GE Healthcare

Monitor UV-900

User Manual







Important user information

All users must read this entire manual to fully understand the safe use of Monitor UV-900.

WARNING!



The WARNING! sign highlights instructions that must be followed to avoid personal injury. It is important not to proceed until all stated conditions are met and clearly understood.

CAUTION!

The Caution! sign highlights instructions that must be followed to avoid damage to the product or other equipment. It is important not to proceed until all stated conditions are met and clearly understood.

Note

The Note sign is used to indicate information important for trouble-free and optimal use of the product.

CE Certifying

This product meets the requirements of applicable CEdirectives. A copy of the corresponding Declaration of Conformity is available on request.

The **CE** symbol and corresponding declaration of conformity, is valid for the instrument when it is:

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

Recycling



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.

WARNING!

This is a Class A pruduct. In a domestic environment this product may cause radio interference in which case the user may be required to make adequate measures

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About this manual

This manual comprises two parts; a practical part (sections 1–5) and a reference part (sections A-D).

Sections 1–5 contain the necessary information for operating the instrument.

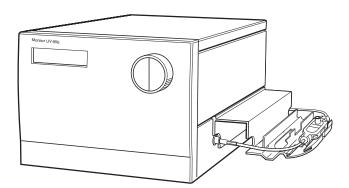
1 Introduction

1.1 General

Monitor UV-900 is a UV-absorption monitor for use in liquid chromatography.

Monitor UV-900 features:

- Variable wavelength for detection in the range 190–700 nm.
- Up to 3 wavelengths can be monitored simultaneously.
- Two alternative flow cells with path length 2 mm and 10 mm.
- Accurate and reliable monitoring through self-test, self-calibration and fiber optics.



1.2 Safety

IMPORTANT! Monitor UV-900 is intended for laboratory use only, not for clinical or in vitro use, or for diagnostic purposes.

- The instrument is designed for indoor use only.
- Do not use in a dusty atmosphere or close to spraying water.



WARNING! When using hazardous chemicals, all suitable protective measures, such as protective glasses, must be taken.



WARNING! The instruments must not be opened by the user. They contain high voltage circuits which can give a lethal electric shock.



WARNING! The system must be connected to a grounded mains socket.



WARNING! Always disconnect the power supply before attempting to replace any item on the instrument.



WARNING! The instrument uses high intensity ultra-violet light. Do not disconnect the optical fibers while the monitor is running.

CAUTION! The flow cell must not be used at pressures above 2 MPa (20 bar, 290 psi). At higher pressures the flow cell may break.



WARNING! 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.



WARNING! There must always be a sample loop connected to ports 2 and 6 of the injection valve. This is to prevent liquid spraying out of the ports when switching the valve. This is especially dangerous if hazardous chemicals are used.



WARNING! This is a Class A product. In domestic environment this product may cause radio interference in which the user may be required to take adequate measures.

2 Installation

2.1 Unpacking

Unpack the instrument and check the items against the supplied packing list. Inspect the items for obvious damage which may have occurred during transportation.

- The system should be installed on a stable laboratory bench providing a suitable working area.
- To maintain correct ventilation, the system requires an appropriate amount of free space. Do not block the ventilation inlets or outlets on the system!

It is recommended that all packing materials should be retained if onward transport of the instrument is expected.

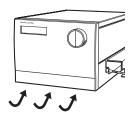
CAUTION! The following information should be read carefully to ensure that the instrument is installed correctly.

2.2 General precautions

The instrument should not be installed in a corrosive atmosphere or in an atmosphere where deposits are liable to form on the optical surfaces.

The instrument should be located in a place of low temperature variations, away from heat sources, draughts and direct sunlight.

The instrument may be operated at normal ambient temperatures in the range +4 to +40 °C.



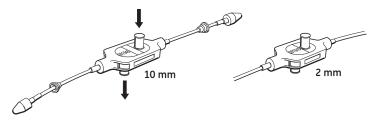
The instrument should be installed on a stable laboratory bench or in ÄKTA™ explorer or ÄKTApurifier. To ensure correct ventilation a free space of 0.1 m is required behind and in front of the instrument. Place the instrument directly on the bench. Do not use any soft material under the instrument, to ensure that the ventilation inlet in the front is not blocked.

2.3 Installing the flow cell

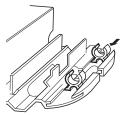
2.3.1 Fixing the flow cell

Handle the optical fibers with care, do not bend excessively.

