# ÄKTAexplorer<sup>TM</sup>, ÄKTApurifier<sup>TM</sup> and ÄKTAmicro<sup>TM</sup>

**Operating Instructions** 







# Table of Contents

1	Intr	oduction
	1.1	Important user information
	1.2	Regulatory information
	1.3	Instrument
	1.4	Control software
2	Safe	ety instructions
	2.1	Safety precautions
	2.2	Labels
	2.3	Emergency procedures
	2.4	Recycling information
3	Inst	allation
	3.1	Site requirements
	3.2	Transport
	3.3	Unpacking
	3.4	Assembly
	3.5	Connections
	3.6	Spare parts and accessories
4	Ope	ration
	4.1	Operation overview
	4.2	Starting the instrument
	4.3	Starting the control system
	4.4	Preparations before start
	4.5	Setting up a run
	4.6	Performing a run
	4.7	Procedures after a run
5	Mai	ntenance
	5.1	General
	5.2	User maintenance schedule
	5.3	Cleaning
	5.4	Component maintenance
	5.5	Disassembly and assembly of components and consumables
	5.6	Replacement of fuses
	5.7	Calibration
	5.8	Storage
6	Tro	ubleshooting
_	6.1	UV curve problems
	6.2	Conductivity curve problems
	6.3	pH curve problems
	6.4	Pressure curve problems
	0. 1	resource out to producting

### Table of Contents

7	Refe	erence information	59
	7.1	Specifications	59
	7.2	Chemical resistance	59
	7.3	System recommendations	63
	7.4	Literature	63
	7.5	Ordering information	63
Α	Elec	trical and communication connections	64
В	Con	nection diagram - Liquid flow path	65
С	Tub	ing	69

# 1 Introduction

# Purpose of the Operating Instructions

The Operating Instructions provides you with the instructions needed to handle the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro systems in a safe way.

#### **Prerequisites**

In order to operate the systems in the way they are intended, the following pre-requisites must be fulfilled:

- You should have a general understanding of how the PC and Windows™ works.
- You should understand the concepts of liquid chromatography.
- You must read and understand the Safety Instructions.
- The instrument and software should be installed, configured and calibrated according to the Installation Guide.

## In this chapter

This chapter contains important user information, and a general description of the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro systems and their intended use.

# 1.1 Important user information

# Read this before using the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro system



All users must read the safety instructions in the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro User Documentation to fully understand the safe use of the systems, before installing, using, or maintaining the systems.

Do not operate the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro systems in any other way than described in the user documentation. Otherwise, you may be exposed to hazards that can lead to personal injury, and you may cause damage to the equipment.

#### Intended use

ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro are liquid chromatography systems intended for protein purification within method development and drug discovery. The systems can be used to screen for optimal choice of columns, media and running parameters to purify selected proteins.

The ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro systems are intended for research use only, and shall not be used in any clinical procedures, or for diagnostic purposes.

# Safety notices

This user documentation contains WARNINGS, CAUTIONS and NOTICES concerning the safe use of the product. See definitions below.

#### Warnings



#### 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.

#### **Cautions**



#### **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.

#### **Notices**



#### 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.

# **Typographical conventions**

Software items are identified in the text by **bold italic** text. A colon separates menu levels, thus **File:Open** refers to the **Open** command in the **File** menu. Hardware items are identified in the text by **bold** text (e.g., **Power** switch).

# 1.2 Regulatory information

This section describes the directives and standards that are fulfilled by the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro systems.

# **Manufacturing information**

The Declaration of Conformity includes the following information:

Requirement	Content
Name and address of manufacturer	GE Healthcare Bio-Sciences AB Björkgatan 30, SE-751 84 Uppsala, Swe- den
Name and address of person responsible for Technical File	Peter Löwendahl, Björkgatan 30 SE-751 84 Uppsala, Sweden
Name and ID of notified body	INTERTEK SEMKO AB, NB 0413
Place and date of declaration	Uppsala, Sweden, May 2009
Identity of person authorized to sign DoC	Peter Löwendahl See EC Declaration of Conformity

# **CE Conformity**

This product complies with the European directives listed in the table, by fulfilling the corresponding harmonized standards. A copy of the Declaration of Conformity is available on request.

Directive	Title
2006/42/EC	Machinery Directive (MD)
2006/95/EC	Low Voltage Directive (LVD)
2004/108/EC	ElectroMagnetic Compatibility (EMC) Directive

### **International standards**

This product fulfills the requirements of the following standards:

Standard	Description	Notes
EN 61010-1, IEC 61010-1, UL 61010-1, CAN/CSA-C22.2	Safety requirements for electrical equipment for measurement, control, and laboratory use	
No. 61010-1		

Standard	Description	Notes
EN 61326-1	EMC emissions and immunity requirements for electrical equipment for measurement, control and laboratory use	Harmonized with 2004/108/EC
EN-ISO 12100-1, 12100-2	Safety of machinery - Basic concepts, general principles for design	Harmonized with 2006/42/EC
EN-ISO 14121-1, 14121-2	Safety of machinery - Principles of risk assessment	Harmonized with 2006/42/EC

### **CE Marking**

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

- used as a stand-alone unit. or
- connected to other CE marked instruments, or
- connected to other products recommended or described in the user documentation, and
- used in the same state as it was delivered from GE Healthcare, except for alterations described in the user documentation.

The Declaration of conformity is valid only for systems that are marked with the CE-marking.



# Regulatory compliance of connected equipment

Any equipment connected to the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro systems should meet the safety requirements of EN 61010-1/IEC 61010-1, or relevant harmonized standards. Within EU, connected equipment must be CE marked.

# 1.3 Instrument

# **Product description**

ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro are high pressure liquid chromatography systems for use in laboratory scale production of biomolecules.

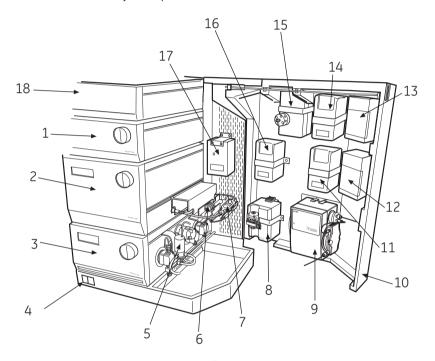


Figure 1.1: The main parts of the instruments. ÄKTAexplorer is shown as an example.

Part	Function
1	Monitor pH/C-900
2	Monitor UV-900
3	Pump P-900 (P-901 alt. P-903)
4	Power switch
5	Switch valve (SV-903)
6	Conductivity cell
7	UV cell

Part	Function
8	Mixer M-925
9	Sample pump P-960 (This is only standard in some versions)
10	Valve door
11	Column selection valve, V3 (PV-908)
12	Sample valve, V5 (PV-908)
13	Injection valve, V1 (INV-907)
14	Column selection valve, V2 (PV-908)
15	Flow direction valve, V7 (INV-907). This is only standard for ÄKTAExplorer 100 and ÄKTAExplorer 100 Air
16	Outlet valve, V4 (PV-908)
17	Buffer valve, V6 (INV-908)
18	Box 900

Detailed information on the components included in each system can be found in their respective User Manual.

# Basic flow path

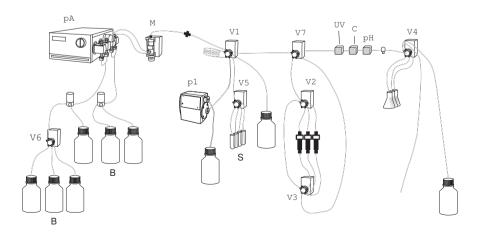


Figure 1.2: Basic flow path. ÄKTAexplorer is shown as an example.

#### 1.3 Instrument

Step	Part	Description
1	В	Buffers from container pass through Buffer valve V6 and/or the switch valve.
2	S	Sample pass through sample valve V5 which selects a sample depending on the setting in the control software.
3	рА	Pump A pumps buffer through the system.
4	М	The buffers pass through Mixer M where they are mixed.
5	V1, p1	This is where the sample is added to the flow path. The sample can be added manually with a suitable syringe or pumped in via the sample pump p1 from a sample chosen via sample valve V5 or an autosampler.
6	V7	The flow direction valve V7 is optional in some systems and used to select the direction of the flow through column.
7	V2, V3	Column selection valves V2 and V3 direct flow through a specified column.
8	V7, UV, C, pH	Liquid returns to flow direction valve V7 and redirects flow to outlet valve via pH, UV and Condictivity monitors.
9	V4	The outlet valve directs the flow either to waste, to fraction collection containers or to a fraction collector device such as Frac- 950.

