cell. Tighten the locking ring by hand to secure the electrode.
6	Connect the pH electrode cable to the connector on front of the pH valve.

4.2.5 Prepare the pump rinsing system

Illustration of the pump piston rinsing systems

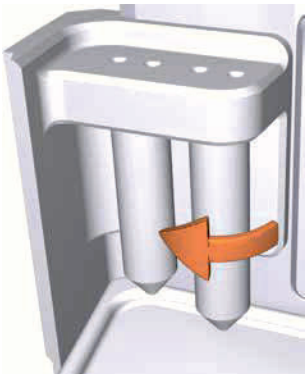


Part	Description
1	Inlet tubing to the sample pump piston rinsing system
2	Outlet tubing from the sample pump piston rinsing system
3	Inlet tubing to the system pump piston rinsing system
4	Outlet tubing from the system pump piston rinsing system

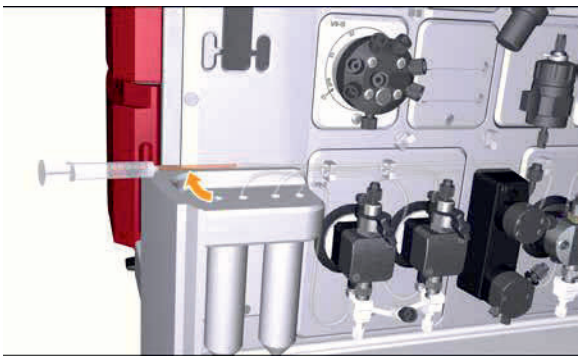
Prime the pump piston rinsing system

Follow the instructions to fill the pump piston rinsing systems with rinsing solution. See the tubing configuration of the rinsing systems in *Illustration of the pump piston rinsing systems on page 62*.

Step	Action
1	Unscrew the rinsing system tubes from the holders.



2	Fill each of the rinsing system tubes with 50 ml of 20% ethanol.
3	Screw the rinsing solution tubes back in the holders.
4	Immerse the inlet tubing to the system pump piston rinsing system in one of the rinsing solution tubes. ***Note:*** *Make sure that the inlet tubing reaches close to the bottom of the rinsing solution tube.*
5	Immerse the inlet tubing to the sample pump piston rinsing system in the other rinsing solution tube. ***Note:*** *Make sure that the inlet tubing reaches close to the bottom of the rinsing solution tube.*
6	Connect a 25 to 30 ml syringe to the outlet tubing of the system pump piston rinsing system. Draw liquid slowly into the syringe.



Step	Action
7	Disconnect the syringe and discard its contents.
8	Immerse the outlet tubing in the rinsing solution tube where the inlet tubing of the system pump piston rinsing system is immersed.
9	Connect a 25 to 30 ml syringe to the outlet tubing from the sample pump piston rinsing system. Draw liquid slowly into the syringe.
10	Disconnect the syringe and discard its contents.
11	Immerse the outlet tubing in the rinsing solution tube where the inlet tubing of the sample pump piston rinsing system is immersed.
12	Fill the rinsing solution tubes so that each of the tubes contains 50 ml of 20% ethanol.

4.2.6 Start the instrument and the computer

Introduction

This section describes how to start the instrument and the computer.

Instruction

Follow the instructions to start the instrument and the computer.

Step	Action
1	Switch on the instrument by pressing the **Power** switch to the **I** position.
	
	Result:
	The instrument starts and the Instrument display states **Not connected**.
2	Turn on the computer and monitor according to the manufacturer's instructions.

4.3 Software installation

Introduction

This section gives an overview of the different UNICORN installation types.

The software should be installed by an assigned UNICORN system administrator. Detailed information about software installation and configuration is available in the *UNICORN Administration and Technical Manual*.

Software installations

You can install UNICORN in one of the following configurations:

- as a complete UNICORN installation on a stand-alone workstation (full installation)
- as a UNICORN database and license server (custom installation)
- as a UNICORN software client and instrument server software on a network client station (custom installation)

4.4 Start UNICORN and connect to system

Introduction

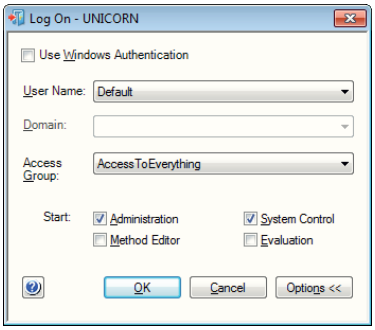
This section describes how to start and log on to UNICORN and how to connect the instrument to UNICORN.

Prerequisites

UNICORN must be correctly installed according to instructions in the *UNICORN Administration and Technical Manual*.

Start UNICORN and log on

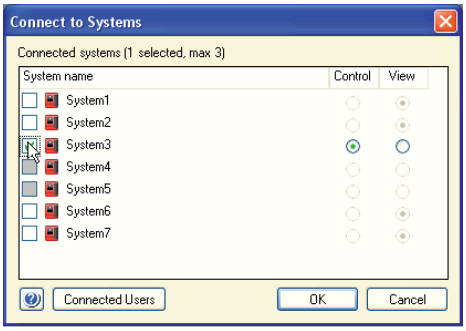
Follow the instructions to start UNICORN and log on to the program. A valid e-license must be available for the workstation. See *UNICORN Administration and Technical Manual* for more information about e-licenses.

Step	Action
1	Double-click the UNICORN icon on the desktop. *Result:* The ***Log On*** dialog box opens.
2	In the ***Log On*** dialog box: • select ***User Name*** and • enter ***Password***. ***Note:*** *It is also possible to select the* ***Use Windows Authentication*** *checkbox and enter a network ID in the* ***User Name*** *field.*  • click ***OK***. *Result:* The selected UNICORN modules open.

Connect to system

Follow the instructions to connect the instrument to UNICORN.

Note: *The system must have been defined by the UNICORN system administrator.*

Step	Action
1	In the ***System Control*** module, click the ***Connect to Systems*** button.  *Result:* The ***Connect to Systems*** dialog box opens. 
2	In the ***Connect to Systems*** dialog box: • Select a system check box. • Click ***Control*** for that system. • Click ***OK***. *Result:* The selected instrument can now be controlled by the software. ***Tip:*** *If UNICORN is unable to connect to the selected instrument, see Chapter Troubleshooting in ÄKTA avant User Manual.*

4.5 Prime inlets and purge pump heads

About this section

Before using the sample pump or system pumps it is important to do the following:

- Prime the inlets (fill the inlets with liquid).
- Purge the pumps (remove air from the pump heads).

This section describes how to prime the buffer inlets, sample inlets, and Q inlets, and how to purge the system pumps and the sample pump.

In this section

4.5.1 Prime buffer inlets and purge system pumps

Overview

The procedure consists of the following stages:

Stage	Description
1	Prime all inlet tubing to be used during the run.
2	Validate priming of inlet tubing.
3	Purge System Pump B if pressure signal indicates air bubbles.
4	Validate purge of System Pump B.
5	Purge System Pump A if pressure signal indicates air bubbles.
6	Validate purge of System Pump A.
7	End the run.

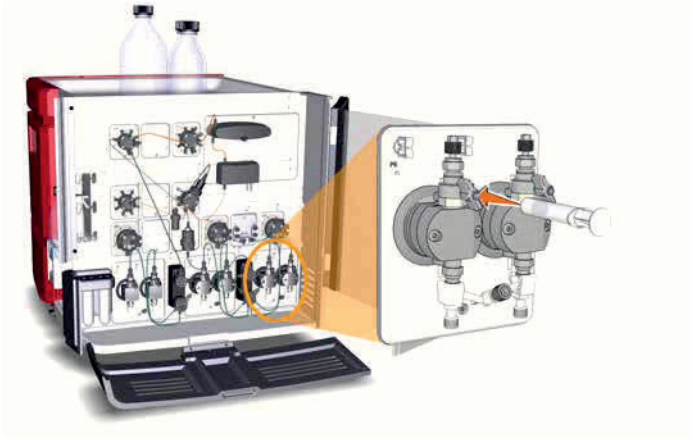
Note: *To increase life length of the pump sealing rings, make sure that the pump rinsing system is filled with fresh rinsing solution.*

Tip: *The procedures for purging the pump heads and priming the inlets using the* ***Process Picture****, are described in the following topic. It is also possible to perform the procedures from the* ***Manual instructions*** *dialog box.*

Prime inlet tubing

Follow the instructions to fill all A and B inlet tubing to be used in the run with appropriate buffer/solution.

Step	Action
1	Make sure that all inlet tubing that is to be used during the method run is placed in the correct buffer.
2	Open the ***System Control*** module.

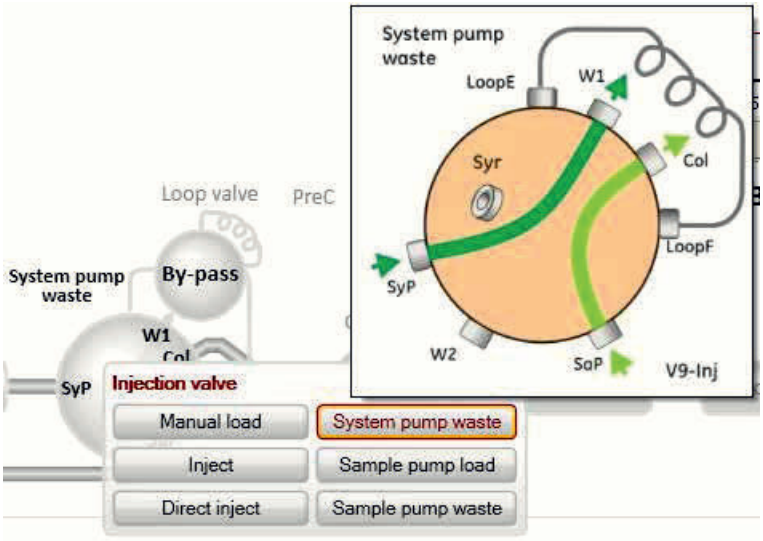
Step	Action
3	In the ***Process Picture***: • Click the inlet valve icons. (Click both the ***Inlet A*** and ***Inlet B*** icons, one at a time, if both inlets are to be primed.) • Click the position of the inlet to be filled. Fill the positions in reverse alphabetical order and start with the highest number. For example, if all the seven inlets in Inlet Valve B are to be filled, click them in the following order: ***B7***, ***B6*** . . . ***B1***, assuming that ***B1*** is the starting buffer.  *Result:* The inlet valve switches to the selected port.
4	Connect a 25 to 30 ml syringe to the purge valve of one of the pump heads of System Pump B. Make sure that the syringe fits tightly into the purge connector. 
5	Open the purge valve by turning it counter-clockwise about three quarters of a turn. Draw liquid slowly into the syringe until the liquid reaches the pump.
6	Close the purge valve by turning it clockwise. Disconnect the syringe and discard its contents.
7	Repeat steps 3 to 6 for each piece of inlet tubing that is to be used during the run. In the final inlet position, draw liquid into the syringe through both purge valves.

Step	Action
8	Check that there is no air left in the pump by following the instructions in *Validate prime or purge of System Pump A or B or Sample Pump on page 75*. If air bubbles are indicated, follow the instructions in *Purge System Pump B on page 72*

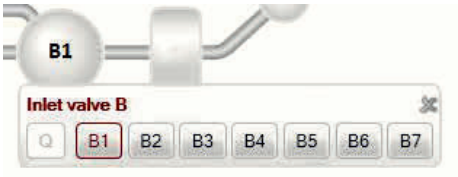
Purge System Pump B

If the priming was done thoroughly and the final buffer was drawn all the way into the syringe and the validation of the priming showed that there was no air left in the pump it is not necessary to purge System Pump B.

However, if the pressure signal indicated air bubbles left in the pump, follow these instructions to purge both pump heads of System Pump B:

Step	Action
1	Make sure that the piece of waste tubing connected to the injection valve port **W1** is placed in a waste vessel.
2	In the ***Process Picture***: • Click the ***Injection valve*** icon and then click ***System pump waste***. **Result**: The injection valve switches to waste position. This is necessary to achieve a low back pressure during the purge procedure. 

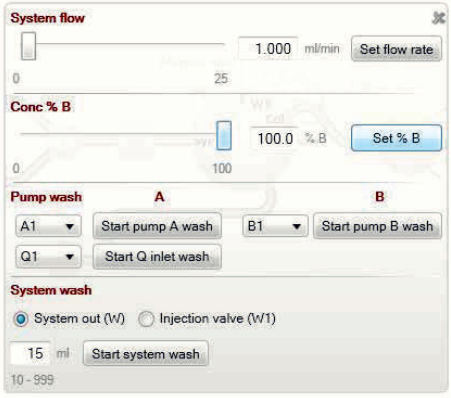
Step	Action
3	In the ***Process Picture***: • Click the ***Inlet valve B*** icon. • Click the position of one of the inlets that will be used at the beginning of the run.



Result:

The inlet valve switches to the selected port.

4	In the ***Process Picture***: • Click the ***System pumps*** icon. • Set ***Conc % B*** to 100% ***B*** and click ***Set % B***.

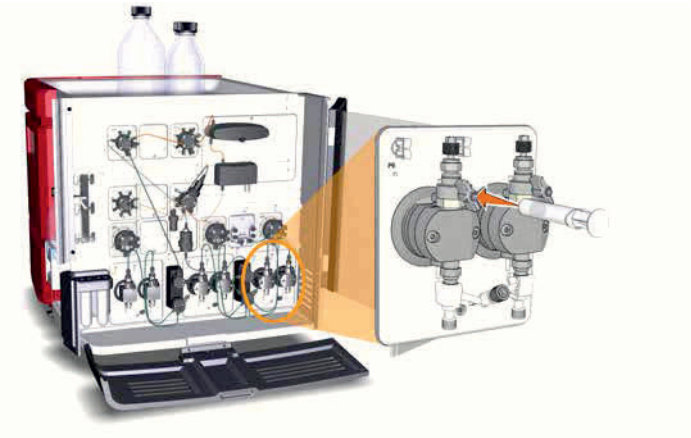


	• Set the ***System flow*** to 1.0 ml/min for ÄKTA avant 25 or 5.0 ml/min for ÄKTA avant 150. • Click ***Set flow rate***.

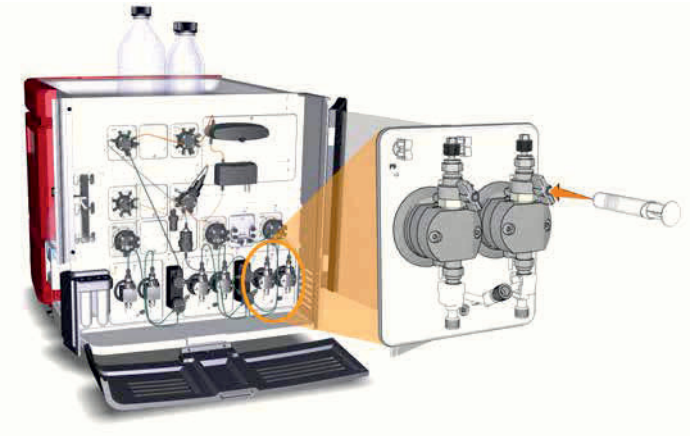
Result:

Only System Pump B is active, and a system flow through injection valve waste starts.

Step	Action
5	Connect a 25 to 30 ml syringe to the purge valve of the left pump head of System Pump B. Make sure that the syringe fits tightly into the purge connector.



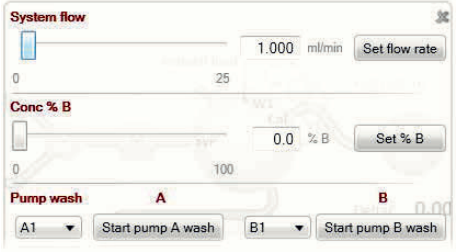
6	Open the purge valve by turning it counter-clockwise about three quarters of a turn. Draw 5 to 10 ml of liquid slowly into the syringe with a rate of about 1 ml/s.
7	Close the purge valve by turning it clockwise. Disconnect the syringe and discard its contents.
8	Connect the syringe to the purge valve on the right pump head of System Pump B, and repeat steps 6 to 8. Keep the system flow running.



9	Check that there is no air left in the pump by following the instructions in *Validate prime or purge of System Pump A or B or Sample Pump on page 75*.

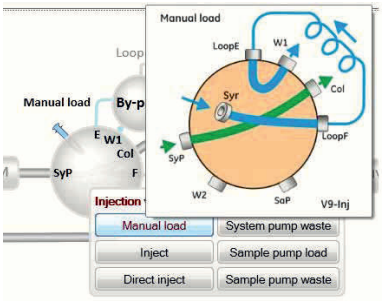
Purge System Pump A

Purge both pump heads of System Pump A by following the same procedure as in *Purge System Pump B, on page 72*, but replace step 3 and 4 with the following:

Step	Action
1	In the ***Process Picture***: • Click the ***Inlet valve A*** icon. • Click the position of one of the inlets that will be used at the beginning of the run.  *Result:* The inlet valve switches to the selected port.
2	In the ***Process Picture***: • Click the ***System pumps*** icon. • Set ***Conc % B*** to 0% B and click ***Set % B***.  *Result:* Only System Pump A is active.

Validate prime or purge of System Pump A or B or Sample Pump

Follow these instructions to check that there is no air left in the pump after performing a prime or a purge.

Step	Action
1	In the ***Process Picture***: • Click on the ***Injection valve*** and select ***Manual load***. *Result:* The injection valve switches to manual load position. 
2	Make sure that the pump flow is on.
3	In the ***Chromatogram*** pane: • Check the ***PreC pressure*** curve. • If the ***PreC pressure*** do not stabilize within a few minutes there may be air left in the pump. Purge the pump once more then see *ÄKTA avant User Manual*.

End the run

Click the ***End*** button in the ***System Control*** toolbar to end the run.

4.5.2 Prime sample inlets and purge Sample Pump

Overview

The procedure consists of the following stages:

Stage	Description
1	Prime all sample inlet tubing to be used during the run.
2	Validate priming of inlet tubing.
3	Purge the sample pump if pressure signal indicates air bubbles.
4	Validate purge of the samle pump.
5	End the run.

Note: *To increase life length of the pump sealing rings, make sure that the pump rinsing system is filled with fresh rinsing solution.*

Prime sample inlets

Follow the instructions below to fill all sample inlet tubing, to be used in the run, with appropriate buffer or sample solution.