Two flow cells are available: UV-900/2 with 2 mm light path length (cell volume 2 μ l) and UV-900/10 with 10 mm path length (8 μ l).



- 1 Unpack the flow cell. Do not remove the red protective caps from the inlet or outlet, or the black protective caps on the optical fiber connectors.
- 2 Slide the rear white clip on the cell holder to its inner position for a 2 mm cell and to its outer position for the 10 mm cell.



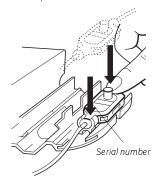
3 Place the flow cell in the opening between the white clips. The cell should be positioned with the text and serial number facing upwards and to the front. Press the cell into the clips to fasten it.

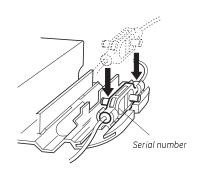
CAUTION! To avoid damaging the optical fibers, press only on the cell body, never on the optical fibers.

To improve the access to the tubing connections, the flow cell can alternatively be positioned with the tubing connections.









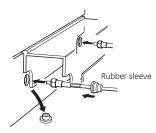
2.3.2 Connecting the optical fibers

1 Remove the 2 black protective caps from the optical fiber connectors.

CAUTION! Do not touch the tips of the optical fibers with your fingers as this will result in poor monitor performance. If you accidentally touch the optical fiber tip, it can be wiped with 30% isopropanol using lens paper.

- 2 Remove the 2 rubber protective caps from the optical fiber receptacles on the right side of the housing.
- 3 Connect the 2 optical fibers to the housing by carefully inserting them into the sockets and tightening the nuts fingertight using the fiber detachment tool supplied.

CAUTION! Do not twist the optical fibers during tightening.



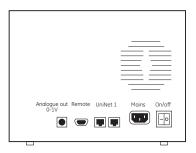
Slide the rubber sleeves on the 2 optical fibers onto the connectors. Make sure that the sleeves are pushed tight to the housing to prevent dust or fluid entering the connection.





2.4 Connecting electrical signal cables

The sockets for electrical signals are located on the back panel.



2.4.1 Connecting to recorder (if used)

1 Connect the recorder to the miniDIN-socket **Analogue out** using the cable supplied. Each wavelength signal is available on a separate channel using the following wires:

λ1	Wire 1	+
	Wire 2	-
λ2	Wire 3	+
	Wire 4	-
λ3	Wire 5	+
	Wire 6	_

Note: The signal cable is delivered with protective covers on each wire. Do not remove the protective covers from unused connections as a short circuit may disturb the measurements.

2 Set the recorder to 0–1 V input, full scale, 0 V offset.

2.5 Connecting to communication link

The monitor can be used in ÄKTAexplorer or ÄKTApurifier and controlled from a PC running UNICORN™ version 3.21 or higher, using UniNet cables.

CAUTION! The mains power to the ÄKTAexplorer or ÄKTApurifier must be switched OFF before connecting the instrument to the UniNet-1 link.



WARNING! Only use mains cables delivered or approved by GE Healthcare.



WARNING! Do not block the rear panel of the system. The mains power switch must always be easy to access.

Connect two UniNet cables to the UniNet-1 connectors. The instrument can be connected in series anywhere in the chain between the PC and the termination plug. The UniNet-1 link connects, in series, the PC with Pump P-905, Monitor pH/C-900 and Monitor UV-900. The termination plug is connected to the last instrument in the chain.

2.6 Connecting to supply voltage

- 1 Make sure the on/off switch is in the OFF position (**O**).
- 2 Connect a mains cable between the instrument and a grounded mains socket. The instrument is delivered with both European and US type mains cables, as standard. Any voltage 100–240 V AC, 50–60 Hz, can be used.

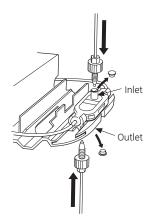


WARNING! The instrument must be connected to a grounded mains socket.

The instrument contains no user replaceable fuse.

2.7 Connecting the tubing

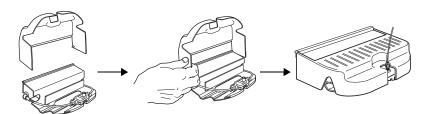
1 Remove the red protective caps from the inlet and outlet of the flow cell and connect the tubing with 1/16" "fingertights".



The inlet is on the top of the flow cell, the same side as the serial number.

2.8 Mounting the cell holder cover

The cover is a simple push fit onto the cell holder. Two small lugs on the cover locate in holes at the front and rear of the cell holder. The cover is then lowered over the cell holder.