The flow path is likely to include air sensors, flow restrictors, online filters, sample loops and so on which vary in number and application depending on the system and its strategy.

Fore more details on liquid flow path, see Appendix B Connection diagram - Liquid flow path, on page 65

# 1.4 Control software

#### UNICORN™ control software

UNICORN is a complete software for control and supervision of ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro. The software runs under Microsoft™ Windows operating system.

UNICORN is supplied with a method wizard which provides easy creation of methods for purification.

For more information about UNICORN control system, see the UNICORN user manuals supplied.

# 2 Safety instructions

This chapter describes safety compliance, safety labels, general safety precautions, emergency procedures, power failure and recycling of ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro.

# 2.1 Safety precautions

#### Introduction

The ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro instruments are powered by mains voltage and handles pressurized liquids 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 injury or damage to the equipment.

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

- General precautions
- Using flammable liquids
- Personal protection
- Installing and moving the instrument
- System operation
- Maintenance

# **General precautions**

Always follow these General precautions to avoid injury when using the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro instrument.



#### WARNING

Do not operate the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro instrument in any other way than described in the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro and UNICORN manuals.



#### WARNING

Operation and user maintenance of the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro instrument should be performed by properly trained personnel only.



#### WARNING

Before connecting a column to the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro instrument, 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 of the column.



#### WARNING

Do not use any accessories not supplied or recommended by GE Healthcare.



#### WARNING

Do not use the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro instrument if it is not working properly, nor 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



#### CAUTION

Waste tubes and containers shall be secured and sealed to prevent accidental spillage.



#### CAUTION

Make sure waste container is dimensioned for maximum possible volume when the instrument is left unattended.



#### NOTICE

Avoid condensation by letting the unit equilibrate to ambient temperature.

# Using flammable liquids

When using flammable liquids with the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro instrument, follow these precautions to avoid any risk of fire or explosion.



#### WARNING

**Fire Hazard**. Before starting the system, make sure that there is no leakage.



#### WARNING

A fume hood or similar ventilation system shall be installed when flammable or noxious substances are used.

# **Personal protection**



#### WARNING

Always use appropriate personal protective equipment during operation and maintenance of ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro system.



#### WARNING

**Hazardous substances.** When using hazardous chemical and biological agents, take all suitable protective measures, such as wearing protective glasses and gloves resistant to the substances used. Follow local and/or national regulations for safe operation and maintenance of the system.



#### WARNING

**Spread of biological agents**. The operator has to take all necessary actions to avoid spreading hazardous biological agents in the vicinity of the instrument. The facility should comply with the national code of practice for biosafety.



#### WARNING

**High pressure.** The ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro instrument operates under high pressure. Wear protective glasses at all times.

# Installing and moving the instrument



#### WARNING

**Supply voltage.** Make sure that the supply voltage at the wall outlet corresponds to the marking on the instrument, before connecting the power cord.



#### WARNING

**Protective ground.** The ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro instrument must always be connected to a grounded power outlet.



#### WARNING

**Power cord.** Only use power cords delivered or approved by GE Healthcare.



#### WARNING

Access to power switch and power cord. Do not block the rear and side panel of the instrument. The **Power** switch must always be easy to access. The power cord must always be easy to disconnect.



#### WARNING

**Installing the computer**. The computer should be installed and used according to the instructions provided by the manufacturer of the computer.



#### **CAUTION**

**Heavy object.** Use suitable lifting equipment when moving the systems. Three people are required to lift the system safely.



#### NOTICE

**Disconnect power.** To prevent equipment damage, always disconnect power from the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro instrument before an instrument module is removed or installed, or a cable is connected or disconnected.



#### NOTICE

ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro shall be installed and prepared by GE Healthcare personnel.

#### **System operation**



#### 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.



#### WARNING

**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.



#### CAUTION

**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.



#### NOTICE

Do not run **Column CIP** if using silica based packing material or RPC columns. Remove the column from the system during CIP.

#### Maintenance



#### WARNING

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



#### WARNING

**Disconnect power.** Always disconnect power from the instrument before replacing any component on the instrument, unless stated otherwise in the user documentation.



#### WARNING

**Hazardous chemicals during maintenance.** When using hazardous chemicals for system or column cleaning, wash the system or columns with a neutral solution in the last phase or step.



#### WARNING

Do not perform any type of maintenance work while the system is powered electrically or when the piping system is pressurized. Note that the piping system can be pressurized even when the system is closed down.



#### WARNING

Only spare parts that are approved or supplied by GE Healthcare may be used for maintaining or servicing the system.



#### WARNING

Make sure that the piping system is completely leakage free before performing any CIP on the system.



#### WARNING

NaOH is corrosive and therefore dangerous to health. When using hazardous chemicals, avoid spillage and wear protective glasses and other suitable personal protective equipment.



#### WARNING

After assembly, the piping system must be tested for leakage at maximum pressure for continued protection against injury risks due to fluid jets, burst pipes or explosive atmosphere.



#### WARNING

Before disassembly, check that there is no pressure in the piping system.



#### WARNING

**Disconnect power.** Always disconnect power from the instrument before replacing fuses.



#### WARNING

Decontaminate the equipment before decommissioning to ensure that hazardous residues are removed.



#### NOTICE

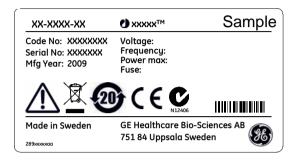
**Cleaning**. Keep the instrument dry and clean. Wipe regularly with a soft damp tissue and, if necessary, a mild cleaning agent. Let the instrument dry completely before use.

# 2.2 Labels

This section describes the safety labels and labels concerning hazardous substances that are attached to the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro instruments. For information about marking of the computer equipment, refer to the manufacturer's instructions.

# Labels on the instrument

The illustration below shows an example of the identification label that is attached to the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro instruments.



# Symbols used in safety labels

Label	Meaning
$\triangle$	<b>Warning!</b> Read the user documentation before using the system. Do not open any covers or replace parts unless specifically stated in the user documentation.
C	The system complies with the requirements for electromagnetic compliance (EMC) in Australia and New Zealand.
CE	The system complies with applicable European directives.

# Labels concerning hazardous substances

Label	Meaning
<b>X</b>	This symbol indicates that the waste of electrical and electronic equipment must not be disposed as unsorted municipal waste and must be collected separately. Please contact an authorized representative of the manufacturer for information concerning the decommissioning of equipment.
<b>@</b>	This symbol indicates that the product contains hazardous materials in excess of the limits established by the Chinese standard SJ/T11363-2006 Requirements for Concentration Limits for Certain Hazardous Substances in Electronics.

# 2.3 Emergency procedures

This section describes how to do an emergency shutdown of the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro systems. The section also describes the result in the event of power failure.

# **Emergency shutdown**

In an emergency situation, do as follows to stop the run:

Step	Action
1	To pause the run from UNICORN, click the <i>Pause</i> button in <i>System Control</i> .  Pause
2	If required, switch off power to the instrument by pressing the $\bf Main\ power$ switch to the $\bf 0$ position.

#### Power failure

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

Power failure to	will result in
ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro sys-	The run is interrupted immediately, in an undefined state
tem	The data collected up to the time of the power failure is available in UNICORN
Computer	The UNICORN computer shuts down in an undefined state
	The run continues, but data cannot be saved in UNI- CORN.

# 2.4 Recycling information

The equipment shall be decontaminated before decommissioning and all local regulations shall be followed with regard to scrapping of the equipment.

# Disposal, general instructions

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

# Recycling of hazardous substances

ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro instruments contain hazardous substances. Detailed information is available from your GE Healthcare representative.

# Disposal of electrical components

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



# 3 Installation



#### NOTICE

ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro shall be installed and prepared by GE Healthcare personnel.

ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro are delivered in protective packing material and shall be unpacked with great care.

Any equipment connected to ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro must fulfill applicable standards and local regulations.

For detailed information on Installation, see ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro Installation Guides.

# 3.1 Site requirements

Parameter	Requirement
Electrical power	100-240 V, 50-60 Hz
Ambient temperature	4°C to 40°C
Placement	Stable laboratory bench min. 200 × 80 cm
Humidity	20 to 95%, non-condensing

# 3.2 Transport

The equipment weights are specified in the table below. Each system requires at least three people to lift and move it unless a suitable lifting device is used.