Step	Action
1	Make sure that all sample inlet tubing that is to be used during the method run is immersed in the correct samples or buffer.
2	Make sure that the waste tubing connected to injection valve port **W2** is immersed in a waste vessel.
3	Open the ***System Control*** module.

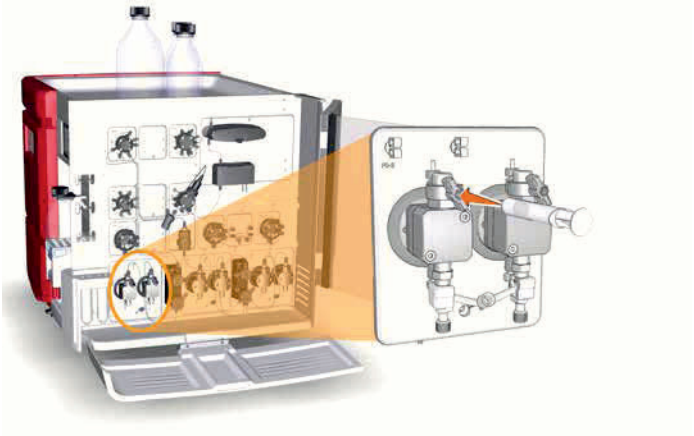
Step	Action
4	In the ***Process Picture***

- Click the ***Sample inlet valve*** icon.
- Select the position of the inlet to be filled. Start at the inlet position with the highest number and end at the position with the lowest number or the buffer position (assuming that the first sample to run is connected to inlet 1 etc.).

Result:

The sample inlet valve switches to the selected port.

5 Connect a 25 to 30 ml syringe to one of the purge valves of the pump heads in the sample pump. Make sure that the syringe fits tightly into the purge connector.



6 Open the purge valve by turning it counter-clockwise about three-quarters of a turn. Draw slowly with the syringe until the sample just passes the Sample inlet valve.

7 Close the purge valve by turning it clockwise. Disconnect the syringe and discard its contents.

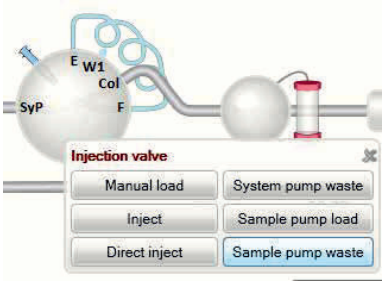
8 Repeat steps 4 to 7 for each sample inlet that is to be used in the method run. The final sample or the buffer from the buffer position should be drawn all the way through both pump heads into the syringe.

Step	Action
9	Check that there is no air left in the pump by following the instructions in *Validate prime or purge of System Pump A or B or Sample Pump on page 75*. If air bubbles are indicated, follow the instructions in *Purge Sample Pump on page 79*.

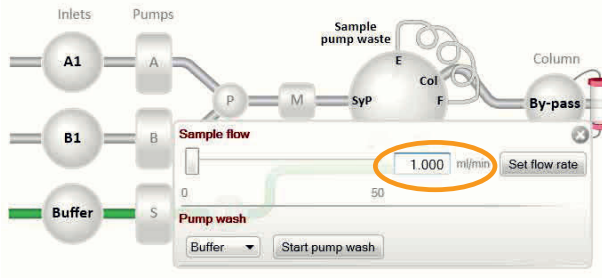
Purge Sample Pump

If the priming was done thoroughly and the final buffer was drawn all the way into the syringe and the validation of the priming showed that there was no air left in the pump it is not necessary to purge the sample pump.

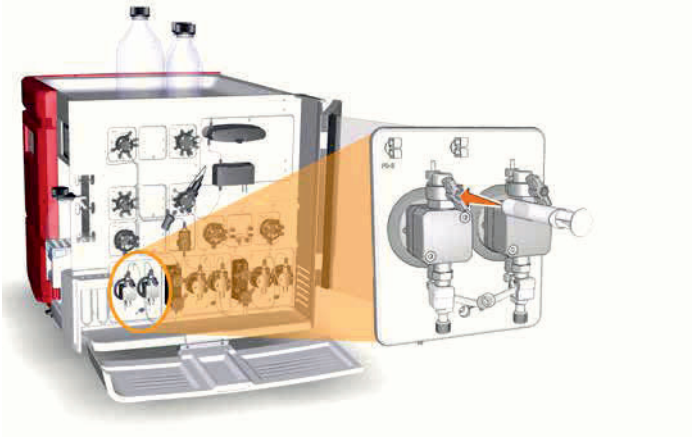
However, if the pressure signal indicated air bubbles left in the pump, follow the instruction below to purge both the pump heads of the sample pump.

Step	Action
1	Make sure that all sample inlet tubing that is to be used during the method run is immersed in the correct buffers.
2	Make sure that the waste tubing connected to injection valve port **W2** is immersed in a waste vessel.
3	Open the ***System Control*** module.
4	In the ***Process Picture***: • Click the ***Injection valve*** icon, and then click ***Sample pump waste***.  *Result:* The injection valve switches to waste position. This is necessary to achieve a low back pressure during the purge procedure.

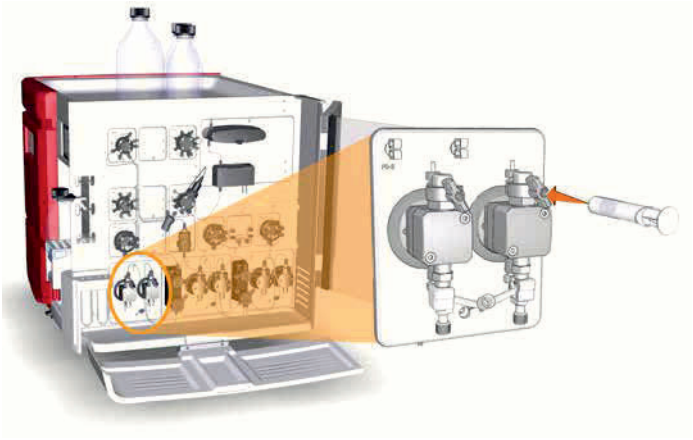
Step	Action
5	In the ***Process Picture***: • Click the ***Sample inlet*** icon, then click ***Buffer***. • Click the ***Sample pump*** icon: Set the ***Sample flow*** to 1.0 ml/min for ÄKTA avant 25 or 5.0 ml/min for ÄKTA avant 150.



	• Click ***Set flow rate***. *Result:* The sample pump flow starts.
6	Connect a 25 to 30 ml syringe to the left purge valve of the sample pump. Make sure that the syringe fits tightly into the purge connector.



7	Open the purge valve by turning it counter-clockwise about three-quarters of a turn. Draw 5 to 10 ml of liquid slowly into the syringe with a rate of about 1 ml/s.
8	Close the purge valve by turning it clockwise. Disconnect the syringe and discard its contents.

Step	Action
9	Connect the syringe to the right purge valve on the sample pump, and repeat step 6 to 8.
	
10	Check that there is no air left in the pump by following the instructions in *Validate prime or purge of System Pump A or B or Sample Pump on page 75*.

End the run

Click the ***End*** button in the ***System Control*** toolbar to end the run.

4.5.3 Prime Q inlets

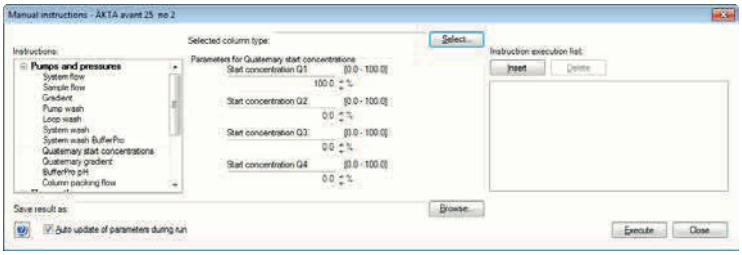
Overview

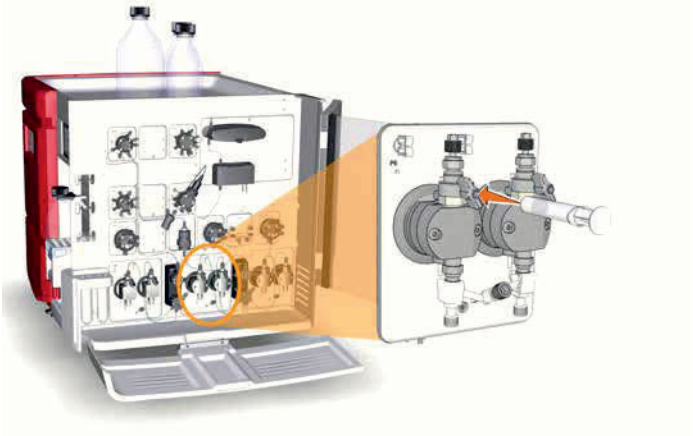
The procedure consists of the following stages:

Stage	Description
1	Prime all Q inlet tubing.
2	Validate priming of Q inlet tubing.
3	Purge Quaternary Valve and the system pumps if pressure signal indicates air bubbles.
4	Validate purge of Quarternary Valve and system pumps.
5	End the run.

Prime the Q inlets

Follow the instructions to prime the Q inlets.

Step	Action
1	Make sure that the pieces of inlet tubing marked **A1**, **B1** and **Q1-Q4** are immersed in the correct buffers. The **A1** and **B1** positions are used for pump synchronization and these lines should already be primed.
2	In the ***Manual instructions*** dialog box: • Select ***Pumps and pressures*** →***Quaternary start concentrations***. • Set ***Start concentration Q1*** to 100%. Make sure that the other start concentrations are set to 0%.  • Select ***Pumps and pressures*** →***System flow*** and set ***Flow rate*** to 0.01 ml/min.

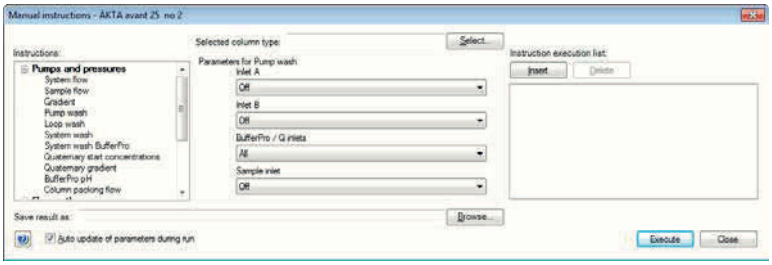
Step	Action
3	Connect a 25 to 30 ml syringe to one of the purge valves of either of the system pumps. Make sure that the syringe fits tightly into the purge connector. 
4	Open the purge valve by turning it counterclockwise about 3 quarters of a turn. Draw 10 ml of liquid into the syringe. Check that the **Q1** inlet is filled with liquid.
5	Close the purge valve by turning it clockwise. Disconnect the syringe and discard its contents.
6	Repeat steps 2 to 5 for **Q2**, **Q3** and **Q4** respectively by setting the respective ***Quaternary start concentration*** to 100%. ***Tip:*** *The inlet tubing that is immersed in distilled water should be the last piece of inlet tubing to be primed.* ***Tip:*** *If you will perform a BufferPro run, end with either* **Q1** *or* **Q2**.
7	Check that there is no air left in the pump by following the instructions in *Validate prime or purge of System Pump A or B or Sample Pump on page 75*. If air bubbles are indicated, follow the instructions in *Purge Quaternary Valve and the system pumps, on page 83*.

Purge Quaternary Valve and the system pumps

If the priming was done thoroughly and the final buffer was drawn all the way into the syringe and the validation of the priming showed that there was no air left in the pump it is not necessary to purge Quaternary Valve and the system pumps.

However, if the pressure signal indicated air bubbles left in the valve or the pump, follow these instructions to purge Quaternary Valve, System Pump A and System Pump B. Note that both pump heads of each system pump have to be purged.

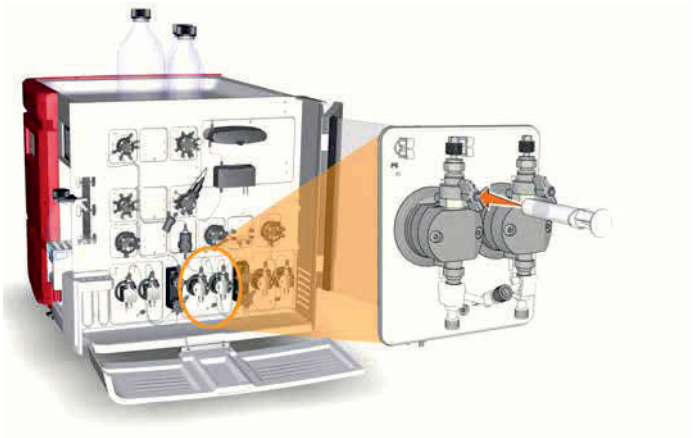
Step	Action
1	In the ***Manual instructions*** dialog box: • Select ***Pumps and pressures*** →***Pump wash***, and click ***All*** in the ***BufferPro / Q inlets*** list.



Result:

A simultaneous pump wash of all the Q inlets is started. This will remove air from Quaternary Valve.

2	Wait until the pump wash is completed. • Select ***Pumps and pressures*** →***Quaternary start concentrations***. • Set ***Start concentration Q1*** to 100%. Make sure that the other start concentrations are set to 0%.
3	Select ***Pumps and pressures*** →***System flow*** and set ***Flow rate*** to 0.01 ml/min.
4	Connect a 25 to 30 ml syringe to the left purge valve of the selected system pump. Make sure that the syringe fits tightly into the purge connector.



Step	Action
5	Open the purge valve by turning it counterclockwise about 3 quarters of a turn. Draw 10 ml of liquid slowly into the syringe with a rate of about 1 ml per second.
6	Close the purge valve by turning it clockwise. Disconnect the syringe and discard its contents.
7	Repeat steps 3 to 5 for the other three purge valves of the system pumps to get rid of air in all pump heads. Keep the system flow running during this procedure.
8	Check that there is no air left in the pump by following the instructions in *Validate prime or purge of System Pump A or B or Sample Pump on page 75*.

End the run

Click the ***End*** button in the ***System Control*** toolbar to end the run.

4.6 Performance tests

Introduction

Before taking the ÄKTA avant instrument into use, run performance tests to check the function of the equipment. See *ÄKTA avant User Manual* for further instructions.

5 Prepare the system for a run

About this chapter

This chapter describes the preparations necessary before starting a run.

In this chapter

5.1 Before you prepare the system

Introduction

It is important to prepare the system in accordance with the settings in the method to be run. Before preparing the system, verify the settings in the ***Method Editor*** and make sure that all accessories to be used are available.

WARNING

- Do not use ÄKTA avant if it is not working properly, or if it has suffered any damage, for example:
  - damage to the power cord or its plug
  - damage caused by dropping the equipment
  - damage caused by splashing liquid onto it
- Always use appropriate Personal Protective Equipment (PPE) during operation and maintenance of this product.
- Do not use any accessories not supplied or recommended by Cytiva.
- **Fire Hazard**. Before starting the system, make sure that there is no leakage.

Checklist

Prepare the instrument and accessories according to the following information from the ***Method Editor***:

- which valve ports to use for inlets and outlets
- which column type to use
- which column position to use
- which buffers and samples to prepare
- which sample application technique to use
- make sure that the the pH electrode is connected, if applicable
- which cassettes with corresponding deep well plates and/or tubes to use in the fraction collector, if applicable
- if it is a reversed phase chromatography (RPC) run, make sure to follow the safety precautions below

WARNING

When using flammable liquids with the ÄKTA avant instrument, follow these precautions to avoid any risk of fire or explosion.

- **Fraction collector**. Do **not** fractionate flammable liquids in the built-in fraction collector. When running RPC methods, collect fractions through the outlet valve or the optional external Fraction collector **F9-R**.
- **RPC runs with 100% acetonitrile and system pressure above 5 MPa (50 bar) in ÄKTA avant 25** . Always replace the green PEEK tubing between the used system pump and the pump pressure monitor with orange PEEK tubing, i.d. 0.5 mm, before running RPC with 100% acetonitrile. Set the system pressure alarm to 10 MPa (100 bar).
- **RPC runs with 100% acetonitrile in ÄKTA avant 150** . Always replace the beige PEEK tubing between the used system pump and the pump pressure monitor before running RPC with 100% acetonitrile. Replace it with green PEEK tubing, i.d. 0.75 mm.

5.2 Prepare the flowpath

Introduction

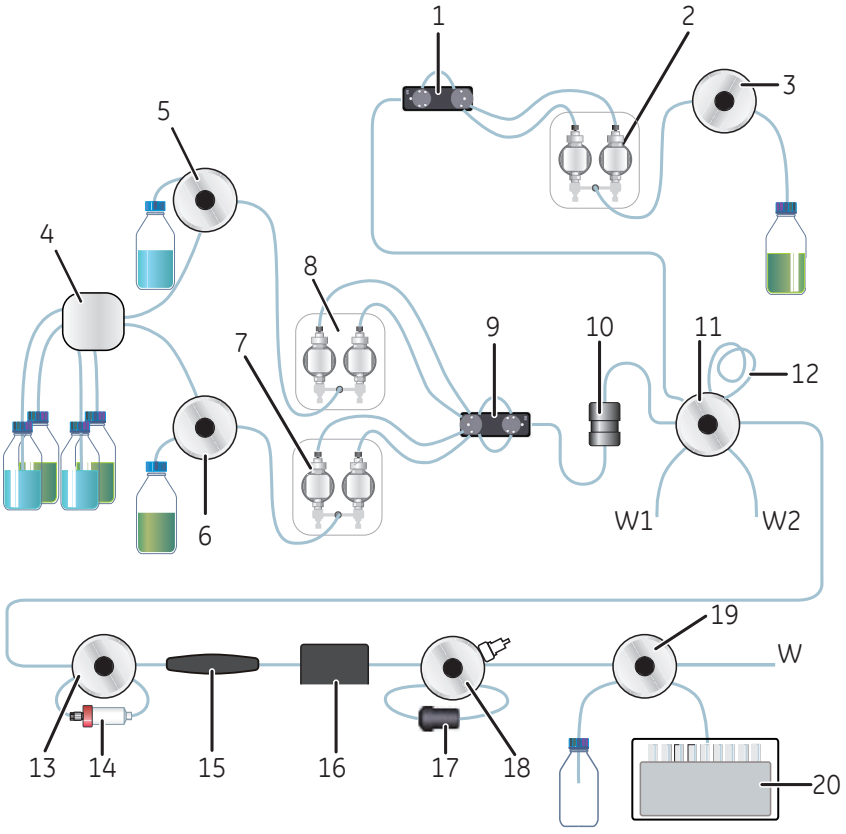
The flow path contains tubing, valves, pumps and monitors. This section gives an overview of the flow path and describes how to prepare the flow path before a run.