3 Operation

3.1 On/off

Selftest Please wait...

1 Switch on the instrument at the mains switch on the rear panel.

Calibrating Please wait...

2 The instrument performs a self-test and then starts calibration.

Please Check UV Cell! Press OK/Esc to cont.

3 If the instrument has a calibrated cell path length set that differs from the nominal length, a warning message appears on the monitor display.

Another message, displaying the calibrated cell length, also appears. The display toggles between these messages.

Real UV Cell Length (?,??mm, SN??????

4 To go to the *Main operating menu*, press **OK** or wait for about 45 seconds to return automatically.

λ1(215) 1.123 AU λ2(254) 0.02345 AU 5 If no such warning messages appear, the display automatically shows the *Main operating menu* after approximately 1 minute. The instrument is ready to use.

All parameters are factory set to default values.

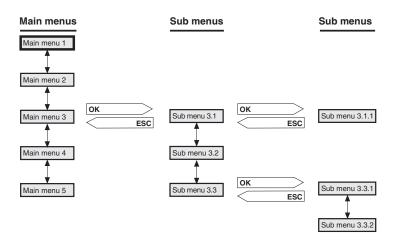
3.2 Menu selection and settings

3.2.1 Menu selection

A specific menu is selected by turning the front selection dial clockwise or counter clockwise. When the required menu is visible the menu or selection is accepted by pressing the **OK** button.



If a menu has sub levels, the sub menu is displayed by pressing the **OK** button. Pressing the **ESC** button moves back one menu level.



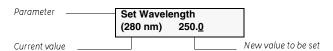
3.2.2 Return to main menu

Pressing **ESC** repeatedly, always returns to the **main menu 2** which is the main operating menu. Press **ESC** once more to return to **main menu 1**, the mode changing menu.



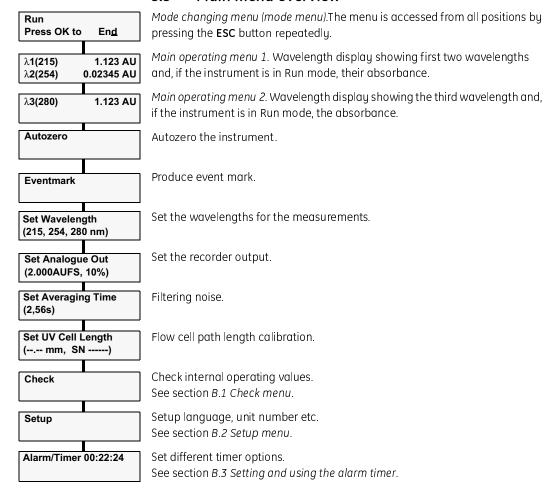
3.2.3 Select value

A cursor below a text or numerical value shows what is affected by the dial. To increase the value turn the dial clockwise. To decrease the value turn the dial counter clockwise.



When setting numerical values, the cursor moves up to the next digit if the dial is turned quickly in one direction, to simplify entering large values. The cursor moves back one place to the right every two seconds if the dial is not turned. The text or numerical value displayed is accepted by pressing the **OK** button. To cancel, press the **ESC** button.

3.3 Main menu overview



3.4 Setting lamp on/off

The lamp should be switched off when no measurement is made to conserve the lamp operating time. The lamp is switched on when a run is started. No warm-up time is required.

- Run
 Press OK to End
- 1 Select the mode changing menu.
- 2 Switch the lamp on (**Run**) or off (**End**) by pressing **OK**. Its current status is shown in the upper left of the display.

When controlled from UNICORN, the lamp is switched on automatically when the flow is started and off when the run is ended

3.5 Reading absorbance values

λ1(215)	1.123 AU
λ2(254)	0.02345 AU
λ3(280)	1.123 AU

The main operating menu shows the absorbance values for up to 3 active wavelengths. The menu is reached from any other menu by pressing the **ESC** button repeatedly. The display for the third wavelength is reached by turning the dial clockwise.

or

215nm	254nm	280nm
1.123	0.023	0.123

As an alternative all 3 wavelengths can be shown in a single display, but limited to 3 decimals. This alternative is reached by turning the dial clockwise.

3.6 Setting wavelength

The instrument can measure up to 3 wavelengths simultaneously and wavelength changes can be set at any time.

Set Wavelength (215, 254, 280 nm) 1 Select main menu **Set Wavelength**, press **OK**.