Instrument	Weight
ÄKTAexplorer	66 kg
ÄKTApurifier	41 kg
ÄKTAmicro	55 kg

The instrument can be transported on a trolley or a suitable lifting device capable of supporting the weight of the instrument.

#### NOTICE

Lift the system in the upright position. Do not use the front panel bar as a lifting handle.

Before moving the system:

- disconnect all cables and tubing connected to peripheral components and liquid containers.
- remove all items from the top of the system.
- close the valve door completely (only for ÄKTAexplorer).
- grasp the system firmly by placing the fingers in the gap between the swivel platform and the base of the main unit and lift.

For more information on Transport, see ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro Installation Guides.

# 3.3 Unpacking

- Check the equipment for damage before starting assembly and installation.
- Document any damage and contact your local GE Healthcare representative.

Remove straps and packing material and stand equipment upright on swivel foot before starting installation.

# 3.4 Assembly

The following parts must be added to the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro instruments before they can be used:

- pH electrode (optional)
- Waste tube
- CU-950 Control unit between unit and computer
- Various buffer and sample bottles

#### Installing the pH electrode

Install the pH electrode in the flow cell according to the picture below.

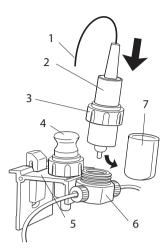


Figure 3.1: Main parts of the pH electrode and holder.

No.	Description	No.	Description
1	To instrument rear "pH probe"	5	Flow cell holder
2	Electrode	6	Flow cell
3	Nut	7	End cover
4	Dummy		

The flow cell holder can be placed on either the optical unit (for ÄKTApurifier) or on the outside of the valve door (for ÄKTAexplorer). For more information on installation, see ÄKTApurifier User Manual and ÄKTAexplorer Installation Guide.

# 3.5 Connections

#### Communication

Connect the network, signal cables and computer according to the electrical drawings in Appendix A Electrical and communication connections, on page 64.

Ensure that UNICORN control software is installed on the computer.

#### **Installing Controller unit CU-950**

Hang the CU-950 on the left side of the system by inserting the hooks on the front of CU-950 into the channel on the side of the UV-900.

Connect according to diagram in Appendix A Electrical and communication connections, on page 64

When using a fraction collector:

- 1 connect a UniNet-1 cable between Monitor UPC-900/Monitor UV-900 and the fraction collector.
- 2 connect a termination plug to the empty UniNet-1 socket (Frac-950 only).

### Flow path

All parts and tubing are mounted on the ÄKTAexplorer and ÄKTAmicro systems at delivery. ÄKTApurifier has no preconnected tubing. It is recommended that the mounting is done by GE Healthcare service engineers. For more information regarding installation, see ÄKTApurifier User Manual.

#### Setting the delay volume in UNICORN

The delay volume is volume of liquid in the flow path from the UV sensor that identifies the peak to the fraction collector. The length of tubes affects the delay volume that needs to be changed in UNICORN.

To change the delay volume in UNICORN:

- 1 Select **System:Settings** in **System Control**.
- 2 Select **Specials** and then **FracParameters**.
- 3 Enter the delay volume and click **OK**.

**Note:** To prevent bacterial growth, the system flow path is filled with 20% ethanol at delivery.

 Connect tubes for reagents, solvents and sample collection to the correct inlet and outlet connections on the system. For more information, see ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro User Manuals.

## **Electrical power**

Connect the power cord to a grounded power outlet specified in Section 3.1 Site requirements, on page 26.

# 3.6 Spare parts and accessories

For correct up to date information on spare parts and accessories visit: www.gelifesciences.com/AKTA

# 4 Operation

This chapter provides instructions for the use of ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro.

# 4.1 Operation overview

#### Workflow

The typical workflow in ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro, after turning on the system and connecting it to UNICORN, can be divided into a number of steps.

Step	Action	Section
1	Create a method	Create a method, on page 35
2	Prepare the system for a run	Section 4.4 Preparations before start, on page 32
3	Start a run using a method	Section 4.6 Performing a run, on page 39
4	During a run - view and change parameters	Viewing the run, on page 40
5	Procedures after a run	Section 4.7 Procedures after a run, on page 42
6	Evaluate the results	See UNICORN user documentation.

# Liquid flow path

See Appendix B Connection diagram - Liquid flow path, on page 65 for an illustration of the liquid flow path in ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro.

# 4.2 Starting the instrument

Ensure that the waste container and needed buffer bottles are correctly connected. Check that all tubing connections are properly tightened and that all valves are connected to a tube or termination.

Turn on the **Power** switch on the instrument.

# 4.3 Starting the control system

### **Starting UNICORN**

- 1 Turn on the monitor, computer and optional printer according to the manufacturer's instructions. Wait for the computer to start up.
- 2 Verify that the power indicator on the CU-950 is on when the computer has been turned on.
- 3 Log on to Windows.
- 4 Start UNICORN by double-clicking on the UNICORN shortcut icon on the Windows desktop.

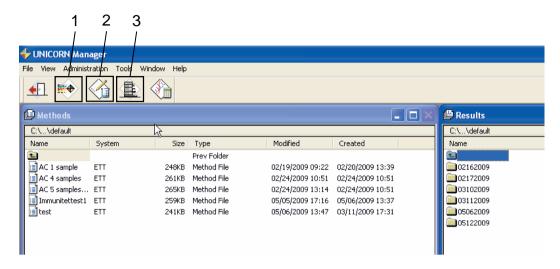


In the Logon dialog, select a user from the User name list and enter the password. If you log on for the very first time, select user default and enter the password default. Click OK.



UNICORN starts and the UNICORN Manager window opens, see Figure 4.1.

**Note:** See the UNICORN user documentation for instructions about how to create new users.



No.	Description
1	The <i>Instant Run</i> icon immediately starts the system control wizard used to start a run.
2	The <b>New Method</b> icon opens the <b>Method Editor</b> module and displays the <b>New Method</b> dialog box.
3	The <b>System Control</b> icon activates the <b>System Control</b> module and displays the <b>Manual instruction</b> dialog box.

Figure 4.1: The UNICORN Manager window.

# Control system in UNICORN

To open the **System Control** module in UNICORN, click the **System Control** icon in the **UNICORN Manager** window, see *Figure 4.1*.

# 4.4 Preparations before start

# Prepare buffers, solutions and inlets

1 Prepare buffers and solutions required for the run.

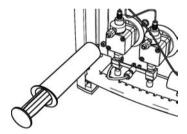
2 Immerse all inlet tubing in the appropriate liquid containers as described in the method.

# Purging the pump and inlet tubing

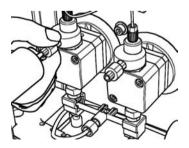
Fill the pump and inlet tubing with liquid if small amounts of air need to be removed or if the inlet tubing is empty.

To fill the inlet tubing manually in **System Control**:

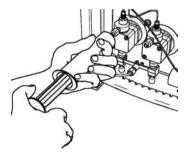
- 1 Make sure that no method has been started.
- 2 Set a low flow in **System Control:Manual:Pump:Flow**, for example 0.5 ml/min.
- 3 Click **Execute**.
- 4 Set the inlet valve to the appropriate position in **System Control:Manual:Flowpath:InletValve**, for example inlet **A11** to **A18** or **A2**.
- 5 Connect a syringe to the purge valve.



6 Turn the purge valve counter clockwise half a turn to open it.



7 Slowly draw solution into the syringe. When fluid starts to enter the syringe, continue to draw a few milliliters before closing the purge valve. Check that there is no visible air left in the tubing.



- 8 Repeat for the other purge valve.
- 9 To fill inlet **B1** and **B2**:
  - a In **System Control:Manual:Pump:Gradient**, select **Target 100% B** and inlet **B1** to fill **B1** or inlet **B2** to fill **B2**. Wait for the valve to turn (a clicking sound) before starting the purging procedure.
  - b When all inlets are filled, click **End**.

# Connect columns and Superloop™

For column positions, see the method.

#### Remove air before connecting columns

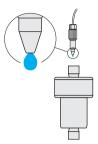
Air remaining in the system may be removed by purging the pump and by selecting *Pump Wash* and *System Wash*.

- 1 Immerse **A1** tubing in the buffer to be used.
- 2 Select **System Control:Manual:Pump:PumpWash**.

#### Connecting tubing to columns

Refer to column manufacturer's instructions.

### Column attachment drop-to-drop



Attach columns manually by starting a low flow (see *Purging the pump and inlet tubing, on page 33*) and selecting *System Control:Manual:Flowpath:ColumnPosition*.

### Preparing the fraction collector

Place the rack chosen in the method in the fraction collector and fill it with appropriate tubes and/or deep well plates.