CAUTION

- **Fasten bottles and cassettes.** Always fasten bottles and cassettes to the rails at the front and side panel. Use appropriate holders for bottles. Shattered glass from falling bottles may cause injury. Spilled liquid may cause fire hazard and personal injury.
- **Max. weight on buffer tray**. Do not place containers with a volume of more than 10 liters each on the buffer tray. The total allowed weight on the buffer tray is 40 kg.
- **Avoid spillage and overflow.** Make sure that the system is prepared according to the settings in the method to be run. For example make sure that the waste tubing is inserted in an appropriate waste container and secured in place.

Illustration of the flow path

The following illustration shows an overview of the standard flow path.



Part	Description
1	Pressure monitor
2	Sample pump
3	Sample inlet valve
4	Quaternary valve
5	Inlet valve A
6	Inlet valve B
7	System pump B
8	System pump A
9	Pressure monitor

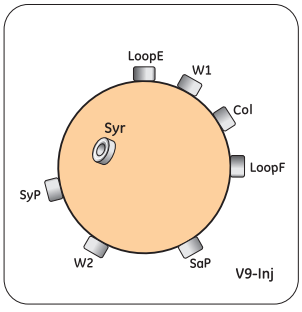
Part	Description
10	Mixer
11	Injection valve
12	Sample loop or Superloop™
13	Column valve
14	Column
15	UV monitor
16	Conductivity monitor
17	Flow restrictor
18	pH valve with pH monitor
19	Outlet valve
20	Fraction collector

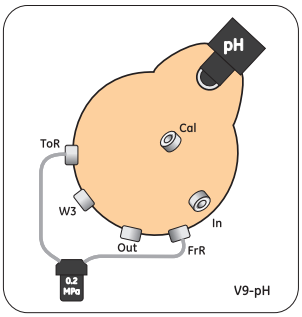
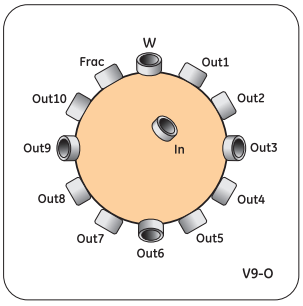
Prepare the inlet tubing

Connect inlet tubing to the inlet ports that are to be used, and immerse all inlet tubing that is to be used during the method run in the correct buffers.

Waste ports

The following table shows the waste ports of Injection Valve, pH Valve and Outlet Valve.

Valves and ports	Illustrations
Injection Valve (label **V9-Inj** and **V9H-Inj**) Waste ports: • **W1, W2**	

Valves and ports	Illustrations
pH Valve (label **V9-pH** and **V9H-pH**) Waste port: • **W3**	
Outlet Valve (label **V9-O** and **V9H-O**) Waste port: • **W**	

Prepare the waste tubing

Make sure that the waste tubing is prepared according to the instructions in *Section 4.2.3 Prepare waste tubing, on page 58*.

Prepare the outlet tubing

Connect outlet tubing to the outlet ports of the outlet valve that are to be used during the run. If a fraction collector is to be used, make sure that tubing is connected between the outlet valve **Frac** port and the fraction collector, and prepare the fraction collector. Otherwise, immerse the outlet tubing in suitable tubes or flasks.

Plug unused valve ports

It is recommended to plug all unused valve ports with stop plugs before starting a run. See *User Manual 29119969* for information about connectors.

5.3 Prime buffer inlets and purge system pumps

Introduction

Before you start the system pumps, it is important to do the following:

- Prime the inlets (fill the buffer inlets with liquid).
- Purge the system pumps (remove air from the pump heads).

For instructions on how to prime the inlets and purge the system pumps, see *Section 4.5 Prime inlets and purge pump heads, on page 69*.

5.4 Connect a column

Introduction

This section describes how to connect a column to the instrument using a column holder and without introducing air into the flow path. Several types of column holders are available for the ÄKTA avant instrumen, see ÄKTA avant User Manual.

WARNING

Before connecting a column, read the instructions for use of the column. To avoid exposing the column to excessive pressure, make sure that the pressure limit is set to the specified maximum pressure for the column.

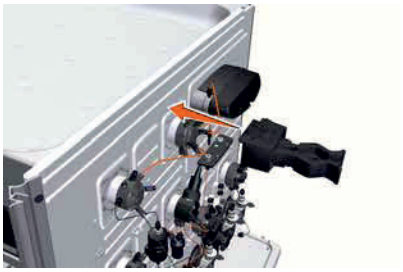
Methods automatically include pressure alarms based on the specifications of the chosen column type. However, when running manual runs you have to set the pressure limits yourself. Also, to protect the column media, special settings are needed. See *Section 5.5 Set pressure alarms, on page 99* for more information on pressure alarms.

Note: *Do not overtighten when connecting columns. Overtightening might rupture the connectors or squeeze the tubing and thereby result in high back pressure.*

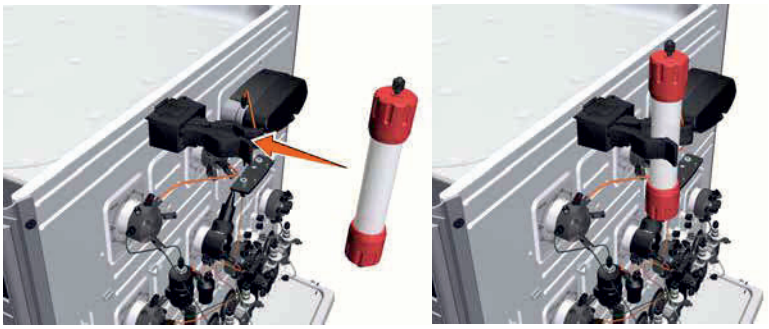
Attach a column holder and connect a column

Follow the instructions to connect a column to the instrument. Always use a column holder. The column is connected to two opposite parts of the column valve, using appropriate tubing and connectors.

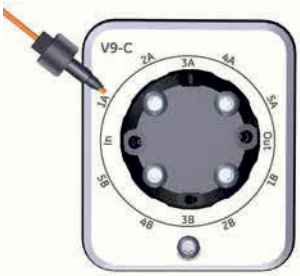
Step	Action
1	Attach an appropriate column holder to the rail on the instrument.



Step	Action
2	Attach the column to the column holder.

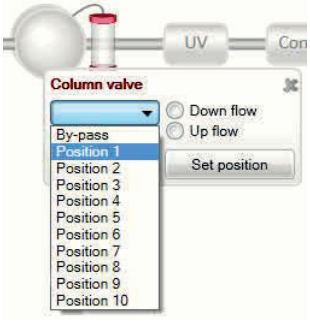


3	Connect a suitable tubing to a column valve port, for example port **1A** if column position 1 was chosen in the method to be run.



4	In the ***Process Picture***:

- Click the ***Column valve*** icon.
- Click, e.g., ***Position 1*** and ***Down flow***.



- Click **Set position**

Result:

The column valve switches to position ***1***.

Step	Action
5	In the ***Process Picture***: • Click the ***System pumps*** icon. • Enter a low ***System flow*** (e.g., 0.2 ml/min). • Click ***Set flow rate***.  *Result:* A system flow of 0.2 ml/min starts.
6	When buffer leaves the tubing on port **1A** (if port **1A** was chosen in the method to be run) in a continuous mode and the top part of the column is filled with buffer, connect the tubing to the top of the column. 
7	Connect a piece of tubing to the bottom of the column. 

Step	Action
8	When buffer leaves the tubing at the bottom of the column in a continuous mode, connect this piece of tubing to the column valve. Use the port opposite to the one already connected to the column, in this example port **2B**. 
9	Click the ***End*** button in the ***System Control*** toolbar to end the run. 

5.5 Set pressure alarms

Introduction

The columns can be protected by two different types of pressure alarms:

- The pre-column pressure alarm protects the column hardware.
- The delta-column pressure alarm protects the column media.

The column valves **V9-C** and **V9H-C** have built-in pressure sensors that automatically measure the pre-column and delta-column pressure.

See the instructions in the next topic to set the pressure alarm for the column to be used in the run and, if applicable, to set the parameters for the tubing dimensions.

Note: *Remember to lower the system pressure alarm and sample pressure alarm if the optional UV Monitor* **U9-L** *and/or the optional second Conductivity Monitor* **C9** *is used on the high pressure side in the system (before the column[s]).*

NOTICE

UV and conductivity flow cells on the high pressure side.
When placing UV and/or conductivity flow cells on the high pressure side of the column, the UV flow cell has a maximum pressure limit of 2 MPa (20 bar) and the conductivity flow cell has a maximum pressure limit of 5 MPa (50 bar).

Pre-column pressure alarms

It is important that the pre-column pressure alarm is set during all runs where a column is used. The pressure alarm can be set in: the method to be run, the ***System Settings*** dialog box, or during a manual run.

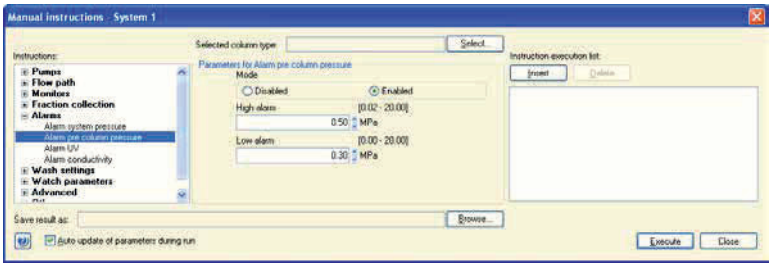
Pre-column pressure alarm limits are automatically set in the method when a column from the column list is selected in the method. Refer to *UNICORN Method Manual* for more information on pressure alarms.

Set pressure alarms

Pressure alarm limits may be set manually in ***System Control***. The example below describes how to set the high pressure limit for the column. Other alarms are set in a corresponding way.

Step	Action
1	In the ***System Control*** module, on the ***Manual*** menu, click ***Execute Manual Instructions*** . *Result:* The ***Manual instructions*** dialog box opens.

Step	Action
2	In the ***Instructions*** box, select ***Alarms*** →***Alarm pre column pressure***.



3	Click ***Enabled*** in the ***Mode*** field.
4	• Enter the high pressure limit in the ***High alarm*** box. • Click ***Execute***.

5.6 Calibrate the pH monitor

Introduction

If pH will be measured during the chromatographic run, the pH monitor should be calibrated before the run is started. Use two pH calibration buffers with a difference of at least one pH unit. Preferably use a pH standard buffer pH 4 or 7 as the first calibration point, and a pH standard buffer close to the lowest or highest pH you need to measure as your second point. Allow the buffers to reach the operating temperature before use.

Note: *Do not run a system flow during pH calibration.*

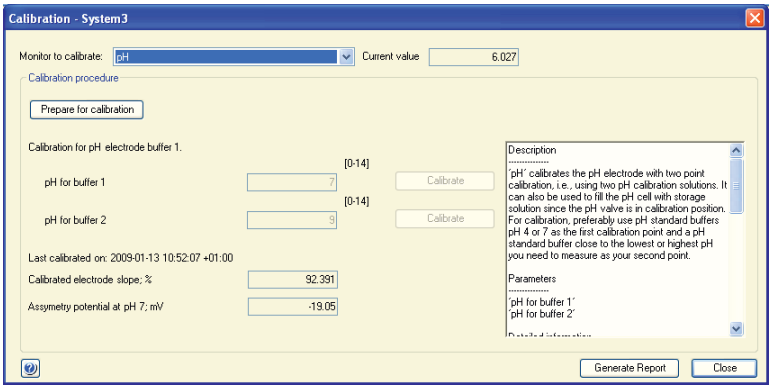
Calibrate the pH monitor

CAUTION

pH electrode. Handle the pH electrode with care. The glass tip may break and cause injury.

Follow the instructions to perform the calibration.

Step	Action
1	Open the ***System Control*** module. On the ***System*** menu, click ***Calibration***. *Result:* The ***Calibration*** dialog box opens.
2	Select ***pH*** in the ***Monitor to calibrate*** list.
3	Click ***Prepare for calibration***. *Result:* The pH valve switches to the calibration position.
4	Enter the pH of the first pH standard buffer in the ***pH for buffer 1*** box.



Step	Action
5	Fill a syringe with approximately 10 ml of the first pH standard buffer. Connect the syringe to the Luer connector in pH valve port **Cal**, and inject the buffer.
6	When the ***Current value*** is stable, click ***Calibrate***.
7	Wash the pH flow cell by injecting water into pH valve port **Cal** using a new syringe.
8	Enter the pH of the second pH standard buffer in the ***pH for buffer 2*** box.
9	Repeat steps 5 to 6 using the second pH standard buffer. *Result:* Calibration date and time are displayed in the dialog box, and also values for ***Calibrated electrode slope*** and ***Asymmetry potential at pH 7***.
10	Is the ***Calibrated electrode slope*** ≥ 80% and the ***Asymmetry potential at pH 7*** inside the interval ±60 mV? • If Yes: Click ***Close*** to switch the pH valve back to the default position, and to close the ***Calibration*** dialog box. • If No: Clean the pH electrode, and repeat the calibration procedure. If this does not help, replace the electrode. For information about cleaning and replacing the pH electrode, see *ÄKTA avant User Manual, Chapter Maintenance*.

5.7 Prepare the built-in fraction collector

Introduction

This section describes how to prepare the built-in fraction collector. For detailed information regarding the types of deep well plates, tubes and cassettes, see *ÄKTA avant User Manual*.

WARNING

Fraction collector. Do **not** fractionate flammable liquids in the built-in fraction collector. When running RPC methods, collect fractions through the outlet valve or the optional external Fraction collector **F9-R**.

Available cassettes, trays and racks

The following Cassettes and racks are available:

- Cassette 3 ml tubes (for 40 tubes)
- Cassette 5 ml tubes (for 40 tubes)
- Cassette 8 ml tubes (for 24 tubes)
- Cassette 15 ml tubes (for 15 tubes)
- Cassette 50 ml tubes (for 6 tubes)
- Cassette for deep well plate (24, 48, 96 wells)
- Cassette tray (for six cassettes)
- Rack for 50 ml tubes (for 55 tubes)
- Rack for 250 ml bottles (for 18 bottles)

Workflow for preparing the built-in fraction collector

Before starting to prepare the built-in fraction collector, check the fractionation settings in the method to be run. Perform the steps described below according to the settings in the method.

Step	Operator actions	Reference to instructions
1	Insert the cassette tray or a rack for tubes or bottles.	See *Prepare and insert the cassette tray* below.
2	Change the ***System Settings*** in UNICORN to set the fractionation mode and other settings for fraction collection.	See *UNICORN System Control Manual*. Available ***System Settings*** are described in *ÄKTA avant User Manual*.

Prepare cassettes and insert the cassette tray

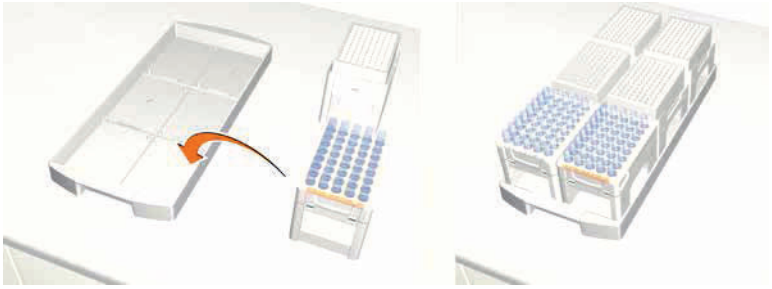
Follow the instructions to prepare the fraction collector before a run.

Step	Action
1	If you are to use cassettes with the QuickRelease function, first open the cassettes.

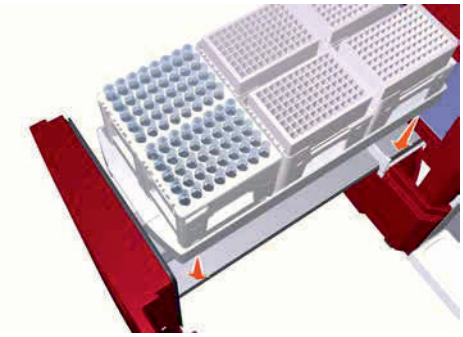
2	Place the tubes and deep well plates in the cassettes. Make sure that the deep well plates are rotated so that the well marked **A1** is positioned above the **A1** marking on the cassette.

Step	Action
3	Close the cassettes that have the QuickRelease function.

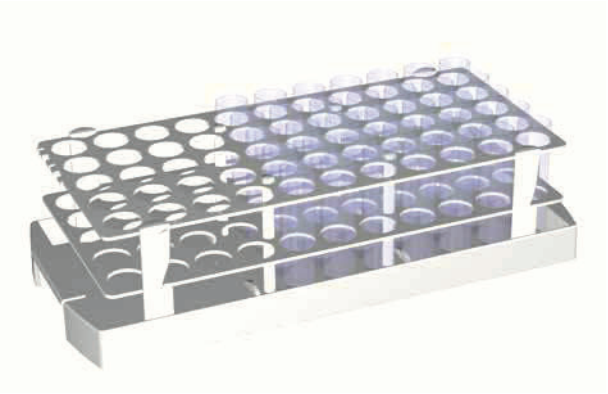
4	Place the cassettes on the cassette tray. Make sure that the cassette type code (see the illustration) faces the front of the tray marked with the Cytiva monogram.



5	Open the fraction collector drawer by pressing the handle upwards, and pulling out the drawer.

Step	Action
6	Place the cassette tray on the tray support of the fraction collector drawer. Make sure that the front of the tray (marked with the Cytiva monogram) faces the front of the drawer and is hooked onto the two pins. 
7	Close the drawer. Make sure that it snaps into closed position. *Result:* After the door has been closed, the fraction collector performs a Full scan of the cassette type code of each cassette to identify the cassette types, see *Cassette and tray identification on page 107*.