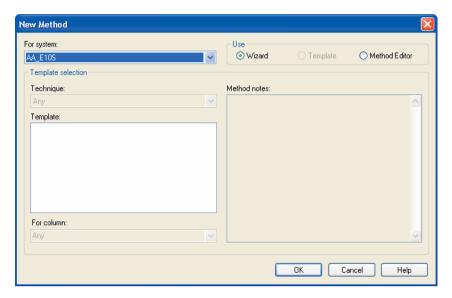
# 4.5 Setting up a run

#### Create a method

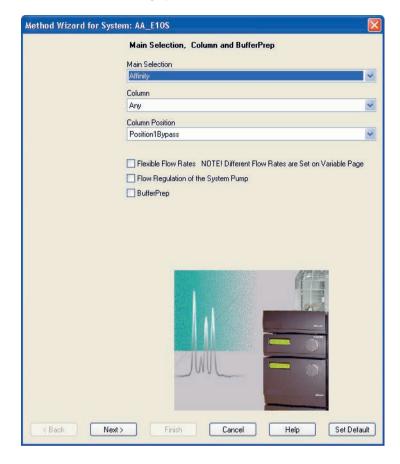
To create a method:

1 Click the **New Method** icon in the **UNICORN Manager** window, see Figure 4.1.

### The **New Method** dialog opens.



- 2 If several systems are available, select which system to use in the *For system* list box.
- 3 Select *Wizard* to create a method using the *Method Wizard*. Click **OK**.



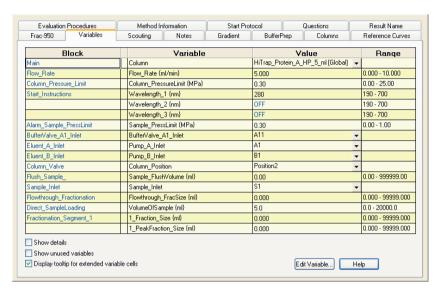
#### The *Method Wizard* dialog opens.

#### 4 In the **Method Wizard** dialog:

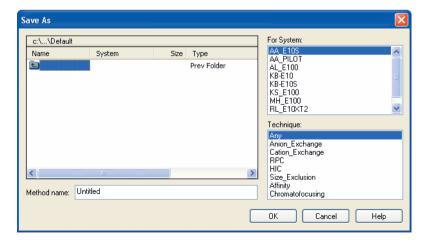
- Select the appropriate chromatographic technique in the Main Selection list box, for example Affinity.
- Select the column you intend to use in the *Column* list box. The correct column volume, the recommended flow rate, and the correct pressure limit for that column will then be automatically implemented in the method.
- Select column position in the *Column Position* list box.
- If required, select *Flexible Flow Rates* and/or *Flow Regulation of the System Pump* and/or *BufferPrep*.
- 5 Click **Next**.
- 6 On each new page in the **Method Wizard**, select the appropriate parameters and click **Next** to continue

7 On the last page, click *Finish*.

The *Run Setup* dialog opens with the *Variables* tab selected by default.



- 8 The method is represented by a number of blocks on the *Variables* tab. The blocks are typical steps in a chromatographic run.
  - Each block contains a number of method variables. If necessary, change the variables to suit your application.
- 9 In the *Method Editor*, select *File:Save As* to save the method. The *Save As* dialog opens.



#### 10 In the Save As dialog:

- Enter method name in the *Method name* field and select folder to save the method in
- If you have more than one system connected to the computer, select system in the *For System* area for which the method is intended.
- Select technique in the **Technique** area for which the method was written.

#### 11 Click **OK**

The method is saved. It can now be started from the **System Control** module.

## 4.6 Performing a run

#### 1 Select method

- a In **System Control**, select **File:Run**.
- b Select the required **Method** from the list.

#### 2 Specify variables

Enter identification names for the samples via the keyboard.

#### 3 Edit result file location and names.

If required, edit the folder path and file names of the result files to be created.

#### 4 Preparations completed?

Ensure that the preparations according to Section 4.4 Preparations before start, on page 32 has been performed.

#### 5 Check the flow path

Make sure that:

- there is enough buffer available
- the correct inlet is placed in each buffer
- the outlets are placed in correct bottles
- the columns are placed in correct positions
- the chosen fraction collector rack is filled and is in correct place.

#### 6 Prepare the samples

The samples should have been prepared and clarified using centrifugation and/or filtration through a 0.45 µm filter <sup>1</sup>.

1 If using HisTrap™ FF crude, clarification is not needed.

- a Place the sample in chosen liquid container, vials for the autosampler or fill the capillary loop or the Superloop with sample depending on chosen method.
   Make sure that no air enters the tubing. Place the tubing close to the bottom of the liquid container but not too tight against the bottom.
- b Secure the tubing.

#### 7 Final check

Perform a final check that tubing, columns, solutions and buffers are placed according to the method

#### 8 Start the run

Click **START** to start the run on the selected systems.

#### Viewing the run

The progress of the run can be viewed in detail in the **System Control** module.



Up to four view panes, *Run Data*, *Curves*, *Flow scheme* and *Logbook* can be displayed showing different aspects of the run in real-time.

- The **Run Data** view pane displays the current values for selected run parameters.
- The *Curves* view pane displays the monitor signal values graphically.
- The Flow scheme view pane displays a graphical representation of the chromatography system that shows the current status of the run. During a run, the flow scheme shows open flow path(s) in color and monitor signals with numerical displays.
- The *Logbook* view pane shows the actions as the run proceeds. All actions and unexpected conditions are logged, with date, time and current user name. The log book provides a complete history of the run and is saved in the result file.

#### Customize the view panes

To customize the view panes, right-click in the respective view pane and select **Properties**. For more information about customizing the view panes, see the UNICORN user documentation

#### **Ending the run**

To stop the run on a system before it is finished:

Click **End** above the **Run data** view pane.

#### Status indicator colors

The status indicator is located at the bottom of **System Control**.

The table below shows how the indicator colors relate to the run status.

Indicator color	Run status
White	End
Green	Run or Manual
Yellow	Hold
Red	Pause

#### **Error** indication

When a warning or an alarm is issued from a system, an error code is displayed. See ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro User Manuals for guidance.

#### **Evaluate the results**

See ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro User Manuals and UNICORN user documentation for how to evaluate the results

## 4.7 Procedures after a run

#### Cleaning system

To keep the system in good shape, it is important to clean both the tubing and the outside of the system regularly.

- 1 In the *Method Editor* module in UNICORN, create a method for cleaning the system.
- 2 Wash the outside of the inlet tubings with water and/or ethanol.
- 3 Immerse the tubing ends to be used in the container with cleaning solution.
- 4 If the column valve is to be cleaned, remove the columns and reconnect the tubings to the column valves.
- 5 Run the cleaning method as described in Section 4.6 Performing a run, on page 39.

## **Cleaning columns**

When running different types of purification methods and different samples after each other, the columns should be cleaned between the runs according to the column instructions. This will remove unspecific bound proteins and prevent column clogging.

- 1 In the *Method Editor* module in UNICORN, create a method for column cleaning in place (CIP).
- 2 Immerse the tubing ends to be used in the correct containers according to the method for the chosen run.
- 3 Run the cleaning in place method as described in Section 4.6 Performing a run, on page 39.

# 5 Maintenance

This chapter provides instructions for routine component maintenance and a maintenance schedule.

#### 5.1 General

Regular maintenance is important for safe and trouble-free operation of your instrument. The user should perform daily and monthly maintenance. Preventive maintenance should be performed on a yearly basis by qualified service personnel.

For maintenance of a specific component, carefully read the component manual and follow the instructions. To avoid personal injury when performing maintenance on the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro instrument, follow the instructions below.



#### WARNING

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



#### WARNING

**Disconnect power.** Always disconnect power from the instrument before replacing any component on the instrument, unless stated otherwise in the user documentation.



#### WARNING

**Hazardous chemicals during maintenance.** When using hazardous chemicals for system or column cleaning, wash the system or columns with a neutral solution in the last phase or step.



#### WARNING

Do not perform any type of maintenance work while the system is powered electrically or when the piping system is pressurized. Note that the piping system can be pressurized even when the system is closed down.



#### NOTICE

When using hazardous chemicals, take all suitable protective measures, such as wearing protective glasses and gloves resistant to the chemicals used. Follow local regulations and instructions for safe operation and maintenance of the system.



#### NOTICE

**Cleaning**. Keep 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.

## 5.2 User maintenance schedule

Table 5.1 provides a guide to maintenance operations and intervals at which these operations should be performed by the user. The user is however responsible for deciding the type of operations and length of intervals necessary to maintain system function and safety.

Table 5.1: User maintenance schedule

Interval	Action	Instructions/reference
Daily	Leak inspection	Visually inspect the system for leaks.
	Wash the system flow path	1 For cleaning the flow path, see Cleaning-In- Place, on page 48.
		2 For leaving the system for a few days, see Section 5.8 Storage, on page 50.
	Calibrate pH electrode (optional)	Calibrate the pH electrode (if applicable) according to Monitor pH/C-900 User Manual.

Interval	Action	Instructions/reference
Weekly	Check inlet filters	Check the inlet filters visually and replace them if necessary.
	Replace on-line filter (if applicable)	Replace the on-line filter.
	Change pump rinsing solution	Change rinsing solution. Always use 20% ethanol with 10 mM NaOH as rinsing solution.
		If the volume of rinsing solution in the storage bottle has increased, it can be an indication of internal pump leakage. Replace the piston seals according to the User manual.
		If the volume of rinsing solution in the storage bottle has decreased significantly, check if the rinsing system connectors are mounted proper- ly.
		If the rinsing system connectors are not leaking, the rinsing membranes or piston seals may be leaking. Replace the membranes and piston seals according to the User manual.

Interval	Action	Instructions/reference
Monthly	Flow restrictor	Check that flow restrictor generates the following back-pressure:
		FR-904: 0.4 ±0.05 MPa
		FR-902: 0.2 ±0.05 MPa
		Check the back-pressure as follows:
		1 Disconnect the flow restrictor.
		2 Connect a capillary (approx. 1 m, i.d. 1 mm) to a free port in the injection valve. Set the valve manually to this port. Put the open end in a waste container.
		3 Run the pump at 10 ml/min with water. Note the back-pressure (Bp1) on the pump display, or in the Run Data window.
		4 Connect the flow restrictor to the open end of the capillary (observe the IN marking). Put the flow restrictor in the waste container.
		5 Run the pump at 10 ml/min with water. Note the back-pressure (Bp2) on the pump display, or in the Run Data window.
		6 Calculate the back-pressure generated by the flow restrictor. Replace it if it is not within limit.

Interval	Action	Instructions/reference
		1 Disconnect the flow restrictor.
		2 Connect a capillary (approx. 1 m, i.d. 1 mm) to the waste port (port 5) on the injection valve. Set the injection valve manually to Waste position. Put the open end in a waste container.
		3 Run the pump manually at 10 ml/min with water. Note the back-pressure (Bp1) on the pump display, or in the Run Data window.
		4 Set the system to Pause and connect the flow restrictor to the open end of the capillary (observe the IN marking). Put the flow restrictor in the waste container.
		5 Press Continue so that the pump run at 10 ml/min with water. Note the back-pressure (Bp2) on the pump display, or in the Run Data window.
		6 Calculate the back-pressure generated by the flow restrictor (Bp2-Bp1). Replace it if it is not within limit.
Yearly	Valve inspection	Check for external or internal leakage. Replace channel plate and distribution plate yearly or when required. Refer to the relevant valve instruction sheet.

# 5.3 Cleaning

# Cleaning before maintenance/service

Before maintenance/service is performed, the system owner must first clean the system and complete a Decontamination Report. Contact GE Healthcare for further information.

#### Cleaning-In-Place

All components in the system are designed for ease of CIP.

After repeated separation cycles, contaminating material might progressively build up in the system and on the column. This material may not have been removed by the cleaning step described above. The nature and degree of contamination depends on the sample and the chromatographic conditions employed. These should be considered when designing a cleaning protocol.

Routine cleaning should be performed at intervals aimed at prevention rather than cleaning the system from growth or contamination.



#### WARNING

Make sure that the piping system is completely leakage free before performing any CIP on the system.

Make sure that the process control method for cleaning flushes all possible flow paths in the system. After cleaning, rinse the entire system with water or suitable liquid until the piping/tubing system is completely free from the CIP solution (monitors in the system can be used as detectors). Do not leave NaOH or other cleaning agents in the system for long periods.



#### WARNING

**Hazardous chemicals during maintenance.** When using hazardous chemicals for system or column cleaning, wash the system or columns with a neutral solution in the last phase or step.



#### WARNING

NaOH is corrosive and therefore dangerous to health. When using hazardous chemicals, avoid spillage and wear protective glasses and other suitable personal protective equipment.

See also Section 5.8 Storage, on page 50.

## 5.4 Component maintenance

Maintenance and preventive replacement of parts of the major components are described in the respective manuals included in the system documentation.

The system documentation also includes a spare part list to be used to find common spare parts and their code numbers for ordering. This list can also be found online at www.gelifesciences.com/AKTA.

# 5.5 Disassembly and assembly of components and consumables

The operator must carefully read and understand the instructions supplied for each component before disassembly and assembly of the component. When replacing consumables, such as tubing and tubing connectors, all neccessary safety precautions must be taken. Contact your local GE Healthcare representative if further information or help is needed.



#### WARNING

**Disconnect power.** Always disconnect power from the instrument before replacing any component on the instrument, unless stated otherwise in the user documentation.



#### WARNING

Before disassembly, check that there is no pressure in the piping system.



#### WARNING

After assembly, the piping system must be tested for leakage at maximum pressure for continued protection against injury risks due to fluid jets, burst pipes or explosive atmosphere.

## 5.6 Replacement of fuses



#### WARNING

**Disconnect power.** Always disconnect power from the instrument before replacing fuses.

Refer to Section 7.1 Specifications, on page 59 for information about the fuse type and rating. If a fuse repeatedly blows, switch off the system mains switch and contact your local GE Healthcare representative.



#### WARNING

For continued protection from fire hazard, replace only with same type and rating of fuse.

## 5.7 Calibration

The table below lists the type and frequency of calibrations that can be done on the instrument. Refer to UNICORN user documentation and to the individual component User Manuals and Instructions for descriptions of how to perform these calibrations. The calibrations are performed from UNICORN by selecting *System:Calibrate* in *System Control*.

Component		How often
pH monitor (if a	pplicable)	Every day.
Pump (if applice	able)	Whenever the running conditions are changed, e.g. viscosity of sample or buffer, temperature, backpressure etc. If the sample pump is not used frequently it should be calibrated before use.
Pressure readin	g	When required.
Conductivity flow cell	Cell constant	Only necessary if specific conductivity with high accuracy is measured ( <i>Cond_Calib</i> ).
	Temperature	Must be done when changing the conductivity flow cell ( <i>Temp</i> ).
	Entering a new cell con- stant	Must be done when changing the conductivity flow cell ( <b>Cond_Cell</b> ).

## 5.8 Storage

#### **General recommendation**

For storage, the system must first be cleaned as described in *Cleaning-In-Place*, on page 48. After cleaning, the system must be filled with 0.01 M NaOH or 20% ethanol solution.

Columns and media shall be stored according to their respective instructions.

## **Storage conditions**

The following conditions shall be maintained while the system is in storage:

- Temperature: +2 to +30°C (preferably room temperature)
- Relative humidity: 0 to 95%, non-condensing (preferably low humidity).

After storage, clean the system, calibrate all monitors, and perform a leakage test before using the system.

# 6 Troubleshooting

# 6.1 UV curve problems

Error symptom	Possible cause	Corrective action
Ghost peak	Dirt or residues in the flow path from previous runs. Air in the eluents.	Clean the system. Make sure air is removed.
	Residue in the column from previous runs	Clean the column according to the column instructions.
	Incorrect mixer function	Check the mixer function by placing a stirrer bar on top of the mixer housing. The stirrer bar should rotate when the system is in <i>Run</i> mode. The mixer function can also be checked by running the installation test.
Noisy UV-signal, sig- nal drift or instability	Bad UV fiber connections	Check the connections of the UV cell optical fiber. Replace if necessary.
	Dirty UV cell	Clean the UV cell by flushing Decone 90, Deconex 11 or equivalent.
	Impure buffer	Check if the signal is still noisy with water.
	Air in the pump or in the UV cell	Purge the pump according to Pump User Manual. Run a system wash with buffer.

Error symptom	Possible cause	Corrective action
Low sensitivity	sitivity Aging UV lamp	Check the lamp run time according to and replace if necessary. Refer to ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro User Manuals.
	UV lamp in wrong position	Check that the lamp position and the filter position are both set to the wavelength to be used, 280 nm or 254 nm. Refer to ÄKTApurifier and ÄKTAmicro User Manuals.  Does not apply to ÄKTAexplorer and ÄKTApurifier without UPC.
	The theoretical extinction coefficient too low	Calculate the theoretical extinction coefficient of the protein. If it is zero or very low at 280 nm, the protein cannot be detected.