Prepare a rack for 50 ml tubes or 250 ml bottles

Step	Action
1	Place 50 ml tubes or 250 ml bottles in the corresponding rack (rack with 50 ml tubes shown in illustration). 

Step	Action
2	Open the fraction collector drawer by pressing the handle upwards, and pulling out the drawer.

3	Place the rack on the tray support of the fraction collector drawer. Make sure that the front of the rack (marked with the Cytiva monogram) faces the front of the drawer and is hooked onto the two pins.

4	Close the drawer. Make sure that it snaps into closed position.

Cassette and tray identification

When the door of the fraction collector is closed automatic scanning is performed. There are two types of scanning procedures:

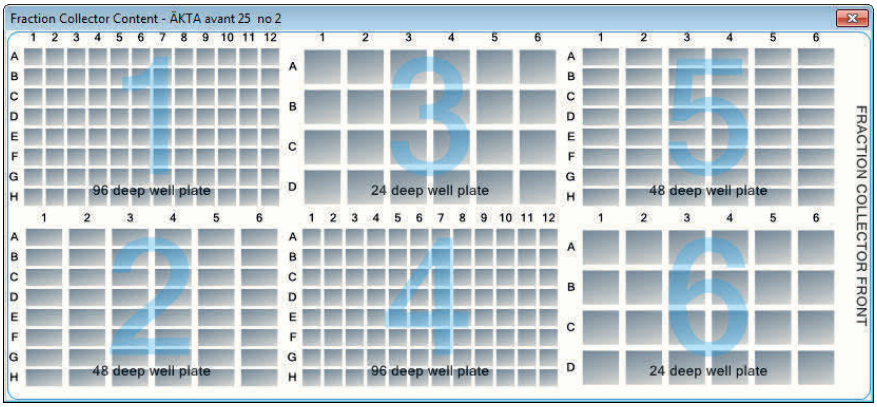
- ***Full scan***: Scanning of cassette type codes to determine which types of cassettes are used, and scanning of rows and columns in deep well plates to identify which types of plates are used (24, 48, or 96 wells). Full scan is performed only when the system is in state ***Ready***.

- ***Quick scan***: Scanning of cassette type codes to determine which type of cassettes are used. Quick scan is performed during the run to ensure that correct cassettes are placed in the fraction collector.

> **WARNING**
>
> **Moving parts in fraction collector**. Do not open the fraction collector drawer when the fraction collector is active. If you need to access the fraction collector, press ***Pause***, and make sure that the movement has stopped before opening the drawer.

View fraction collector content

To view the content of the Fraction collector, open the ***System control*** module. On the ***View*** menu, click ***Fraction Collector Content***.



5.8 Prepare for a run at cold temperature

Introduction

To fit the ÄKTA avant instrument in a cold cabinet, the foldable door and pump cover can be removed. For instructions, see *ÄKTA avant User Manual*. When using the instrument in a cold room or cold cabinet, make sure to follow the precautions listed in the next topic.

Precautions concerning runs in a cold temperature

NOTICE

- **Avoid condensation**. If ÄKTA avant is kept in a cold room, cold cabinet or similar, keep it switched on in order to avoid condensation.
- **Avoid overheating.** If ÄKTA avant is kept in a cold cabinet and the cold cabinet is switched off, make sure to switch off ÄKTA avant and keep the cold cabinet open to avoid overheating.
- **Place the computer in room temperature**. If the ÄKTA avant instrument is placed in a cold room, use a cold room compatible computer or place the computer outside the cold room and use the Ethernet cable delivered with the instrument to connect to the computer.

Note: *When the instrument is kept in a cold room, it is important to tighten all tubing connectors, also the inlet manifold connectors. Otherwise air might get into the flow path.*

Note: *Make sure that the instrument, buffers and sample have had time to reach the ambient temperature. When the instrument has reached the ambient temperature, calibrate all pressure sensors.*

Tip: *When runs are performed in a cold cabinet, make sure to adjust the target temperature of the built-in fraction collector temperature control function. The target temperature is 20°C by default. Settings for the temperature control function can be edited in the* ***System Settings*** *dialog box of* ***System Control****, or in the* ***Text Instructions*** *pane in* ***Method Editor****.*

6 Run a method

About this chapter

This chapter describes how to start up and run a method, and also how to handle the system after the run.

In this chapter

6.1 Before you start

Introduction

Before starting a run, it is necessary to read and understand the information in this section and to perform the checks listed in the next topic.

WARNING

- Always use appropriate Personal Protective Equipment (PPE) during operation and maintenance of this product.
- **Hazardous substances.** When using hazardous chemicals, take all suitable protective measures, such as wearing protective clothing, glasses and gloves resistant to the substances used. Follow local and/or national regulations for safe operation and maintenance of the product.
- **High pressure**. The product operates under high pressure. Wear protective glasses and other required Personal Protective Equipment (PPE) at all times.

Checklist

Make sure that the system is correctly prepared:

- Prepare the system according to the settings in the method to be run.
- Select a suitable column for the application.
- Immerse the buffer inlet tubing in correct buffer vessels.
- Immerse all waste tubing in appropriate waste vessels (consider vessel size, placement and material).
- Verify that no tubing is twisted and that the flow path is free from leakage.

Warnings concerning use of hazardous substances

WARNING

- **Hazardous chemicals during run.** When using hazardous chemicals, run ***System CIP*** and ***Column CIP*** to flush the entire system tubing with distilled water, before service and maintenance.
- **Hazardous biological agents during run.** When using hazardous biological agents, run ***System CIP*** and ***Column CIP*** to flush the entire system tubing with bacteriostatic solution (e.g. NaOH) followed by a neutral buffer and finally distilled water, before service and maintenance.

Hold, pause or stop the run

At the end of a method the run stops automatically. All pumps stop and an acoustic end signal sounds and ***End*** is displayed in the ***Run Log***.

To interrupt a method during a run you may click the ***Hold***, ***Pause*** or ***End*** buttons in ***System Control***. A held or paused method run can be resumed by clicking the ***Continue*** button. See the instructions in the following table.

If you want to...	then...
temporarily hold the method, with current flow rate and valve positions sustained	click the ***Hold*** button. 
temporarily pause the method, and stop all pumps	click the ***Pause*** button. 
resume, for example, a held or paused method run.	click the ***Continue*** button.  ***Note:*** *An ended method cannot be continued.*
permanently end the run	click the ***End*** button. 

Note: *When ending a method run in advance, it is possible to save the partial result.*

6.2 Applying the sample

Introduction

A number of different sample application techniques are available. The sample can be applied either directly onto the column using the sample pump, or via a loop. A loop can be filled either manually or by using the sample pump.

This section describes sample application using a syringe to manually fill a sample loop.

Sample application stages

The two stages of the sample application are described in the following table. For detailed instructions and information regarding the different sample applications techniques, see *ÄKTA avant User Manual*.

Stage	Description
Load	The sample loop is filled with sample.
Inject	The sample is injected onto the column.

How to fill a sample loop

Follow the instructions to fill the sample loop with sample.

Step	Action
1	Connect a suitable sample loop to the Injection Valve ports **LoopF** (fill) and **LoopE** (empty).
	
2	Fill a syringe with sample.

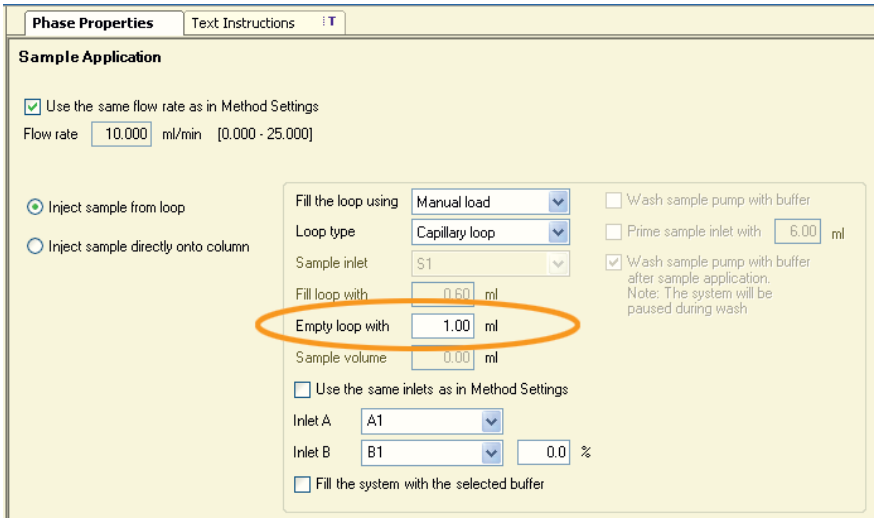
Step	Action
3	Connect the syringe to the Injection Valve port **Syr**.

4	Load sample into the sample loop. To avoid sample loss due to siphoning, leave the syringe in the port until the sample has been injected onto the column during the run. ***Tip:*** *It is recommended to overload the loop to make sure that the loop is completely filled. Excess of sample will leave the valve through port* **W1**.

Sample application/injection through a sample loop

A sample loop is manually filled with sample using a syringe connected to the Injection Valve. During the method run, the sample is automatically injected onto the column. The loop is emptied and washed out using buffer from the system pumps.

The total buffer volume to be used for emptying and washing the sample loop is set in the ***Phase Properties*** tab of the ***Sample Application*** phase in the ***Empty loop with*** box.



Tip: *Empty the sample loop with a buffer volume that exceeds the volume of the loop. This will ensure that the loop is completely emptied.*

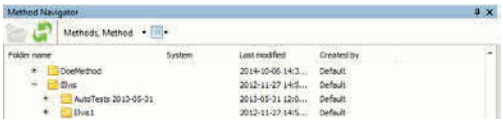
6.3 Start a method run

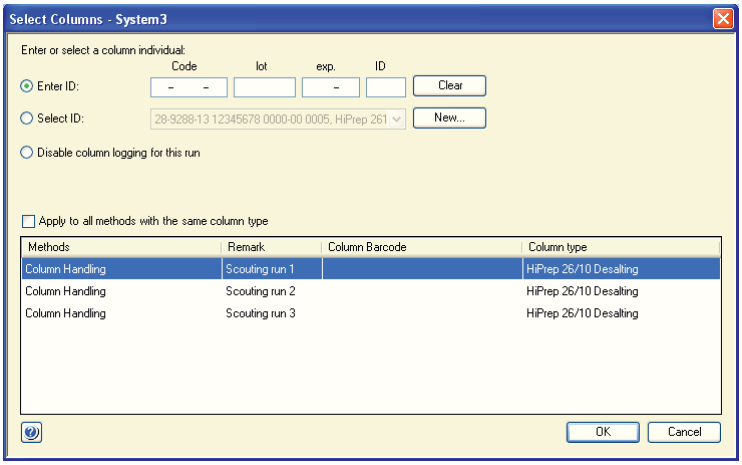
Introduction

This section describes how to start a run using a previously created method. If ***Column Logbook*** was enabled during installation of the software, registration and selection of individual columns is possible at method start. For further information on method creation, please refer to *UNICORN Method Manual*.

Choose and start a method

The following instructions describe how to open a method and start a run.

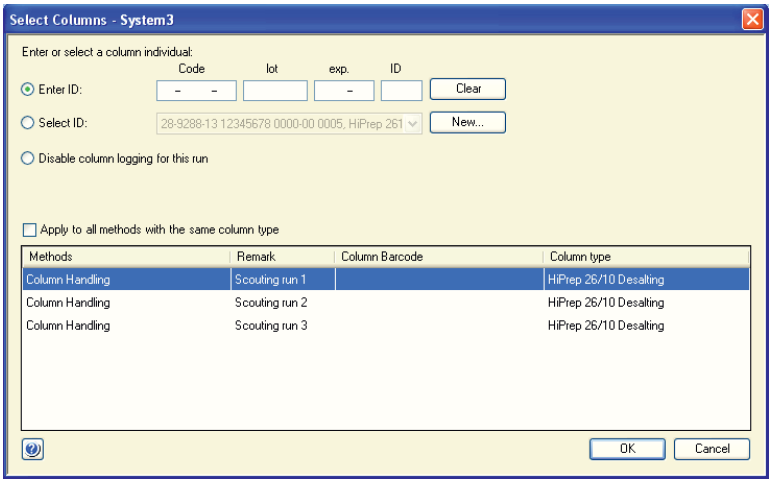
Step	Action
1	Open the ***System Control*** module and click the ***Open Method Navigator*** button.  *Result:* The ***Method Navigator*** pane opens. 
2	Select the method to run, and click the ***Run*** button.  *Result:* The ***Start Protocol*** dialog box opens.
3	Step through the displayed pages in the ***Start Protocol***, add requested input and make appropriate changes if necessary. Click ***Next***.

Step	Action
4	Click ***Start*** on the last page of the ***Start Protocol***. *Result:* • If column logging was chosen at installation of UNICORN and a column type was selected at method creation, the ***Select Columns*** dialog box opens. Continue with the steps outlined in the next topic.  • If column logging was *not* chosen at installation of UNICORN and/or *no* column type was selected at method creation, the run starts directly.

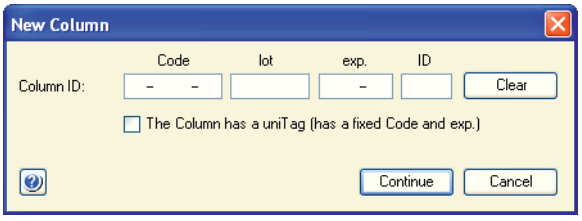
Register a column and start a run

The following instructions describe how to register a column and start a run.

Step	Action
1	Is the column to be used already registered? • If No, continue to step 2. • If Yes, continue to step 5.



2	In the ***Select Columns*** dialog box, click ***New***. *Result:* The first ***New Column*** dialog box opens.



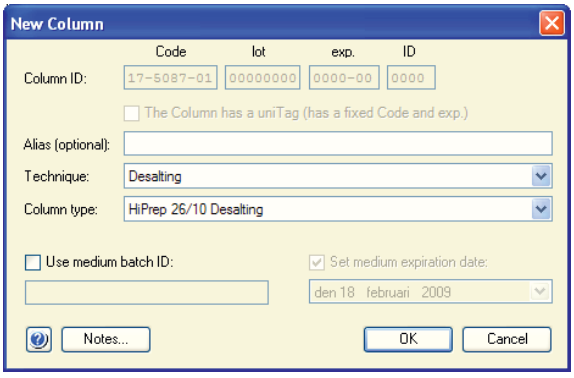
Step	Action
3	Register the column using the Barcode Scanner 2-D as follows:

- Make sure that the pointer is placed in the first position of the ***Code*** box.
- Point the Barcode Scanner 2-D towards the data matrix tag on the column.
- Press and hold the trigger to create a beam.
- When the scanner beeps, the column ID is registered and displayed in the dialog box.

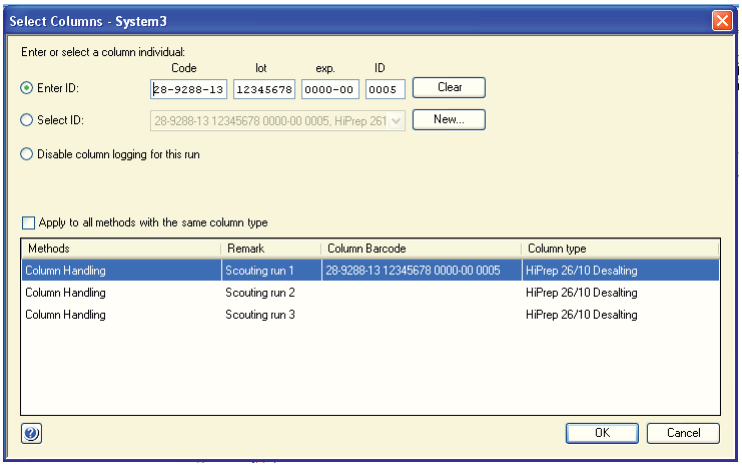
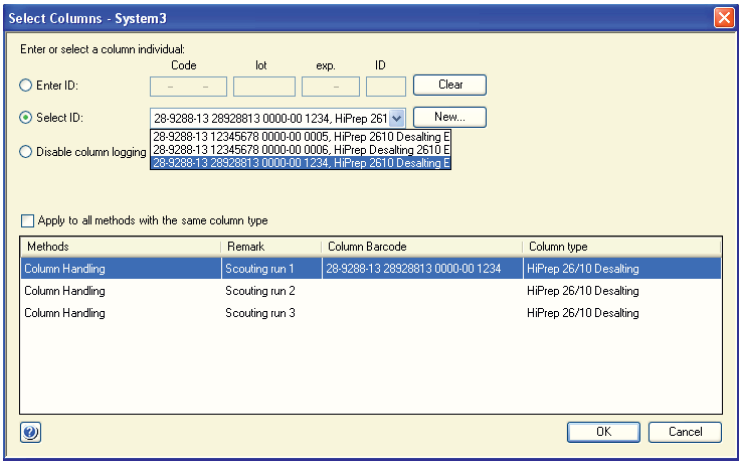
- Alternatively, manually enter the column ID, that you find on the column label, in the dialog box using your keyboard.
- Click ***Continue***.

Result:

The expanded ***New Column*** dialog box opens.



Step	Action
4	In the expanded ***New Column*** dialog box: • Enter an column alias in the ***Alias*** box (optional). • Click a chromatography technique on the ***Technique*** menu. • Click a column type on the ***Column type*** menu. • Select the ***Set medium expiration date*** check box and click a date on the menu. • Click ***OK***. ***Tip:*** *Alias can be used for easy identification of a column.* *Result:* The entered information is saved and the dialog box closes.

Step	Action
5	In the ***Select Columns*** dialog box: • Click ***Enter ID***. • Use the Barcode Scanner 2-D (see step 3) to enter the column ID.  • Alternatively, click ***Select ID*** and click the column individual to be used in the run from the menu.  • Click ***OK***. *Result:* The run starts. All necessary actions occur automatically according to the method, including ending of the run.

6.4 Monitor the run

Introduction

You may follow the on-going method run in the ***System Control*** module. The current system status is shown in the ***System state*** panel in the ***Run Data*** pane. For example, it may state ***Run***, ***Wash*** or ***Hold***. The same information is also shown in the instrument display.

- Selected curves are shown in the ***Chromatogram*** pane.
- All registered actions during the run are displayed in the ***Run Log*** pane.
- The current flow path is shown in the ***Process Picture*** pane.