# 6.2 Conductivity curve problems

Error symptom	Possible cause	Corrective action
Baseline drift or noisy signal	Air in the pump or the flow cell	Use a flow restrictor after the flow cell.
	Leaking tube connections	Tighten the connectors. If necessary, replace the connectors.
	Incorrect mixer function	Check the mixer function by placing a stirrer bar on top of the mixer housing. The stirrer bar should rotate when the system is in <i>Run</i> mode. The mixer function can also be checked by running the installation test.
	Dirty conductivity cell	Clean the conductivity cell by flushing 1 M NaOH or 20% ethanol.
	Column not equilibrated	Equilibrate the column. If necessary, clean the column. Refer to ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro User Manuals.

Error symptom	Possible cause	Corrective action
Conductivity measure- ment with the same	Dirty flow cell	Clean the flow cell according to procedure in Monitor User Manual.
buffer appears to de- crease over time	Decrease in ambient temperature	Use a temperature compensation factor. See Monitor User Manual.
Waves on the gradi- ent	Incorrect pump function	Check that the pump is operating and is programmed correctly.
	Dirty mixing chamber	Check that the mixing chamber is free from dirt or particles.
	Insufficient mixing chamber volume	Change to a larger mixing chamber volume if necessary.
	Incorrect motor function	Check the motor operation. Place a hand on the mixer and start it by starting the pump at a low flow rate. You should both hear and feel the mixer motor and stirrer when they are spinning.
Ghost peaks appear in the gradient profile	Air in the flow cell	Check for loose tubing connections. Use the flow restrictor.
Unlinear gradients or slow response to %B	Dirty tubing	Wash the tubing and check pump is operating properly.
changes	Incorrect mixer vol- ume	Change to smaller mixer volume.

Error symptom	Possible cause	Corrective action
Incorrect or unstable reading	Loose connection of conductivity flow cable	Check that the conductivity flow cell cable is connected properly.
	Incorrect pump and valves function	Check that the pump and valves operate correctly.
	Incorrect temperature compensation factor	If temperature compensation is being used, check that the temperature sensor is calibrated, and that the correct temperature compensation factor is in use.
	Dirty or incorrectly equilibrated column	Check that the column is equilibrated. If necessary clean the column.
	Incorrect mixer function	Check the operation of the mixer. The mixer function is checked by placing a stirrer bar on top of the mixer housing. The stirrer bar should rotate when the system is in <i>Run</i> mode. The mixer function can also be checked by running the installation test.

# 6.3 pH curve problems

Error symptom	Possible cause	Corrective action
No response to pH changes	Faulty electrode con- nection	Check that the electrode cable is connected properly.
	Damaged electrode	The electrode glass membrane may be cracked. Replace the electrode.
	Incorrectly connected pH monitor	Check that the pH monitor is correctly connected according to the User Manual.
Small response to pH changes	Dirty pH electrode	Clean the pH electrode as detailed in Monitor pH/C-900 User Manual or UPC-900 User Manual.
		If the problem remains, replace the pH electrode.

# 6 Troubleshooting 6.3 pH curve problems

Error symptom	Possible cause	Corrective action
Slow pH response or Calibration impossible	Contaminated electrode glass membrane	Check the electrode glass membrane. If it is contaminated, clean the electrode following the instructions in Monitor pH/C-900 User Manual or UPC-900 User Manual.
	Membrane has dried out	If the membrane has dried out, the electrode may be restored by soaking it in buffer overnight.

Error symptom	Possible cause	Corrective action
Incorrect or unstable pH reading	Problem with electrode	Check that the electrode cable is connected properly.
		Check that the electrode is correctly inserted in the flow cell and, if necessary, hand-tighten the nut.
		Check that the pH electrode is not broken.
		Calibrate the pH electrode.
		Clean the pH electrode if required, see Monitor pH/C-900 User Manualor UPC-900 User Manual.
		Compare the response of the pH electrode with that of another pH electrode. If the response differ greatly, the electrode may require cleaning or replacement.
		In organic solvents such as ethanol, methanol and acetonitrile, stable pH measurements are not possible since dehydration of the membrane will occur. It is recommended that the pH electrode is not used in applications using organic solvents. Mount the dummy electrode instead.
	Incorrect pump or valve operation	Check that the pump and valves operate correctly.
	Air in the flow cell	If air in the flow cell is suspected, tap the flow cell carefully or tilt it to remove the air. Alternatively, flush the cell with buffer at 20 ml/min (ÄKTAexplorer 100 and ÄK-TApurifier 100) or 10 ml/min (ÄKTAexplorer 10 and ÄKTApurifier 10) or 0.5 ml/min (ÄKTAmicro) for 1/2 min. Use the flow restrictor FR-902 after the pH electrode.
	Static interference	

Error symptom	Possible cause	Corrective action
		There may be interference from static fields. Connect the pH flow cell and the rear panel of the monitor using a standard laboratory 4 mm "banana plug" cable.
pH values vary with varied back pressure	Problem with the electrode	Replace the pH electrode.

# 6.4 Pressure curve problems

Error symptom	Possible cause	Corrective action
Erratic flow, noisy baseline signal, irregu- lar pressure trace	Air bubbles passing through or trapped in the pump	Check all connections for leaks. Check that there is sufficient eluent present in the reservoirs. Use degassed solutions. Purge the pump. Follow the instructions in Pump P-900 User Manual.
	Inlet or outlet check valves not functioning correctly	Clean the valves in according to Pump P-900 User Manual.
	Piston seal leaking	Replace the piston seal according to the instructions in Pump P-900 User Manual.
	Blockage or part blockage of flowpath	Flush through to clear blockage. If necessary, replace tubing. Check inlet tubing filter. It can become clogged if unfiltered buffers or samples are applied. See instructions for flushing through at the end of the run in Pump P-900 User Manual.

# 7 Reference information

This chapter contains technical data, regulatory and other information.

# 7.1 Specifications

Parameter	ÄKTAexplorer	ÄKTAmicro	ÄKTApurifier
Ingression protection	IP20	IP20	IP20
Supply Voltage	100-120/220-240 V ~, 50 to 60 Hz	100-120/220-240 V ~, 50 to 60 Hz	100-120/220-240 V ~, 50 to 60 Hz
Power consump- tion	600 VA	370 VA	600 VA
Fuse specification	T 6.3 AL 250 V	T 6.3 AL 250 V	T 6.3 AL 250 V
Dimensions (H × W × D)	450 × 480 × 610 mm	450 × 480 × 610 mm	450 × 490 × 610 mm
Weight	66.8 kg	55 kg	41 kg
Ambient tempera- ture	+4 to +40 °C	+4 to +40 °C	+4 to +40 °C
Relative humidity tolerance (non- condensing)	10 to 95%	10 to 95%	10 to 95%
Atmospheric pressure	84 to 106 kPa (840 to 1060 mbar)	84 to 106 kPa (840 to 1060 mbar)	84 to 106 kPa (840 to 1060 mbar)

## 7.2 Chemical resistance

Chemical	Exposure < 1 day	Exposure up to 2 months	CAS no.	EEC no.	Comments
Acetaldehyde	OK	ОК			
Acetic acid, < 5%	ОК	ОК			

## 7.2 Chemical resistance

Chemical	Exposure < 1 day	Exposure up to 2 months	CAS no.	EEC no.	Comments
Acetic acid, 70%	OK	OK	64-19-7	200-580-7	
Acetonitrile	OK	OK	75-05-8	200-835-2	FFKM, PP and PE swell.
Acetone, 10%	OK	Avoid			PVDF is affected by long term use.
Ammonia, 30%	OK	OK	7664-41-7	231-635-3	Silicone is affected by long-term use.
Ammonium chlo- ride	OK	OK	12125-02-9	235-186-4	
Ammonium bicar- bonate	OK	OK			
Ammonium nitrate	ОК	OK			
Ammonium sul- phate	OK	ОК	7783-20-2	231-984-1	
1-Butanol	ОК	ОК			
2-Butanol	OK	OK			
Citric acid	OK	OK	29340-81-6	249-576-7	
Chloroform	OK	Avoid			Kalrez™, CTFE, PP and PE are affected by long term use.
Cyclohexane	ОК	ОК			
Detergents	ОК	ОК			
Dimethyl sulphox- ide	Avoid	Avoid	67-68-5	200-664-3	PVDF is affected by long term use.
1, 4-Dioxane	Avoid	Avoid			ETFE, PP, PE and PVDF are affected by long term use.
Ethanol, 100%	ОК	ОК	75-08-1	200-837-3	

Chemical	Exposure < 1 day	Exposure up to 2 months	CAS no.	EEC no.	Comments
Ethyl acetate	OK	Avoid			Silicone not resistant. Pressure limit for PEEK decreases.
Ethylene glycol, 100%	OK	OK	107-21-1	203-473-3	
Formic acid, 100%	ОК	ОК	64-18-6	200-579-1	Silicone not resistant.
Glycerol, 100%	ОК	ОК	56-81-5	200-289-5	
Guanidinium hy- drochloride	OK	ОК			
Hexane	ОК	Avoid			Silicone not resistant. Pressure limit for PEEK decreases.
Hydrochloric acid, 0.1 M	ОК	ОК	7647-01-0	231-595-7	Silicone not resistant.
Hydrochloric acid, > 0.1 M	ОК	Avoid			Silicone not resistant. Titanium is affected by long term use.
Isopropanol, 100%	ОК	ОК	67-63-0	200-661-7	
Methanol, 100%	ОК	ОК	74-93-1	200-659-6	
Nitric acid, diluted	OK	Avoid			Silicone not resistant.
Nitric acid, 30%	Avoid	Avoid			Elgiloy is affected by long term use.
Phosphoric acid, 10%	ОК	Avoid	7664-38-2	231-633-2	Titanium, alumini- um oxide and glass are affected by long term use.
Potassium carbon- ate	OK	OK	584-08-7	209-529-3	
Potassium chloride	ОК	ОК	7447-40-7	231-211-8	

Chemical	Exposure < 1 day	Exposure up to 2 months	CAS no.	EEC no.	Comments
Pyridine	Avoid	Avoid			ETFE, PP and PE not resistant.
Sodium acetate	ОК	OK			
Sodium bicarbon- ate	OK	OK			
Sodium bisulphate	ОК	OK			
Sodium borate	ОК	OK			
Sodium carbonate	OK	ОК			
Sodium chloride	ОК	OK	7647-14-5	231-598-3	
Sodium hydroxide, 2 M	OK	Avoid	1310-73-2	215-185-5	PVDF and borosili- cate glass are af- fected by long term use.
Sodium sulphate	ОК	OK	7757-82-6	231-820-9	
Sulphuric acid, dilut- ed	OK	Avoid			PEEK and titanium are affected by long term use.
Sulphuric acid, medium concentra- tion	Avoid	Avoid			
Tetrachloroethy- lene	Avoid	Avoid			Silicone, PP and PE are not resistant.
Tetrahydrofuran	Avoid	Avoid			ETFE, CTFE, PP and PE are not resistant.
Toluene	OK	Avoid			Pressure limit for PEEK decreases.
Trichloroacetic acid, 1%	OK	OK	76-03-9	200-927-2	
Trifluoroacetic acid, 1%	OK	OK	176-05-1	200-929-3	
Urea, 8M	OK	OK	57-13-6	200-315-5	

Chemical	Exposure < 1 day	Exposure up to 2 months	CAS no.	EEC no.	Comments
o-Xylene and p-Xy- lene	OK	Avoid			PP and PE are af- fected by long term use.

# 7.3 System recommendations

Refer to ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro User Manuals, or contact your local GE Healthcare representative for the most current information.

## 7.4 Literature

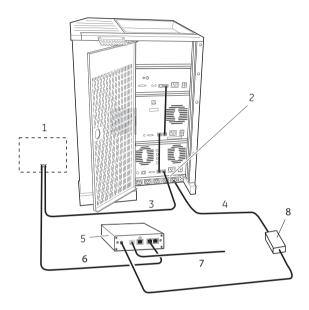
For further information regarding the ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro system, refer to the following:

- ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro User Manuals
- ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro Safety Handbooks
- ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro Installation Guides
- UNICORN User Manual

## 7.5 Ordering information

For ordering information visit www.gelifesciences.com/AKTA.

# Appendix A Electrical and communication connections

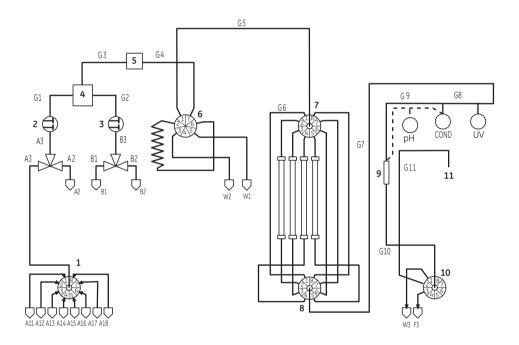


No.	Description	No.	Description
1	Fraction collector (optional)	5	CU-950
2	Mains supply socket	6	UniNet-1-cable
3	UniNet-1-cable	7	USB cable (to computer)
4	Power cord	8	Power converter

Figure A.1: Electrical and communication connections for ÄKTAexplorer, ÄKTApurifier and ÄKTAmicro

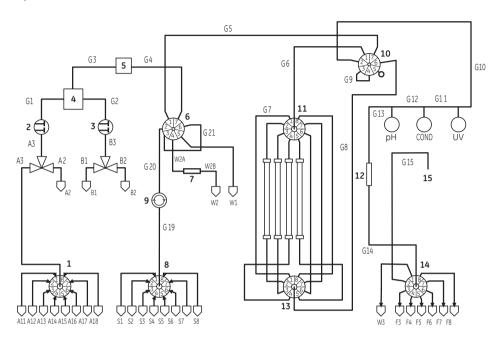
# Appendix B Connection diagram - Liquid flow path

# Liquid flow path for ÄKTAexplorer 10



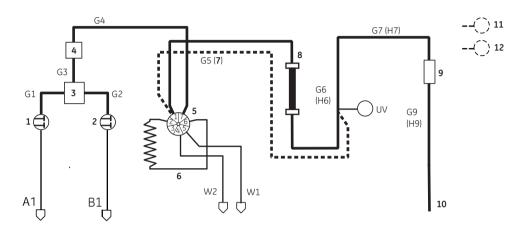
No.	Description	No.	Description
1	Buffer Valve (V6)	7	Column Selection Valve (V2)
2	Pump A	8	Column Selection Valve (V3)
3	Pump B	9	Flow restrictor
4	Mixer	10	Outlet Valve (V4)
5	On-line filter	11	To Fraction Collector (optional)
6	Injection Valve (V1)		

# Liquid flow path for ÄKTAexplorer 100



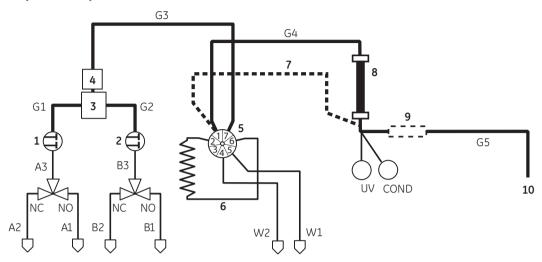
No.	Description	No.	Description
1	Buffer Valve (V6)	9	Sample Pump
2	Pump A	10	Flow Direction Valve (V7)
3	Pump B	11	Column Selection Valve (V2)
4	Mixer	12	Restrictor
5	On-line filter	13	Column Selection Valve (V3)
6	Injection Valve (V1)	14	Outlet Valve (V4)
7	Restrictor	15	To Fraction Collector (optional)
8	Sample Valve (V5)		

# Liquid flow path for ÄKTApurifier 10 and 100



No.	Description	No.	Description
1	Pump A	7	Bypass
2	Pump B	8	Column
3	Mixer	9	Flow restrictor
4	On-line filter	10	To Fraction Collector (optional)
5	Injection Valve (V1)	11	Cond (optional). Connected after the UV cell.
6	Sample loop	12	pH (optional). Connected after the conductivity cell.

## Liquid flow path for ÄKTAmicro



No.	Description	No.	Description
1	Pump A	6	Sample loop
2	Pump B	7	Bypass
3	Mixer	8	Column
4	On-line filter	9	Flow restrictor (supplied)
5	Injection Valve (V1)	10	To Fraction Collector (optional)

# Appendix C Tubing

# Tubing specifications for ÄKTAexplorer

Names in the Label column in *Table C.1* and *Table C.2* refer to tubing labels in the liquid flow path connection diagram, see *Liquid flow path for ÄKTAexplorer 10*, on page 65 and *Liquid flow path for ÄKTApurifier 10 and 100*, on page 67, respectively.