For details on the ***System Control*** interface, see *Section 3.2.2 The System Control module, on page 38*.

Monitor the run

To interrupt a method during a run you may click the ***Hold***, ***Pause*** or ***End*** buttons in ***System Control***. A held or paused method run can be resumed by clicking the ***Continue*** button. See the following table.

If you want to...	then...
temporarily hold the method, with current flow rate and valve positions sustained	click the  button.
temporarily pause the method, and stop all pumps	click the  button.
resume, for example, a held or paused method run.	click the  button. ***Note:*** *An ended method cannot be resumed.*
permanently end the run	click the  button.

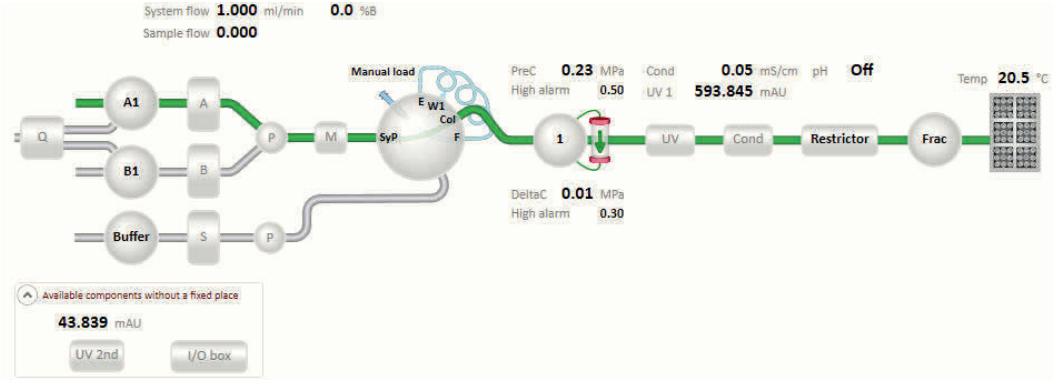
Note: *When ending a method run in advance, it is possible to save the partial result.*

Process Picture

The ***Process Picture*** displays the current flow path, run parameters and real-time data from monitors during a run. It also allows manual interactions with the system.

Tubing colors indicate flow path states, as shown in the following illustration and described in the following table.

Modules without a fixed place in the system are shown in a panel below the process picture (modules are called components in the process picture).

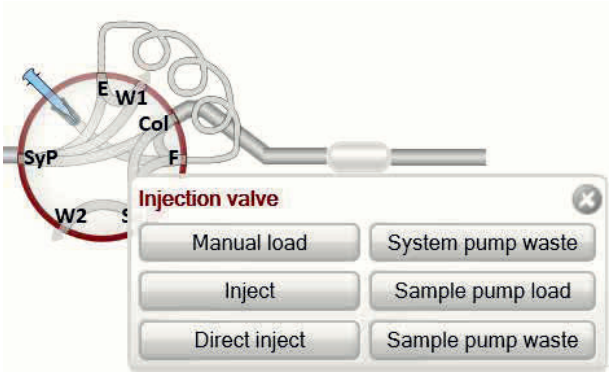


Color	Indication
Green	Open flow path with flow.
Grey	Closed flow path or an open path without flow.
Blue	Syringe port in loop open for manual injection.

Actions in the Process Picture pane

It is possible to interact with the ***Process Picture*** pane.

- To open a related instruction, click the component icon. The example below shows the pop-up toolbar for the ***Injection valve*** icon. Instructions can be given from the pop-up toolbar of each component icon.



- To display a detailed picture with explanations, for example for a valve, right-click the component icon and click ***Detailed picture***.

6.5 After run procedures

Introduction

This section describes how to clean the instrument and columns after a chromatographic run, and how to prepare the system for storage.

The instrument and the columns should be cleaned between the runs. This will prevent, for example, sample contamination, protein precipitation and column clogging. If the instrument is not going to be used for a couple of days or longer, the instrument, columns and the pH flow cell should be filled with storage solution. For further information about cleaning and maintenance procedures, see *Chapter 7 Maintenance, on page 127*.

Tip: *To clean and fill the instrument and columns with storage solution, use the **System CIP** and **Column CIP** methods. Either as separate, predefined methods or as phases included in a chromatographic method.*

WARNING

Corrosive chemicals during maintenance. If the system or column is cleaned with a strong base or acid, flush with water afterwards and wash with a weak neutral buffer solution in the last step or phase.

System cleaning

After a method run is completed, perform the following:

- Rinse the instrument with one or several cleaning solution(s) (e.g., NaOH, buffer solution or distilled water) using the ***System CIP*** method.
- If applicable, empty the fraction collector.
- Clean all spills on the instrument and on the bench using a moist tissue.
- Empty the waste vessel.
- Clean the manual injection port of the injection valve, see *ÄKTA avant User Manual* for detailed instructions.
- If applicable, clean the pH electrode manually and make sure to leave it in an appropriate buffer. See *ÄKTA avant User Manual* for detailed instructions.

System storage

If the instrument is not going to be used for a couple of days or longer, also perform the following:

- Fill the system and inlets with storage solution (e.g., 20% ethanol) using the ***System CIP*** method.

Column cleaning

After a method run is completed, perform the following:

- Clean the column with one or several cleaning solution(s) using the ***Column CIP*** method.

Column storage

If the column is not going to be used for a couple of days or longer, also perform the following:

- Fill the column with storage solution (e.g., 20% ethanol) using the ***Column CIP*** method.

pH electrode storage

If pH monitoring will not be used for a week or longer, perform one of the following actions:

- Inject new storage solution into the pH flow cell.
- Replace the pH electrode with the dummy electrode that is installed in the pH valve on delivery.

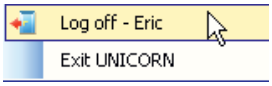
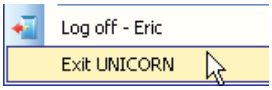
In the following situations, in order to increase the lifetime of the pH electrode, use the ***By-pass*** position and store the electrode in storage solution inside the pH flow cell:

- pH monitoring is not needed during the run.
- Organic solutions are used.
- Extremely acidic or extremely basic solutions are used.

For further information on how to prepare the pH electrode for storage, refer to *ÄKTA avant User Manual*.

Log off or exit UNICORN

Follow the instructions to log off or exit UNICORN. This can be performed from any of the UNICORN modules.

If you want to...	then...
log off UNICORN	on the ***File*** menu, click ***Log off***.  *Result*: All open UNICORN modules close and the ***Log On*** dialog box opens.
exit UNICORN	on the ***File*** menu, click ***Exit UNICORN***.  *Result*: All open UNICORN modules close.

Note: *If an edited method or result is open and not saved when you try to exit or log off UNICORN, you will see a warning. Click **Yes** to save, **No** to exit without saving, or **Cancel** to stay logged on.*

Shut down the instrument

Switch off the instrument by pressing the **Power** switch to the **O** position.

7 Maintenance

About this chapter

Regular maintenance is essential for reliable function and results. This chapter provides schedules for preventive maintenance that should be performed by the user of the ÄKTA avant instrument.

Instructions for replacing pump parts and cleaning pump head check valves are included in this chapter.

Refer to the *ÄKTA avant User Manual* for detailed instructions for maintenance and replacement of parts.

WARNING

Always use appropriate Personal Protective Equipment (PPE) during operation and maintenance of this product.

In this chapter

7.1 Maintenance program

Introduction

An overview of the preventive maintenance to be performed on the ÄKTA avant instrument is outlined in the following list. See *ÄKTA avant User Manual* for detailed information about the maintenance procedures.

Maintenance is divided into:

- Daily maintenance
- Weekly maintenance
- Monthly maintenance
- Bi-annual maintenance
- Maintenance when required

WARNING

Electrical shock hazard. All repairs should be done by service personnel authorized by Cytiva. Do not open any covers or replace parts unless specifically stated in the user documentation.

Periodic maintenance program

The following periodic maintenance should be performed by the user of the ÄKTA avant instrument.

Interval	Maintenance action
Daily	Calibrate the pH monitor
Weekly	Change pump rinsing solution
Weekly	Replace inline filter in the mixer
Weekly	Clean the fraction collector
Monthly	Check the flow restrictor
Bi-annual	Clean the UV flow cell
Bi-annual	Replace the pH electrode

Maintenance when required

The following maintenance should be performed by the user of the ÄKTA avant instrument when required. Refer to the *ÄKTA avant User Manual* for detailed instructions.

Maintenance action
Clean the instrument externally
Run System CIP (System cleaning-in-place)
Run Column CIP (Column cleaning-in-place)
Clean the fraction collector
Replace tubing and connectors
Storage of pH electrode
Clean the pH electrode
Clean the conductivity flow cell
Calibrate the conductivity monitor
Calibrate the UV monitor
Calibrate pressure monitors
Replace the mixer
Replace O-ring in mixer
Replace the UV flow cell
Replace the flow restrictor
Replace inlet filters
Wipe off excess oil from pump heads
Clean the check valves. See *Section 7.5 Clean pump head check valves, on page 148*
Replace check valves
Replace pump piston seal, O-rings, and rinse membrane. See *Section 7.2 Replace pump piston seal, O-rings, and rinse membrane of Pump P9 or Pump P9H, on page 130* and *Section 7.3 Replace pump piston seal, O-ring, and rinse membrane of Pump P9-S, on page 139*.
Replace pump pistons. See *Section 7.4 Replace pump pistons, on page 147*.
Replace pump rinsing system tubing
Replace valve modules

7.2 Replace pump piston seal, O-rings, and rinse membrane of Pump P9 or Pump P9H

Introduction

Follow the instructions to replace the O-rings, piston seal, and rinse membrane of pumps **P9**, **P9H A**, **P9H B**, or **P9H S**.

Note: *Always replace the O-rings, piston seals, and rinse membranes of both pump heads of a pump at the same time.*

Tip: *A sign of internal leakage is that the pump rinsing solution volume starts to increase.*

NOTICE

- Do not disassemble the pump head unless there is good reason to believe that there is an internal leakage. A sign of leakage is increasing pump rinsing solution volume. Always make sure that sufficient spare components are available before attempting to replace a spare part.
- **Replacing spare parts**. Read the instructions carefully. For example, some individual parts of the pump head can be assembled incorrectly. Check the orientation of each part before continuing with the next instruction.

Maintenance interval

Replace the O-rings, piston seals, and rinse membranes the pumps **P9**, **P9H A**, **P9H B**, and **P9H S** if they are damaged. After replacement, perform a run to break in the new piston seals.

NOTICE

Advanced maintenance. Read the instruction carefully before disassembly of the pump head.

Required material

The following materials are required:

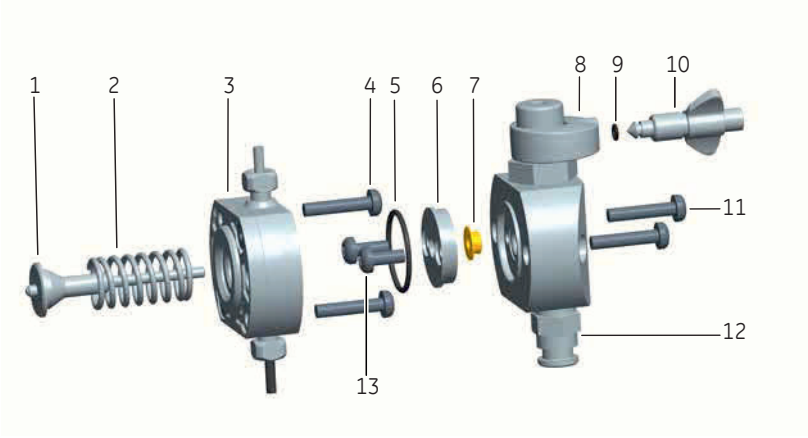
- Adjustable wrench
- For Pump **P9**: Star screwdriver, T20
- For Pump **P9H**: Star screwdriver, T10 and T20
- Ultrasonic bath
- Ethanol, 20%
- For Pump **P9**: Tubing giving a back pressure of 6 to 8 MPa (60 to 80 bar).

- For Pump **P9H**: Tubing giving a back pressure of 2 to 3 MPa (20 to 30 bar)
- For Pump **P9**: P9 Seal kit, 25 ml
- For Pump **P9H**: P9H Seal kit, 150 ml

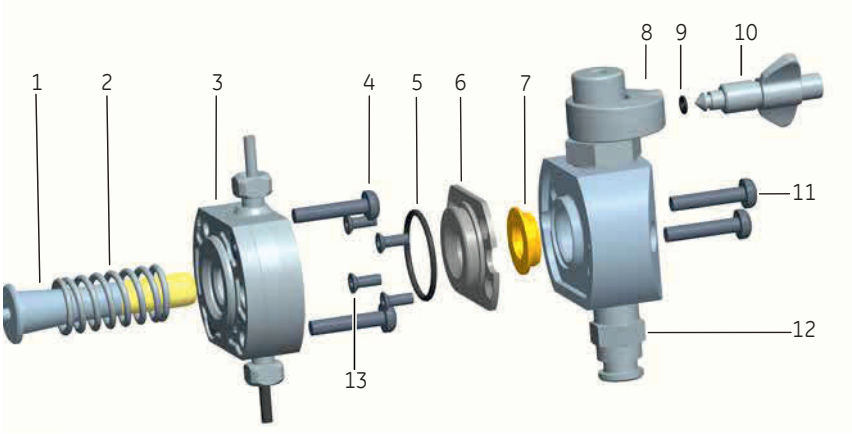
Illustrations

The illustrations below show the parts of the pump heads of the pumps **P9** and **P9H**.

Pump P9



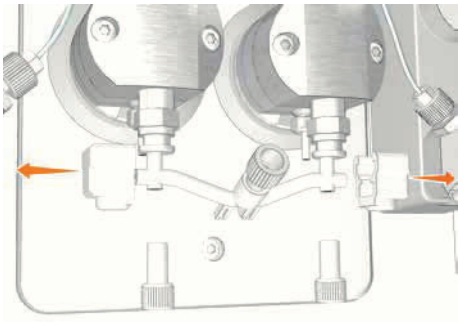
Pump P9H



Part	Description	Part	Description
1	Piston	7	Piston seal
2	Return spring	8	Outlet check valve
3	Pump membrane housing	9	O-ring
4	Star screws	10	Purge valve
5	O-ring	11	Star screws
6	Support washer	12	Inlet check valve
13	Star screws		

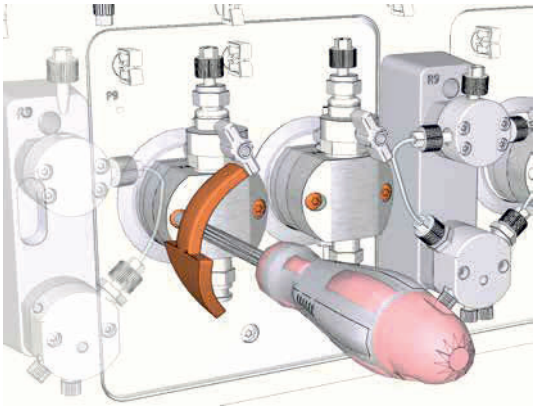
Disassemble the pump head

Step	Action
1	Make sure that no run is in progress on the instrument.
2	Disconnect the tubing from the pump head, and disconnect the pump inlet tubing.
3	Unscrew the two white plastic screws located below each pump head by hand. Pull the plastic connectors to the sides to release the inlet manifold.



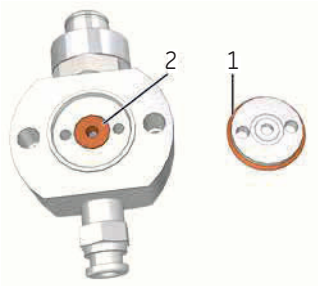
4	Disconnect the tubing of the pump piston rinsing system.

Step	Action
5	Unscrew the two screws of the front section of the pump head using a star screwdriver T20, and pull off the front section.

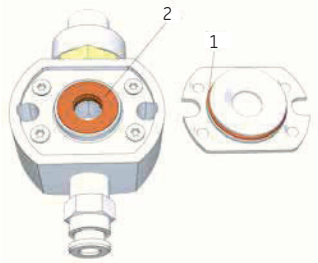


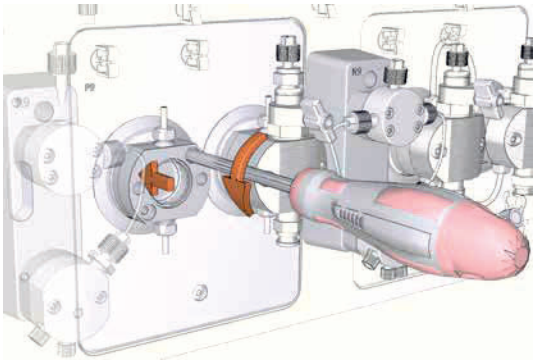
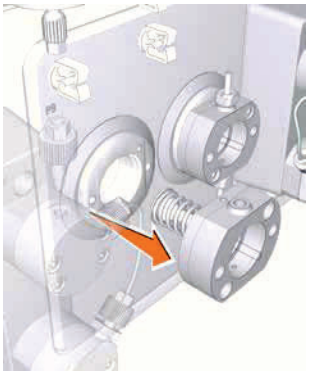
6	Place the front section of the pump head face down on the bench. For Pump **P9**, unscrew the two screws of the support washer using a star screwdriver, T20. For Pump **P9H**, unscrew the four screws of the support washer using a star screwdriver, T10. Discard the O-ring (1) on the support washer, and the discard the piston seal (2) located in the front section of the pump head. ***Note:*** *Be careful not to scratch the metal surfaces.*

Pump P9



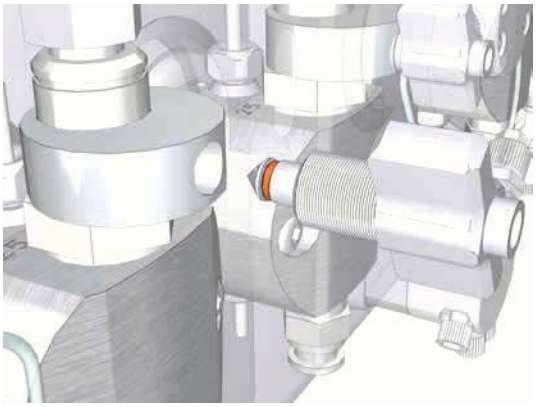
Pump P9H



Step	Action
7	Unscrew *one* of the two screws securing the pump membrane housing using a star screwdriver, T20. Unscrew the second screw, and at the same time push firmly on the front of the pump membrane housing to compensate for the pressure of the piston return spring. 
8	Carefully pull off the pump membrane housing together with the piston and return spring. 
9	Inspect the piston and return spring for signs of damage. If damaged, discard the piston and return spring and use a new piston and return spring when assembling the pump head.
10	Clean the pump head and pump membrane housing in an ultrasonic bath. If there are particles on any surfaces, the check valves should be removed and cleaned separately, see *Section 7.5 Clean pump head check valves, on page 148*.