Table C.1: Tubing specifications for ÄKTAexplorer 10

Use	Label	Material	Length (mm)	I.D. (mm)	Volume (µl)
Inlets A11 to A18	A11-18	Teflon™	1250	1.6	$2.5 \times 10^3$
Inlet A1	A1	Teflon	750	1.6	$1.5 \times 10^3$
Inlet A2	A2	Teflon	2000	1.6	$4.0 \times 10^3$
Inlet B1	B1	Teflon	1800	1.6	$3.6 \times 10^3$
Inlet B2	B2	Teflon	1800	1.6	$3.6 \times 10^3$
Switch valve - pump	A3, B3	Teflon	150	1.6	302
Pump - Mixer	G1, G2	PEEK	300	0.50	59
Mixer - On-line filter	G3	PEEK	150	0.50	29
Filter - V1 (Inj. Valve)	G4	PEEK	460	0.50	90
V1 - V2	G5	PEEK	270	0.25	13
V2 - V3 (bypass)	G6	PEEK	620	0.50	122
V3 - UV	G7	PEEK	550	0.25	27
UV - Cond. Cell	G8	PEEK	160	0.25	8
Cond. Cell - Flow restrictor	G9	PEEK	450	0.25	22
Flow restrictor - V4	G10	PEEK	120	0.25	6
V4 - Fraction collector	G11	PEEK	600	0.25	29
V4 - Flow through	F3	PEEK	1000	0.50	196
V1 - Waste	W1, W2	Tefzel™	1300	0.75	574

Use	Label	Material	Length (mm)	I.D. (mm)	Volume (µl)
V4 - Waste	W3	Tefzel	1000	0.75	442

Table C.2: Tubing specifications for ÄKTAexplorer 100

Use	Label	Material	Length (mm)	I.D. (mm)	Volume (µl)
Inlets A11 to A18	A11-18	Teflon	1250	2.9	8.3 × 10 <sup>3</sup>
Inlet A1	A1	Teflon	750	2.9	4.9 × 10 <sup>3</sup>
Inlet A2	A2	Teflon	2000	2.9	$13.2 \times 10^3$
Inlet B1	B1	Teflon	1800	2.9	11.9 × 10 <sup>3</sup>
Inlet B2	B2	Teflon	1800	2.9	11.9 × 10 <sup>3</sup>
Switch valve - pump	A3, B3	Teflon	150	2.9	991
Pump - Mixer	G1, G2	PEEK	300	0.75	133
Mixer - On-line filter	G3	PEEK	150	0.75	66
Filter - V1 (Inj. Valve)	G4	PEEK	460	0.75	203
V1 - V7	G5	PEEK	470	0.75	208
V7 - V2	G6	PEEK	410	0.75	181
Bypass	G7	PEEK	620	0.75	274
V3 - V7	G8	PEEK	470	0.75	208
V7 loop	G9	PEEK	180	0.75	80
V7 - UV	G10	PEEK	370	0.75	163
UV - Cond. Cell	G11	Tefzel	160	0.75	71
Cond. Cell - pH Cell	G12	Tefzel	450	0.75	199
pH Cell - Flow restrictor <sup>1</sup>	G13	Tefzel	110	0.75	49
Flow restrictor - V4 <sup>1</sup>	G14	Tefzel	120	0.75	53
V4 - Fraction collector	G15	Tefzel	600	0.75	265
Flow through	F3	Tefzel	1000	1.0	785
V1 - Waste	W1	Tefzel	1300	0.75	574

Use	Label	Material	Length (mm)	I.D. (mm)	Volume (µl)
Sample pump - Waste	W2	Teflon	1300	1.6	$2.6 \times 10^3$
V4 - Waste	W3	Tefzel	1000	1.0	785

<sup>1</sup> Not mounted at factory

# Tubing specifications for ÄKTApurifier

Names in the Label column in *Table C.3* and *Table C.4* refer to tubing labels in the liquid flow path connection diagram, see *Liquid flow path for ÄKTApurifier 10 and 100, on page 67.* 

Table C.3: Tubing specifications for ÄKTApurifier 10

Use	Label	Material	Length (mm)	I.D. (mm)	Volume (µl)
Inlet A1	A1	Teflon	1700	1.6	$3.4 \times 10^3$
Inlet A2	A2	Teflon	1900	1.6	$3.8 \times 10^{3}$
Inlet B1	B1	Teflon	1500	1.6	$3.0 \times 10^3$
Inlet B2	B2	Teflon	1700	1.6	$3.4 \times 10^3$
Switch valve - pump	A3, B3	Teflon	150	1.6	302
Pump - Mixer	G1, G2	PEEK	330	0.50	65
Mixer - On-line filter	G3	PEEK	200	0.50	39
Filter - V1 (Inj. Valve)	G4	PEEK	180	0.50	35
V1 - UV (bypass)	G5	PEEK	400	0.50	79
UV - Cond. Cell	G6	PEEK	160	0.25	8
Cond. Cell - Flow restrictor	G7	PEEK	80	0.25	4
Flow restrictor - V4	G8	PEEK	140	0.25	7
V4 - Fraction collector	G9	PEEK	600	0.25	29
V4 - Flow through	F3	PEEK	1000	0.50	196
V1/V4 - Waste	W1, W2, W3	Tefzel	100	0.75	442

Table C.4: Tubing specifications for ÄKTApurifier 100

Use	Label	Material	Length (mm)	I.D. (mm)	Volume (µl)
Inlet A1	A1	Teflon	1700	2.9	11.2 × 10 <sup>3</sup>
Inlet A2	A2	Teflon	1900	2.9	12.5 × 10 <sup>3</sup>
Inlet B1	B1	Teflon	1500	2.9	9.9 × 10 <sup>3</sup>
Inlet B2	B2	Teflon	1700	2.9	11.2 × 10 <sup>3</sup>
Switch valve - pump	A3, B3	Teflon	150	2.9	991
Pump - Mixer	G1, G2	PEEK	330	0.75	146
Mixer - On-line filter	G3	PEEK	200	0.75	88
Filter - V1 (Inj. Valve)	G4	PEEK	180	0.75	80
V1 - UV (bypass)	G5	PEEK	400	0.75	177
UV - Cond. Cell	G6	Tefzel	160	0.75	71
Cond. Cell - Flow restrictor	G7	Tefzel	80	0.75	35
Flow restrictor - V4	G8	Tefzel	140	0.75	62
V4 - Fraction collector	G9	Tefzel	600	0.75	265
V4 - Flow through	F3	Tefzel	1000	1.0	785
V1/V4 - Waste	W1, W2, W3	Tefzel	100	0.75	442

# Tubing specifications for ÄKTAmicro

Names in the Tubing i.d. column in *Table* refer to tubing labels in the liquid flow path connection diagram, see *Liquid flow path for ÄKTAmicro*, on page 68.

Tubing i.d.	Tubing o.d.	Material	Color	Max. pressure	Volume of 10 cm	Connection points
0.35 mm (G1, G2)	1.6 mm	Titanium	Grey	> 35 MPa	9.6 µl	From Pump P-905 to Mixer

Tubing i.d.	Tubing o.d.	Material	Color	Max. pressure	Volume of 10 cm	Connection points
Union, m/m	-	PEEK	Black	> 35 MPa	-	Between Mixer and On-line filter. Between UV cell and Conductivity cell
0.15 mm (G3 to G5) <sup>1</sup>	1/16"	PEEK	Violet	> 35 MPa	1.8 µl	From On-line filter to outlet (or to fraction collector/MS, optional). (PEEK tubing i.d. 0.15 mm is installed at delivery)
0.15 mm (Di. 0.15)	375 μm	Fused sili- ca	Brown	> 35 MPa	1.8 µl	From On-line filter to outlet (or to fraction collector/MS, optional).  (Tubing kit 0.15 is supplied with the system)
0.10 mm (Di. 0.10)	200 μm	Fused sili- ca	Brown	> 35 MPa	0.8 μΙ	From On-line filter to outlet (or to fraction collector/MS, optional).  (Tubing kit 0.10 is supplied with the system)
0.75 mm (W1, W2)	1/16"	Tefzel	Clear	7 MPa	44.2 µl	Waste tubing
1.6 mm (A1 to A3, B1 to B3)	1/8"	Teflon	Clear	3.4 MPa	201.1 μΙ	Inlet tubing to Pump P-905

 $<sup>1\,</sup>$   $\,$  0.25 and 0.5 mm i.d. tubing is supplied for non-analytical applications.

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