Replace O-rings, piston seal and pump membrane housing

Step	Action
1	Unscrew the purge valve of the pump head. Replace the O-ring on the purge valve with a new O-ring, , and screw the purge valve back into the pump head.

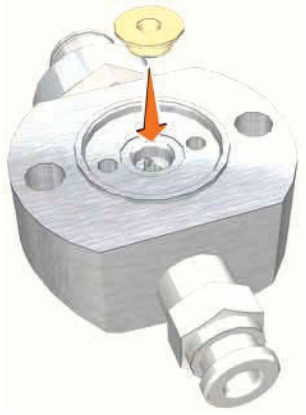


Note:

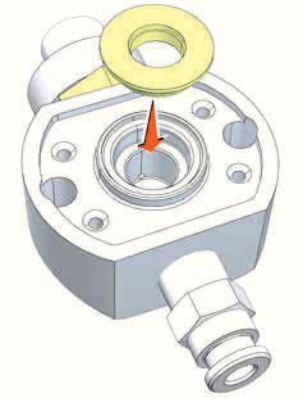
Always use Lubricant 56686700 when exchanging the O-ring 3 x 1 mm.

2	Wet a new seal with 20% ethanol. Place the new seal in the hole in the front section of the pump head and press it into position.

Pump P9

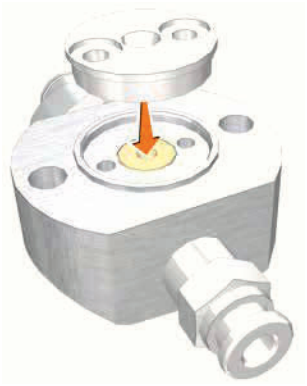


Pump P9H

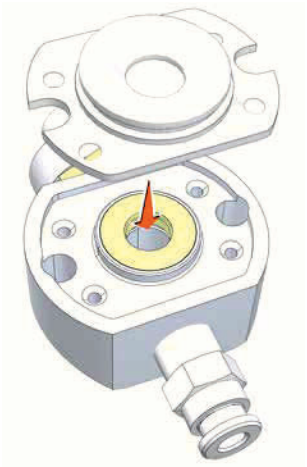


Step	Action
3	Place the support washer on top of the new seal in the front section of the pump head. Screw the two or four screws of the support washer. Make sure to tighten the screws fully.

Pump P9



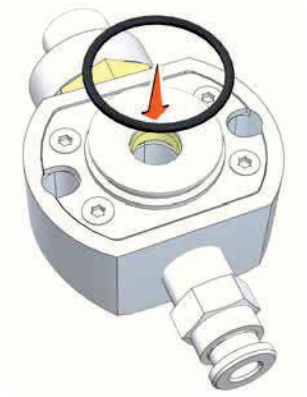
Pump P9H



4	Wet a new O-ring, 21.4 x 1.6 mm, with 20% ethanol. Fit the O-ring around the support washer.

Pump P9

Pump P9H



Assemble the pump head

Make sure to assemble the pump head correctly. Refer to *Illustrations on page 131*.

Step	Action
1	Insert the piston into the return spring. Insert piston and return spring into hole in the pump module. ***Note:*** *Do no touch the ceramic or glass part of the pump piston.*
2	Wet the membrane in the hole with 20% ethanol before mounting.
3	Place the pump membrane housing onto the locating pins on the front of the pump module.
4	Screw one of the two screws securing the pump membrane housing using a star screwdriver, T20. Push firmly on the front of the pump membrane housing to compensate for the pressure of the piston and then screw the second screw.
5	Make sure that the new seal is wetted with 20% ethanol and then tighten both screws fully.
6	Reconnect the tubing of the pump piston rinsing system.
7	Reconnect the inlet manifold.
8	Reconnect the tubing to the pump head, and reconnect the pump inlet tubing.

Break in the new pump piston seal

Follow the instruction below to break in the new pump piston seal of Pump **P9** or **P9H**.

Step	Action
1	Fill a buffer vessel with 20% ethanol in water. Immerse the inlet tubing, for example **A1** for System Pump A, **B1** for System Pump B, or **S1** for the Sample Pump in the buffer vessel. Place the buffer vessel on the buffer tray.
2	Prime the inlets and purge the pump, see *Section 4.5.1 Prime buffer inlets and purge system pumps, on page 70*.
3	• For Pump **P9**: Connect the reference capillary **Ref 2** (or an equivalent capillary that gives a backpressure of 6 to 8 MPa [60 to 80 bar]) to one of the column positions of the column valve (e.g., ports **1A** and **1B**). • For Pump **P9H**: Connect the reference capillary **Ref 1** (or an equivalent capillary that gives a backpressure of 2 to 3 MPa [20 to 30 bar] to one of the column positions of the column valve (e.g., ports **1A** and **1B**).
4	Immerse the waste tubing in the buffer vessel to recirculate the liquid.

Step	Action
5	• If breaking in a pump piston seal of a system pump, follow the instruction below: In the ***Manual instructions*** dialog box: - Select ***Flow path*** →***Column valve***, and select the position of the capillary connected to the column valve. Click ***Insert***. - Select ***Flow path*** →***Inlet A*** (for System Pump A) or ***Flow path*** →***Inlet B*** (for System Pump B) and select a ***Position***. Click ***Insert***. - Select ***Pumps and Pressures*** →***Gradient*** and set ***Target*** to 0% B (for System Pump A) or 100% B (for system pump B). - For Pump**P9**: Select ***Pumps and Pressures*** →***System flow*** and set the ***Flow rate*** to 5.0 ml/min. Click ***Insert***. For Pump**P9H**: Select ***Pumps and Pressures*** →***System flow*** and set the ***Flow rate*** to 25.0 ml/min. Click ***Insert***. - Click ***Execute***. *Result:* A system flow starts. • If breaking in a pump piston seal of a sample pump, follow the instruction below: In the ***Manual instructions*** dialog box: - Select ***Flow path*** →***Column valve***, and select the position of the capillary connected to the column valve. Click ***Insert***. - Select ***Flow path*** →***Sample inlet*** and select a ***Position***. Click ***Insert***. - Select ***Flow path*** →***Injection valve*** and click ***Direct inject*** on the ***Position*** menu. Click ***Insert***. - Select ***Pumps and Pressures*** →***Sample flow*** and set the ***Flow rate*** to 25.0 ml/min. Click ***Insert***. - Click ***Execute***. *Result:* A sample flow starts.
6	Run the flow for 2 hours.
7	Discard the used buffer.

7.3 Replace pump piston seal, O-ring, and rinse membrane of Pump P9-S

Introduction

Follow the instructions to replace the O-ring, piston seal, and rinse membrane of Pump **P9-S**.

Note: *Always replace the O-rings, piston seals, and rinse membranes of both pump heads of a pump at the same time.*

NOTICE

- Do not disassemble the pump head unless there is good reason to believe that there is an internal leakage. A sign of leakage is increasing pump rinsing solution volume. Always make sure that sufficient spare components are available before attempting to replace a spare part.
- **Replacing spare parts**. Read the instructions carefully. For example, some individual parts of the pump head can be assembled incorrectly. Check the orientation of each part before continuing with the next instruction.

Maintenance interval

Replace the O-ring, piston seal, and rinse membrane of Pump **P9-S** if they are damaged. After replacement, perform a run to break in the new piston seal.

NOTICE

Advanced maintenance. Read the instruction carefully before disassembly of the pump head.

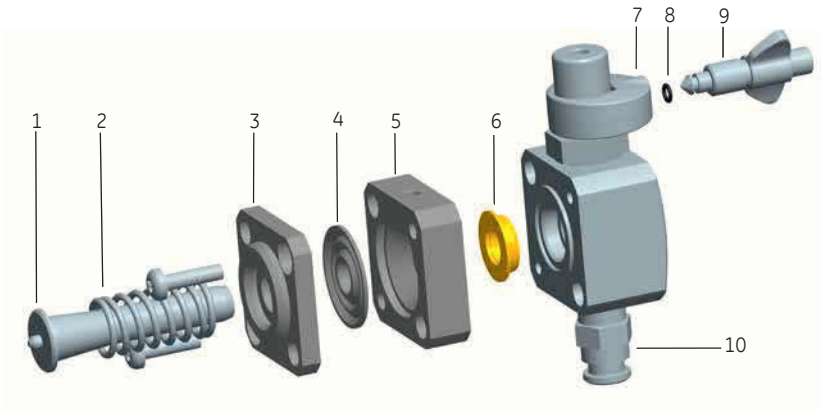
Required material

The following materials are required:

- Adjustable wrench
- Star screwdriver, T20
- Cross-headed screwdriver
- Hex wrench
- Ultrasonic bath
- Ethanol, 20%
- Reference capillary **Ref 1**
- P9-S Seal kit, 65 ml

Illustration

The illustration below shows the parts of the pump heads of Pump **P9-S**.

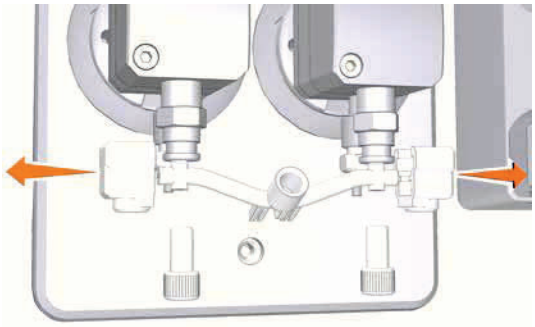


Part	Description	Part	Description
1	Piston	6	Piston seal
2	Return spring	7	Outlet check valve
3	Drain plate	8	O-ring
4	Rinse membrane	9	Purge valve
5	Rinse chamber	10	Inlet check valve

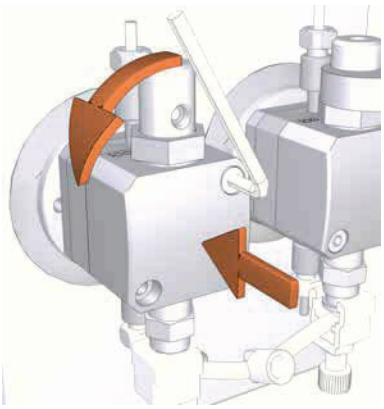
Disassemble the pump head

Step	Action
1	Make sure that no run is in progress on the instrument.
2	Disconnect the tubing from the pump head, and disconnect the pump inlet tubing.

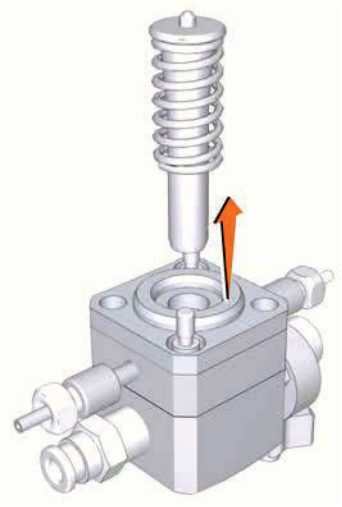
Step	Action
3	Unscrew the two white plastic screws located below each pump head by hand. Pull the plastic connectors to the sides to release the inlet manifold.



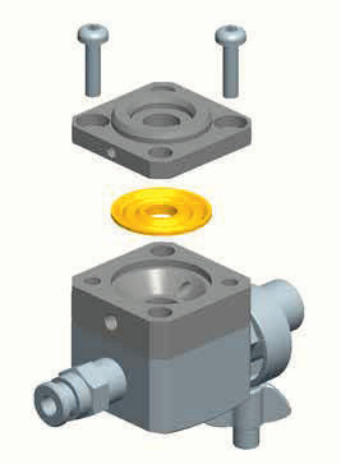
4	Disconnect the tubing of the pump piston rinsing system.
5	Unscrew *one* of the two screws of the pump head using a hex wrench. Unscrew the second screw, and at the same time push firmly on the front of the rinse chamber to compensate for the pressure of the piston return spring.



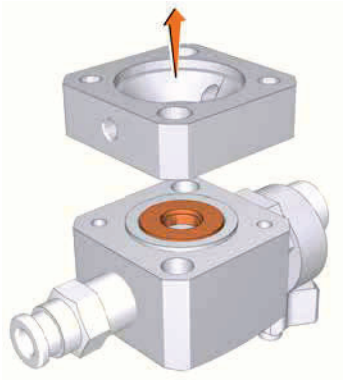
Step	Action
6	Place the pump head face down on the bench. Pull out the piston together with the return spring.



7	Inspect the piston and return spring for sign of damage. If damaged, discard the piston and return spring and use a new piston and return spring when assembling the pump head.
8	Unscrew the two screws securing the drain plate and the rinse chamber. Lift off the drain plate, and discard the membrane located between the drain plate and the rinse chamber.



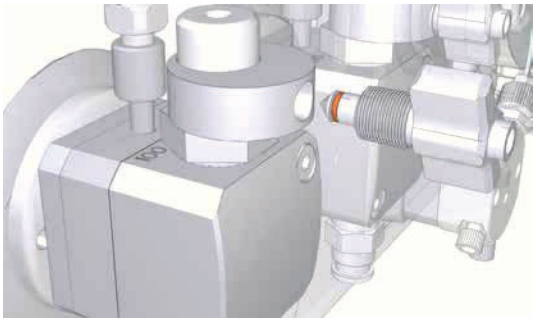
Step	Action
9	Lift off the rinse chamber. Gently pull off the piston seal. Discard the used seal.



10	Clean the pump head, rinse chamber and drain plate in an ultrasonic bath. If there are particles on any surfaces, the check valves should be removed and cleaned separately, see *Section 7.5 Clean pump head check valves, on page 148*.

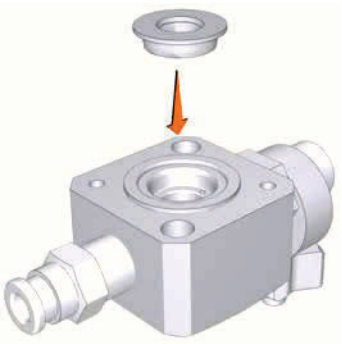
Replace O-ring, piston seal, and rinse membrane

Step	Action
1	Unscrew the purge valve of the pump head. Replace the O-ring on the purge valve with a new O-ring, 3x1 mm, and screw the purge valve back into the pump head.



Note:

Always use Lubricant 56686700 when exchanging the O-ring 3 x 1 mm.

Step	Action
2	Wet a new seal with 20% ethanol. Place the new seal in the hole in the front section of the pump head and press it into position. 
3	With the pump head facing downwards on the bench, place the rinse chamber onto the front section of the pump head with the rinse ports in line with the check valves. The conical depression in the rinse chamber shall be facing upwards. Wet a new membrane with 20% ethanol, and place the membrane into the rinse chamber with the conical face upwards. 

Assemble the pump head

Make sure to assemble the pump head correctly. Refer to *Illustrations on page 131*.

Step	Action
1	Place the drain plate on top of the assembly. Screw the two screws through the drain plate and the rinse chamber using a cross-headed screwdriver.
2	Wipe clean the piston and remove all finger prints. Wet the piston with 20% ethanol, and insert the piston into the return spring. With the pump head facing downwards on the bench, insert the piston into the pump head by pushing it gently but firmly vertically downwards into the seal.

Step	Action
3	Place the complete pump head over the locating pins on the front panel of the sample pump module. Turn the pump head so that the text **UP** on the drain plate is facing upwards. Push firmly on the front of the pump head, and at the same time screw one of the screws to fasten the pump head onto the front of the module using a hex wrench. Screw the second screw of the pump head. Make sure to tighten both screws fully.
4	Reconnect the tubing of the pump piston rinsing system.
5	Reconnect the inlet manifold.
6	Reconnect the tubing to the pump head, and reconnect the pump inlet tubing.
7	Break in the new pump piston seal, see instruction below.

Break in the new pump piston seal

Follow the instruction to break in the new pump piston seal of Pump **P9-S**.

Step	Action
1	Fill a buffer vessel with 20% ethanol in water. Immerse a piece of sample inlet tubing, for example **S1**, in the buffer vessel. Place the buffer vessel on the buffer tray.
2	Prime the inlets and purge the pump, see *Section 4.5.2 Prime sample inlets and purge Sample Pump, on page 77*.
3	Connect the reference capillary **Ref 1** (or an equivalent capillary that gives a backpressure of 2 to 3 MPa [20 to 30 bar]) to one of the column positions of the column valve (e.g., ports **1A** and **1B**).
4	Immerse the waste tubing in the buffer vessel to recirculate the liquid.
5	In the ***Manual instructions*** dialog: • Select ***Flow path*** →***Column position***, and select the ***Position*** of the capillary connected to the column valve. Click ***Insert***. • Select ***Flow path*** →***Sample inlet*** and select a ***Position***. Click ***Insert***. • Select ***Flow path*** →***Injection valve*** and click ***Direct inject*** on the ***Position*** menu. Click ***Insert***. • Select ***Pumps*** →***Sample flow*** and set the ***Flow rate*** to 25.0 ml/min. Click ***Insert***. • Click ***Execute***. *Result:* A sample flow of 25.0 ml/min starts.

Step	Action
6	Run the flow for 2 hours.
7	Discard the used buffer.

7.4 Replace pump pistons

Introduction

Follow the instructions to replace the pump pistons of the pumps **P9**, **P9H** and **P9-S**.

Note: *Always replace the O-rings, piston seals, and rinse membranes of both pump heads of a pump at the same time.*

NOTICE

- Do not disassemble the pump head unless there is good reason to believe that there is an internal leakage. A sign of leakage is increasing pump rinsing solution volume. Always make sure that sufficient spare components are available before attempting to replace a spare part.
- **Replacing spare parts**. Read the instructions carefully. For example, some individual parts of the pump head can be assembled incorrectly. Check the orientation of each part before continuing with the next instruction.

Maintenance interval

Replace the pump pistons if they are damaged.

Required material

The following materials are required:

- Adjustable wrench
- Star screwdriver, T20
- Piston kit

Replace pump pistons of Pump P9 and P9H

If a damaged piston has been in operation, the piston seal will be destroyed and should also be replaced. To replace the piston and the seal of a system pump, see *Section 7.2 Replace pump piston seal, O-rings, and rinse membrane of Pump P9 or Pump P9H, on page 130*.

Replace pump pistons of Pump P9-S

If a damaged piston has been in operation, the piston seal will be destroyed and should also be replaced. To replace the piston and the seal of Pump **P9-S**, see *Section 7.3 Replace pump piston seal, O-ring, and rinse membrane of Pump P9-S, on page 139*.

7.5 Clean pump head check valves

Introduction

Clean the check valves when required, for example if particles like dust or salt crystals in the check valve cause irregular or low flow. The cleaning procedure is the same for the system pumps and the sample pump.

Required material

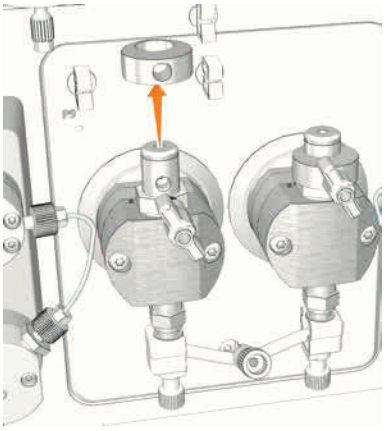
The following materials are required:

- Adjustable wrench
- 100% Methanol
- Distilled water
- Ultrasonic bath

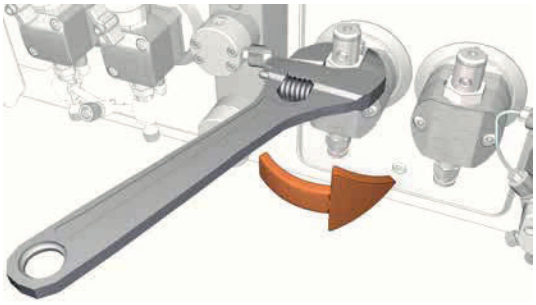
Instruction

Follow the instructions to remove and clean the pump head check valves.

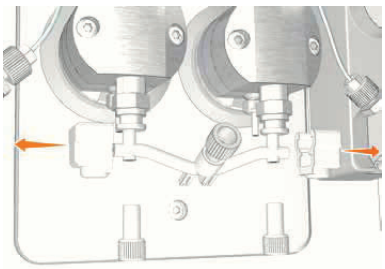
Step	Action
1	Before taking the check valve apart, always try to clean the check valves by priming the pump heads first with distilled water, then with 100% Methanol and then with distilled water again.
2	Switch off the instrument.
3	Disconnect the tubing from the pump head and disconnect the pump inlet tubing. Disconnect the tubing of the pump rinsing system.
4	Unscrew the purge valve by turning it counter-clockwise, and lift off the metal ring.



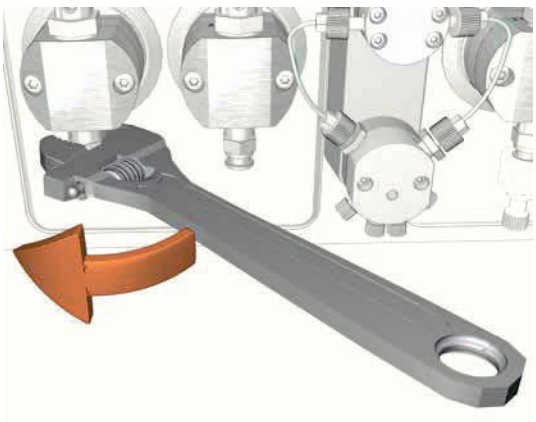
Step	Action
5	Unscrew the plastic nut of the upper check valve using an adjustable wrench, and gently lift off the upper check valve.



6	Unscrew the two white plastic screws located below each pump head. Pull the plastic connectors to the sides to release the inlet manifold.



7	Unscrew the lower check valve using an adjustable wrench.



Step	Action
8	 **WARNING** **Hazardous substances.** When using hazardous chemicals, take all suitable protective measures, such as wearing protective clothing, glasses and gloves resistant to the substances used. Follow local and/or national regulations for safe operation and maintenance of the product. Immerse the valves completely in methanol and place them in an ultrasonic bath for a few minutes. Repeat the ultrasonic bath with deionized water.
9	Refit the check valves.
10	Tighten the nut until fully finger-tight and then use the adjustable wrench to tighten a further 90 degrees.
11	Refit the inlet manifold and reconnect the tubing to the pump head.

7.6 Cleaning before planned service

Cleaning before planned maintenance/service

To ensure the protection and safety of service personnel, all equipment and work areas must be clean and free of any hazardous contaminants before a Service Engineer starts maintenance work.

Please complete the checklist in the *On Site Service Health and Safety Declaration Form* or the *Health and Safety Declaration Form for Product Return or Servicing*, depending on whether the instrument is going to be serviced on site or returned for service, respectively.

Health and safety declaration forms

Health and safety declaration forms are available for copying or printing in the *Reference information* chapter of this manual, or on digital media supplied with the user documentation.

8 Reference Information

About this chapter

This chapter lists the technical specifications of the ÄKTA avant instrument. The chapter also includes a chemical resistance guide. See also ÄKTA avant 25 Product Documentation and ÄKTA avant 150 Product Documentation for detailed technical specifications.

In this chapter

8.1 System specifications

Technical specifications

Parameter	Data
System configuration	Benchtop system, external computer
Control system	UNICORN™ 6.3.2 or later version
Connection between PC and instrument	Ethernet
Dimensions (Length x Depth x Height)	860 x 710 x 660 mm
Weight (excluding computer)	116 kg
Power supply	100-240 VAC, 50-60 Hz
Power consumption	800 VA
Enclosure protective class	IP 21, wet side IP 22
Tubing and connectors	ÄKTA avant 25: • Inlet: FEP tubing, inner diameter (i.d.) 1.6 mm, Tubing connector 5/16″ + Ferrule (yellow), 1/8″ • Pump to Injection valve: PEEK tubing, i.d. 0.75 mm, Fingertight connector, 1/16″ • After Injection valve: PEEK tubing, i.d. 0.50 mm, Fingertight connector, 1/16″ • Outlet and waste: ETFE tubing, i.d. 1.0 mm, Fingertight connector, 1/16″ ÄKTA avant 150: • Inlet: FEP tubing, i.d. 2.9 mm, Tubing connector 5/16″ + Ferrule (blue), 3/16″ • After pumps: PEEK tubing, 1.0 mm i.d., Finger-tight connector, 1/16″ • Outlet: FEP o.d.1/8″, i.d. 1.6 mm, Tubing connector 5/16″ + Ferrule (yellow), 1/8″ • Waste: ETFE tubing, i.d. 1.0 mm, Fingertight connector, 1/16″

Enviromental requirements

Parameter	Data
Storage and transport temperature range	-25°C to 60°C
Chemical environment	See *Section 8.2 Chemical resistance guide, on page 155*.
Operating temperature range	4°C to 35°C
Relative humidity	20% to 95%, noncondensing

Equipment noise level

Equipment	Acoustic noise level
ÄKTA avant instrument	< 70 dBA

8.2 Chemical resistance guide

Introduction

This section provides general information about biocompatibilty and detailed information about chemical resistance of the ÄKTA avant instrument.

In this section

8.2.1 General information about biocompatibility and chemical resistance

Biocompatibility

The ÄKTA avant instrument is designed for maximum biocompatibility, with biochemically inert flow paths constructed mainly from titanium, PEEK and highly resistant fluoropolymers and fluoroelastomers. Titanium is used as far as possible to minimize contribution of potentially deactivating metal ions such as iron, nickel and chromium. There is no standard stainless steel in the flow path. Plastics and rubber materials are selected to avoid leakage of monomers, plasticizers or other additives.

Cleaning chemicals

Strong cleaning works well with 2 M sodium hydroxide, 70% acetic acid or the alcohols methanol, ethanol and isopropyl alcohol. Complete system cleaning using 1 M hydrochloric acid should be avoided in order to not damage the pressure sensors. If you are cleaning separation media using 1 M hydrochloric acid, use loop injections of the acid and make sure that the column is not mounted on the Column Valve **V9-C**. The Column Valve **V9-C** contains a pressure sensor which can be damaged by 1 M hydrochloric acid.

Long time use of 0.2 M HCl connected to the Quaternary Valve **Q9** as part of a ***BufferPro*** recipe is acceptable. The solution becomes diluted further down in the system.

If sodium hypochlorite is used as sanitizing agent instead of 2 M sodium hydroxide, use a concentration up to 10%.

Organic solvents

Reversed phase chromatography of proteins works well with 100% acetonitrile and additives trifluoroacetic acid (TFA) up to 0.2% or formic acid up to 5%.

Strong organic solvents like ethyl acetate, 100% acetone or chlorinated organic solvents should be avoided. These might cause swelling of plastic material and reduce the pressure tolerance of PEEK tubing. For this reason, flash chromatography and straight ("normal") phase chromatography is generally not recommended on the system.

Assumptions made

The ratings are based on the following assumptions:

- Synergy effects of chemical mixtures have not been taken into account.
- Room temperature and limited overpressure is assumed.

Note: *Chemical influences are time and pressure dependent. Unless otherwise stated, all concentrations are 100%.*

8.2.2 Chemical resistance specifications

Introduction

This section provides detailed information about chemical resistance of the ÄKTA avant instrument to some of the most commonly used chemicals in liquid chromatography. Regarding exposure to solutions not covered by this information, contact your Cytiva representative for recommendations.

Note: *A user can be exposed to large volumes of chemical substances over a long time period. Material Safety Data Sheets (MSDS) provide the user with information regarding characteristics, human and environmental risks and preventive measures. Make sure that you have the MSDS available from your chemical distributor and/or databases on the internet.*

Aqueous buffers

The specified aqueous buffers are suitable for continuous use.

Chemical	Concentration	CAS no/EC no
Aqueous buffers pH 2-12	N/A	N/A

Strong chemicals and salts for CIP

The following chemicals are suitable for up to 2 h contact time at room temperature.

Chemical	Concentration	CAS no/EC no
Acetic acid	70%	75-05-8/ 200-835-2
Decon™ 90	10%	N/A
Ethanol	100%	75-08-1/ 200-837-3
Methanol	100%	67-56-1/ 200-659-6
Hydrochloric acid[1]	0.1 M	7647-01-0/ 231-595-7
Isopropanol	100%	67-63-0/ 200-661-7
Sodium hydroxide	2 M	1310-73-2/ 215-185-5
Sodium hydroxide/ethanol	1 M/40%	N/A
Sodium chloride	4 M	7647-14-5/ 231-598-3

Chemical	Concen-tration	CAS no/EC no
Sodium hypochlorite	10%	7681-52-9/231-668-3

[1] If hydrochloric acid, HCl, is used as a cleaning agent when columns are connected to the system, the HCl concentration should not exceed 0.1 M in the pressure sensors. Remember that the ÄKTA avant system has pressure sensors in the column valve **V9-C**.

For other parts of the system up to 1 M HCl is acceptable for short periods of use. See *Cleaning chemicals on page 156*

Solubilization and denaturing agents

The following chemicals are suitable for continuous use, as additives in separation and purification methods.

Chemical	Concen-tration	CAS no/EC no
Guanidinium hydrochloride	6 M	50-01-1/ 200-002-3
Sodium dodecyl sulfate (SDS)	1%	151-21-3/ 205-788-1
Tween™ 20	1%	9005-64-5/ 500-018-3
Urea	8 M	57-13-6/ 200-315-5

Chemicals used in reversed phase chromatography (RPC)

The following chemicals are suitable for continuous use.

Chemical	Concen-tration	CAS no/EC no
Acetonitrile[1]	100%	75-05-8/ 200-835-2
Acetonitrile/Tetrahydro-furan[1]	85%/15%	109-99-9/ 203-726-8
Acetonitrile/water/ Trifluoroacetic acid (TFA)[2]	Max 0.2% TFA	N/A
Ethanol	100%	75-08-1/ 200-837-3
Isopropanol	100%	67-63-0/ 200-661-7
Methanol	100%	74-93-1/ 200-659-6

Chemical	Concentration	CAS no/EC no
Water/organic mobile phase/formic acid	Max 5% formic acid	N/A

[1] Organic solvents can penetrate weaknesses in PEEK tubing walls more easily than water based buffers. Special care should therefore be taken with prolonged use of organic solvents close to pressure limits.

Note: *Quaternary valve is not resistant.*

Depending on pressure, tubing between pump head and pressure monitor needs to be changed. See *ÄKTA avant User Manual* for more information.

[2] Mobile phase system.

Note: *It is recommended to replace the mixer sealing ring with the highly resistant O-ring (product code 29011326) if the system is to be exposed to organic solvents or high concentrations of organic acids, such as acetic acid and formic acid, for a longer period of time.*

Salts and additives for hydrophobic interaction chromatography (HIC)

The following chemicals are suitable for continuous use.

Chemical	Concentration	CAS no/EC no
Ammonium chloride	2 M	12125-02-9/ 235-186-4
Ammonium sulfate	3 M	7783-20-2/ 231-984-1
Ethylene glycol	50%	107-21-1/ 203-473-3
Glycerol	50%	56-81-5/ 200-289-5

Reducing agents and other additives

The following chemicals are suitable for continuous use.

Chemical	Concentration	CAS no/EC no
Arginine	2 M	74-79-3 / 200-811-1
Benzyl alcohol	2%	100-51-6 / 202-859-9
Dithioerythritol (DTE)	100 mM	3483-12-3 / 222-468-7
Dithiothreitol (DTT)	100 mM	6862-68-8 / 229-998-8
Ethylenediaminetetraacetic acid (EDTA)	100 mM	60-00-4 / 200-449-4

Chemical	Concen-tration	CAS no/EC no
Mercaptoethanol	20 mM	37482-11-4 / 253-523-3
Potassium chloride	4 M	7447-40-7 / 231-211-8

Other substances

The following chemicals are suitable for continuous use.

Chemical	Concen-tration	CAS no/EC no
Acetone	10%	67-64-1/ 200-662-2
Ammonia	30%	7664-41-7/ 231-635-3
Dimethyl sulphoxide (DMSO)	5%	67-68-5/ 200-664-3
Ethanol for long-term storage	20%	75-08-1/ 200-837-3
Phosphoric acid	0.1 M	7664-38-2/ 231-633-2

8.3 Recycling information

Introduction

This section contains information about the decommisioning of ÄKTA avant.

Decontamination

The product must be decontaminated before decommissioning. All local regulations must be followed with regard to scrapping of the equipment.

Disposal of the product

When taking the product out of service, the different materials must be separated and recycled according to national and local environmental regulations.

CAUTION

Always use appropriate personal protective equipment when decommissioning the equipment.

Disposal of electrical components

Waste comprising electrical and electronic equipment must not be disposed of as unsorted municipal waste and must be collected separately. Please contact an authorized representative of the manufacturer for information concerning the decommissioning of equipment.

8.4 Regulatory information

Introduction

This section lists the regulations and standards that apply to ÄKTA avant.

In this section

8.4.1 Contact information

Contact information for support

To find local contact information for support and sending troubleshooting reports, visit *cytiva.com/contact*.

Manufacturing information

The table below summarizes the required manufacturing information.

Requirement	Information
Name and address of manufacturer	Cytiva Sweden AB Björkgatan 30 SE 751 84 Uppsala Sweden
Telephone number of manufacturer	+ 46 771 400 600

8.4.2 European Union and European Economic Area

Introduction

This section describes regulatory information for the European Union and European Economic Area that applies to the equipment.

Conformity with EU Directives

See the EU Declaration of Conformity for the directives and regulations that apply for the CE marking.

If not included with the product, a copy of the EU Declaration of Conformity is available on request.

CE marking

The CE marking and the corresponding EU Declaration of Conformity is valid for the instrument when it is:

- used according to the *Operating Instructions* or user manuals, and
- used in the same state as it was delivered, except for alterations described in the *Operating Instructions* or user manuals.

8.4.3 Eurasian Economic Union Евразийский экономический союз

This section describes the information that applies to the product in the Eurasian Economic Union (the Russian Federation, the Republic of Armenia, the Republic of Belarus, the Republic of Kazakhstan, and the Kyrgyz Republic).

Introduction

This section provides information in accordance with the requirements of the Technical Regulations of the Customs Union and (or) the Eurasian Economic Union.

Введение

В данном разделе приведена информация согласно требованиям Технических регламентов Таможенного союза и (или) Евразийского экономического союза.

Manufacturer and importer information

The following table provides summary information about the manufacturer and importer, in accordance with the requirements of the Technical Regulations of the Customs Union and (or) the Eurasian Economic Union.

Requirement	Information
Name, address and telephone number of manufacturer	See *Manufacturing information*
Importer and/or company for obtaining information about importer	LLC Global Life Sciences Solutions Rus Russian Federation, 123112 Presnenskaya nab., 10, fl. 12, pr. III, room 6 Telephone: + 7 495 739 6931 Fax nr: + 7 495 739 6932 E-mail: *rucis@cytiva.com*

Информация о производителе и импортере

В следующей таблице приводится сводная информация о производителе и импортере, согласно требованиям Технических регламентов Таможенного союза и (или) Евразийского экономического союза.

Требование	Информация
Наименование, адрес и номер телефона производителя	См. *Информацию об изготовлении*

Требование	Информация
Импортер и/или лицо для получения информации об импортере	ООО "Глобал Лайф Сайэнсиз Солюшнз Рус" Российская Федерация, 123112 Пресненская наб., д. 10, эт. 12, пом. III, ком. 6 Телефон: + 7 495 739 6931 Факс: + 7 495 739 6932 Адрес электронной почты: *rucis@cytiva.com*

Description of symbol on the system label
Описание обозначения на этикетке системы

This Eurasian compliance mark indicates that the product is approved for use on the markets of the Member States of the Customs Union of the Eurasian Economic Union

Данный знак о Евразийском соответствии указывает, что изделие одобрено для использования на рынках государств-членов Таможенного союза Евразийского экономического союза

8.4.4 Regulations for North America

Introduction

This section describes the information that applies to the product in the USA and Canada.

FCC compliance

This device complies with part 15 of the FCC Rules. Operation is subject to the following two conditions: (1) This device may not cause harmful interference, and (2) this device must accept any interference received, including interference that may cause undesired operation.

Note: *The user is cautioned that any changes or modifications not expressly approved by Cytiva could void the user's authority to operate the equipment.*

This equipment has been tested and found to comply with the limits for a Class A digital device, pursuant to part 15 of the FCC Rules. These limits are designed to provide reasonable protection against harmful interference when the equipment is operated in a commercial environment. This equipment generates, uses, and can radiate radio frequency energy and, if not installed and used in accordance with the instruction manual, may cause harmful interference to radio communications. Operation of this equipment in a residential area is likely to cause harmful interference in which case the user will be required to correct the interference at his own expense.

8.4.5 Regulatory statements

Introduction

This section shows regulatory statements that apply to regional requirements.

EMC emission, CISPR 11: Group 1, Class A statement

NOTICE

This equipment is not intended for use in residential environments and may not provide adequate protection to radio reception in such environments.

South Korea

Regulatory information to comply with the Korean technical regulations.

NOTICE

Class A equipment (equipment for business use).

This equipment has been evaluated for its suitability for use in a business environment.

When used in a residential environment, there is a concern of radio interference.

주의

A급 기기 (업무용 방송통신 기자재)

이 기기는 업무용환경에서 사용할 목적으로 적합성평가를 받은 기기

로서 가정용 환경에서 사용하는 경우 전파간섭의 우려가 있습니다.

8.4.6 Declaration of Hazardous Substances (DoHS)

根据 SJ/T11364-2014《电子电气产品有害物质限制使用标识要求》特提供如下有关污染控制方面的信息。

The following product pollution control information is provided according to SJ/T11364-2014 Marking for Restriction of Hazardous Substances caused by electrical and electronic products.

电子信息产品污染控制标志说明
Explanation of Pollution Control Label

该标志表明本产品含有超过中国标准 GB/T 26572《电子电气产品中限用物质的限量要求》中限量的有害物质。标志中的数字为本产品的环保使用期，表明本产品在正常使用的条件下，有毒有害物质不会发生外泄或突变，用户使用本产品不会对环境造成严重污染或对其人身、财产造成严重损害的期限。单位为年。

为保证所申明的环保使用期限，应按产品手册中所规定的环境条件和方法进行正常使用，并严格遵守产品维修手册中规定的定期维修和保养要求。

产品中的消耗件和某些零部件可能有其单独的环保使用期限标志，并且其环保使用期限有可能比整个产品本身的环保使用期限短。应到期按产品维修程序更换那些消耗件和零部件，以保证所申明的整个产品的环保使用期限。

本产品在使用寿命结束时不可作为普通生活垃圾处理，应被单独收集妥善处理。

This symbol indicates the product contains hazardous materials in excess of the limits established by the Chinese standard GB/T 26572 Requirements of concentration limits for certain restricted substances in electrical and electronic products. The number in the symbol is the Environment-friendly Use Period (EFUP), which indicates the period during which the hazardous substances contained in electrical and electronic products will not leak or mutate under normal operating conditions so that the use of such electrical and electronic products will not result in any severe environmental pollution, any bodily injury or damage to any assets. The unit of the period is “Year”.

In order to maintain the declared EFUP, the product shall be operated normally according to the instructions and environmental conditions as defined in the product manual, and periodic maintenance schedules specified in Product Maintenance Procedures shall be followed strictly.

Consumables or certain parts may have their own label with an EFUP value less than the product. Periodic replacement of those consumables or parts to maintain the declared EFUP shall be done in accordance with the Product Maintenance Procedures.

This product must not be disposed of as unsorted municipal waste, and must be collected separately and handled properly after decommissioning.

有害物质的名称及含量
Name and Concentration of Hazardous Substances

产品中有害物质的名称及含量
Table of Hazardous Substances' Name and Concentration

部件名称 Compo-nent name	有害物质 Hazardous substance					
	铅 (Pb)	汞 (Hg)	镉 (Cd)	六价铬 (Cr(VI))	多溴联苯 (PBB)	多溴二苯醚 (PBDE)
28930842	X	0	X	0	0	0
28976337	X	0	X	0	0	0
29011360	X	0	0	0	0	0
29011361	X	0	0	0	0	0
29011362	X	0	0	0	0	0
29090689	X	0	0	0	0	0
29090691	X	0	0	0	0	0
29011353	X	0	0	0	0	0
29011358	X	0	0	0	0	0

0: 表示该有害物质在该部件所有均质材料中的含量均在 GB/T 26572 规定的限量要求以下。

X: 表示该有害物质至少在该部件的某一均质材料中的含量超出 GB/T 26572 规定的限量要求。

- 此表所列数据为发布时所能获得的最佳信息.

0: Indicates that this hazardous substance contained in all of the homogeneous materials for this part is below the limit requirement in GB/T 26572.

X: Indicates that this hazardous substance contained in at least one of the homogeneous materials used for this part is above the limit requirement in GB/T 26572

- Data listed in the table represents best information available at the time of publication.

8.5 Ordering information

Introduction

This chapter lists accessories and user replaceable spare parts available for ÄKTA avant.

Tubing

Item	Code no.
Reference capillary 1	28950749
Reference capillary 2	28950750
ETFE Tubing kit 10×1.0 m, id 1.0 mm, od 1/16″ ***Note:*** *Outlet tubing for ÄKTA avant 25.*	28980995
Tubing kit 10×1.5m, FEP i.d.1.6 mm, o.d. 1/8″ ***Note:*** *Inlet tubing for ÄKTA avant 25/Outlet tubing for ÄKTA avant 150*	28980984
Tubing kit 10×1.5m, FEP id 2.9 mm, o.d 3/17″ ***Note:*** *Inlet tubing for ÄKTA avant 150.*	28980987
Sample tubing kit for 7 inlets i.d. 0.75 mm ***Note:*** *Narrow inlet tubing for ÄKTA avant 25.*	28957217
Rinse system tubing	28956504
Replacement tubing kit, ÄKTA avant 25	28956606
Replacement Tubing Kit, ÄKTA avant 150	28979446
BufferPro InA and InB tubing kit, FEP i.d. 1.6 mm o.d. 1/8″	28980998
Complete tubing marking kit ***Note:*** *Tags for all tubing in ÄKTA avant 25 and ÄKTA avant 150.*	28956608
Inlet/outlet tubing tag kit, ÄKTA avant 25 ***Note:*** *Tags for all inlet and outlet tubing in ÄKTA avant 25.*	28981001

Item	Code no.
Inlet/outlet tubing tag kit, ÄKTA avant 150 ***Note:*** *Tags for all inlet and outlet tubing in ÄKTA avant 150.*	28981004
Union 1/16″ male/male, i.d. 0.5 mm (5-pack)	28954326
Tubing cutter	18111246
Inlet filter holder kit	11000407
Inlet filter set	11000414

Fittings and connectors

Item	Product code
Fingertight connector, 1/16″ male	18111255
Tubing connector for o.d. 1/16″ tubing	18112707
Tubing connector for o.d. 1/8″ tubing	18112117
Tubing connector for o.d. 3/16″ tubing	18111249
Union, 1/16" female to 1/16" female, (5 pcs)	11000339
Union, Fingertight 1/16" female to 1/16" female, i.d. 0.3 mm (4 pcs)	11000852
Union, 1/16" male to M6 female	18111258
Union, 1/16" female to M6 male	18111257
Union, Luer female to 1/16" male	18111251
Union, 1/16" male to 1/16" male, i.d. 0.5 mm (2 pcs)	18112093
Union, 5/16" female to M6 male	18112776
Union, 5/16" female to 1/16" male	18114208
Union, M6 female to 1/16" male	18385801
Ferrule for 1/16" tubing connector, blue	18112706
Ferrule for 1/8" tubing connector, yellow	18112118
Ferrule for 3/16" tubing connector, blue	18111248
Stop plug, 5/16" male	18111250
Stop plug, 1/16" male	18111252

Holders

Item	Code no.
Adapter for air sensor	28956342
Bottle holder	28956327
Column clamp (for columns o.d. 10 to 21 mm)	28956319
Column holder	28956282
Column holder rod	28956270
Flexible column holder	28956295
Loop holder	29011350
Multi-purpose holder	29011349
Rail extension	29011352
Tube holder (5-pack)	28954329
Tubing holder comb	28956286
Tubing holder, spool for small tubing (o.d. 1/8" and smaller)	28956274
Tubing holder, spool for large inlet tubing (o.d. 3/16") for ÄKTA avant 150	29014283
Inlet filter holder kit	11000407
Screw lid kit, GL45	11000410

Conversion kits

Item	Code no.
ÄKTA avant Conversion kit, 25 to 150 ***Note:*** *Contact Service for installation.*	28980168
ÄKTA avant Conversion kit, 150 to 25 ***Note:*** *Contact Service for installation.*	28981861

Pump spare parts

Item	Code no.
P9-S Seal kit, 65 ml	28960250
P9 Seal kit, 25 ml (seal kit, P9)	28952642
Piston kit, 100 ml (piston kit, P9-S)	18111213
P9 Piston, kit 25 ml	28952640
Check valve kit (check valves in/out for P9, P9H and P9-S)	18112866
P9H Piston kit, 150 ml	28979368
P9H Seal kit, 150 ml	28979373

Mixer

Item	Code no.
Mixer chamber 0.6 ml	28956186
Mixer chamber 1.4 ml (mounted at delivery in ÄKTA avant 25)	28956225
Mixer chamber 5 ml (mounted at delivery in ÄKTA avant 150)	28956246
Mixer chamber 15 ml	28980309
O-ring 13.1 × 1.6 mm ***Note:*** *For Mixer chamber 0.6, 1.4, and 5 ml.*	28953545
O-ring 13.1 × 1.6 mm High resistance (can be used as an alternative to 28953545)	29011326
O-ring 22.1 × 1.6 mm ***Note:*** *For Mixer chamber 15 ml.*	28981857
Online filter kit	18102711

Valves

ÄKTA avant 25

Item	Code no.
Column Valve **V9-C**	28956506
pH Valve **V9-pH**	28956508
Inlet Valve **V9-IA**	28956510
Inlet Valve **V9-IB**	28962006
Inlet Valve **V9-IS**	28962007
Outlet Valve **V9-O**	28956512
Injection Valve **V9-Inj**	28956514
Inlet Valve **V9-A2**	28957221
Inlet Valve **V9-B2**	28957223
Inlet Valve **V9-S2**	28957225
Inlet Valve **V9-X1**	28957227
Inlet Valve **V9-X2**	28957234
Column Valve **V9-C2**	28957236
Outlet Valve **V9-O2**	28957238
Outlet Valve **V9-O3**	28957240
Versatile Valve **V9-V**	29011353
Loop Valve **V9-L**	29011358

ÄKTA avant 150

Item	Code no.
Column Valve **V9H-C**	28979241
pH Valve **V9H-pH**	28979246
Inlet Valve **V9H-IA**	28979248
Inlet Valve **V9H-IB**	28979277
Inlet Valve **V9H-IS**	28979279
Outlet Valve **V9H-O**	28979281

Item	Code no.
Injection Valve **V9H-Inj**	28979283
Inlet Valve **V9H-A2**	28979303
Inlet Valve **V9H-B2**	28979315
Inlet Valve **V9H-S2**	28979320
Inlet Valve **V9H-X1**	28979326
Inlet Valve **V9H-X2**	28979328
Column Valve **V9H-C2**	28979330
Outlet Valve **V9H-O2**	28979332
Outlet Valve **V9H-O3**	28979337
Versatile Valve **V9H-V**	29090691
Loop Valve **V9H-L**	29090689

Injection valve accessories

Item	Code no.
Sample loop, 10 µl	18112039
Sample loop, 100 µl	18111398
Sample loop, 500 µl (mounted at delivery)	18111399
Sample loop, 1 ml	18111401
Sample loop, 2 ml	18111402
Sample loop, FEP 10 ml	18116124
Superloop, M6 fitting, 10 ml	19758501
Superloop, 1/16" fittings (ÄKTA design), 50 ml	18111382
Superloop, M6 fitting, 150 ml	18102385
Fill port, INV-907	18112766
Injection kit, INV-907	18111089
Connector 1/16" Male/Luer Female	28985812

Built-in fraction collector

Item	Code no.
Cassette tray	28954209
Cassette, for 50 ml tubes (2-pack) ***Note:*** *For 6 tubes*	28956402
Cassette, for 15 ml tubes (2-pack)	28956404
Cassette for 5 ml tubes (2-pack)	29133422
Cassette, for 8 ml tubes (2-pack)	28956425
Cassette for 3 ml tubes (2-pack)	28956427
Cassette, for deep-well plate (2-pack)	28954212
Rack, for 50 ml tubes ***Note:*** *For 55 tubes*	28980319
Rack, for 250 ml bottles	28981873

Fraction Collector F9-R

Item	Code no.
Fraction Collector F9-R	29011362
Tube Rack Complete, 175 × 12 mm	19868403
Tube Rack Complete, 95 × 10–18 mm	18305003
Tube Rack Complete, 40 x 30 mm	18112467
Bowl	18305103
Tube Support	18305402
Tubing Holder	18646401
Tube Holder and Guide, 175 × 12 mm	19724202
Tube Holder and Guide, 95 × 10–18 mm	19868902
Tube Holder and Guide, 40 × 30 mm	18112468
Drive sleeve	19606702

pH monitor

Item	Code no.
pH electrode	28954215
O-ring 5.3 × 2.4 mm	28956497

UV monitor

Item	Code no.
UV flow cell U9-0.5, 0.5 mm for **U9-M**	28979386
UV flow cell U9-2, 2 mm for **U9-M**	28979380
UV flow cell U9-10, 10 mm for **U9-M**	28956378
UV Monitor U9-L (fixed wavelength)	29011360
UV flow cell 2 mm for U9-L	29011325
UV flow cell 5 mm for U9-L	18112824

Conductivity monitors

Item	Code no.
Conductivity Monitor **C9** (standard module)	28956495
Conductivity monitor (**C9n**) (optional)	29011363

External air sensors

Item	Code no.
Air Sensor **L9-1.2** mm	28956502
Air Sensor **L9-1.5** mm	28956500

I/O box

Item	Code no.
I/O-box E9	29011361

Module components

Item	Code no.
Module Panel	29011364
Dummy Module	28956493
Extension Box	29110806

Cables

Item	Code no.
Jumper 1 IEC 1394 (F-type)	28956489
External module cable, short (F-type)	29012474
External module cable, long (F-type)	29011366

Flow restrictor

Item	Code no.
Flow Restrictor FR-902	18112135

Barcode scanner

Item	Code no.
Barcode Scanner 2-D with USB	28956452

UniTag

Item	Code no.
UniTag (sheet with 108 labels)	28956491

Trays

Item	Code no.
Wet side waste tray	28956487
Front side waste tray	28956485

User Documentation

Item	Code no.
ÄKTA avant User Manual ***Note:*** *Covers ÄKTA avant 25 and ÄKTA avant 150.*	29035184

8.6 Health and Safety Declaration Form

On site service

cytiva

On Site Service Health & Safety Declaration Form

Service Ticket #:	

To make the mutual protection and safety of Cytiva service personnel and our customers, all equipment and work areas must be clean and free of any hazardous contaminants before a Service Engineer starts a repair. To avoid delays in the servicing of your equipment, complete this checklist and present it to the Service Engineer upon arrival. Equipment and/or work areas not sufficiently cleaned, accessible and safe for an engineer may lead to delays in servicing the equipment and could be subject to additional charges.

Yes	No	Review the actions below and answer "Yes" or "No". Provide explanation for any "No" answers in box below.
○	○	**Instrument has been cleaned of hazardous substances**. Rinse tubing or piping, wipe down scanner surfaces, or otherwise make sure removal of any dangerous residue. Make sure the area around the instrument is clean. If radioactivity has been used, perform a wipe test or other suitable survey.
○	○	**Adequate space and clearance is provided to allow safe access** for instrument service, repair or installation. In some cases this may require customer to move equipment from normal operating location prior to Cytiva arrival.
○	○	**Consumables, such as columns or gels, have been removed or isolated from the instrument and from any area that may impede access to the instrument.**
○	○	**All buffer / waste vessels are labeled. Excess containers have been removed from the area to provide access.**
Provide explanation for any "No" answers here:		

Equipment type / Product No:		**Serial No:**	
I hereby confirm that the equipment specified above has been cleaned to remove any hazardous substances and that the area has been made safe and accessible.			
Name:		**Company or institution:**	
Position or job title:		**Date (YYYY/MM/DD):**	
Signed:			

Product return or servicing

Health & Safety Declaration Form for Product Return or Servicing

Return authorization number:		*and/or* **Service Ticket/Request:**	

To make sure the mutual protection and safety of Cytiva personnel, our customers, transportation personnel and our environment, all equipment must be clean and free of any hazardous contaminants before shipping to Cytiva. To avoid delays in the processing of your equipment, complete this checklist and include it with your return.

1. Note that items will NOT be accepted for servicing or return without this form
2. Equipment which is not sufficiently cleaned prior to return to Cytiva may lead to delays in servicing the equipment and could be subject to additional charges
3. Visible contamination will be assumed hazardous and additional cleaning and decontamination charges will be applied

Yes	No	Specify if the equipment has been in contact with any of the following:	
◯	◯	Radioactivity (specify)	
◯	◯	Infectious or hazardous biological substances (specify)	
◯	◯	Other Hazardous Chemicals (specify)	

Equipment must be decontaminated prior to service / return. Provide a telephone number where Cytiva can contact you for additional information concerning the system / equipment.

Telephone No:		
Liquid and/or gas in equipment is:	☐	**Water**
	☐	**Ethanol**
	☐	**None, empty**
	☐	**Argon, Helium, Nitrogen**
	☐	**Liquid Nitrogen**
	Other, specify	

Equipment type / Product No:		**Serial No:**	

I hereby confirm that the equipment specified above has been cleaned to remove any hazardous substances and that the area has been made safe and accessible.

Name:		**Company or institution:**	
Position or job title:		**Date (YYYY/MM/DD)**	
Signed:			

To receive a return authorization number or service number, call local technical support or customer service.

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For local office contact information, visit cytiva.com/contact

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