Set Wavelength λ1 (215, 254, 280 nm)

2 Select sub menu **Set Wavelength** $\lambda 1$, press **OK**.

Set Wavelength λ1 (215nm) 23<u>4</u>

3 Set the value, press **OK**.

Set Wavelength $\lambda 2$ (254nm)

4 The next wavelength $\lambda 2$ is now shown. Set the value or turn the wavelength off, by turning the dial counter clockwise until the value passes through 190 nm. If only one wavelength is to be used, $\lambda 2$ and $\lambda 3$ should be set to off. Press **OK**

The first wavelength $\lambda 1$ can never be set to off.

- 5 Repeat step 4 in the menu **Set Wavelength** λ **3**.
- 6 Press **ESC** to return to the main operating menu.

In UNICORN, the wavelengths are set with the instruction **Wavelength** in **System Control:Manual:Alarm&Mon**.

3.7 Autozero

The autozero function sets the detected absorbance to zero when the **OK** button is pressed. All three wavelengths are autozeroed. Autozero is recommended after wavelength changes in a method and before the sample is injected.

Autozero

1 Select main menu Autozero, press OK. The normal absorbance value display is then shown.

In UNICORN, Autozero is set with the instruction **AutoZeroUV** in **System Control:Manual:Alarm&Mon**.

3.8 Storage and shut-down

Overnight: The flow cell can be left filled with buffer.

Weekend and long term storage: Flush the flow cell with distilled water and then fill it with 20% ethanol.

The flow cell can also be stored dry by flushing as above with distilled water and then blowing a compressed inert gas such as nitrogen (N_2) through the cell. Replace the red protective caps. Never use compressed air as this may contain droplets of oil.

CAUTION! Do not allow solutions which contain dissolved salts, proteins or other solid solutions to dry out in the flow cell. Do not allow particles to enter the flow cell as damage to the flow cell may occur.

3.9 Using an external chart recorder

3.9.1 Setting range and zero

The external chart recorder output from the instrument is always 0–1 V, but the absorbance value for full scale deflection and the Zero Absorbance level on the recorder can be set

Set Analogue Out (2.000AUFS, 10%)

1 Select main menu **Set Analogue Out**, press **OK**.

Set Range (2.000AUFS) <0.1> 2 Set the range value in the sub menu Set Range and press OK. The range is the full scale absorbance range for the chart recorder (1 V). Only fixed steps between 5.0 AU and 0.01 AU can be set and the value is the same for all 3 wavelengths.

 Set Zero Level

 (10% of FS)
 20

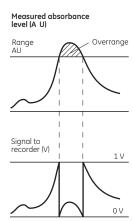
- 3 Select menu **Set Zero Level**, press **OK**.
- 4 Set the value and press **OK**. This value determines where the zero absorbance level should be in relation to full scale on the recorder.

The instrument has an automatic overrange function. If during a peak the monitor signal reaches the full scale value, the signal will drop instantly to 0 V and give an accurate display of the peak starting from this position.

3.9.2 Event mark

Event marks can be set, for example when the sample is injected, and displayed as spikes on all three channels of the chart recorder. The spikes are 10% of the full scale of the chart recorder which corresponds to 0.1 V.

1 Select main menu **Event mark**, press **OK**.



3.10 Filtering noise

To filter the noise in the UV-signal, a moving average filter is used. The averaging time is the time interval used for calculating the moving average of the absorbance signal. A long averaging time will smooth out noise efficiently, but it will also distort the peaks. Peaks narrower than the minimum peak width value may be distorted. Because of this the averaging time should be as short as possible, see the table below. On delivery the averaging time is set to 2.56 s.

1 Select menu **Set Averaging Time**, press **OK**.

Set Averaging	Time
(2.56s)	<2. <u>5</u> 6>

2 Set the value, press **OK**. Use the fixed **values** between 5.12 and 0.08 s. When monitoring more than one wavelength, the recommended averaging time is a minimum of 1.28 s.

Averaging time (s)	Time constant (s) (approximate)	Min. peak width at half height (s)
5.12	2.0	32
2.56	1.0	16
1.28	0.5	8.0
0.64	0.2	3.2
0.32	0.1	1.6
0.16	0.05	0.8
0.08	0.03	0.5

In UNICORN the averaging time is set with the instruction **AveragingTime** in **System Control:Manual:Alarm&Mon**.

3.11 UV cell calibration

The path length in the UV flow cell might differ from the nominal length (1, 2, 5 respective 10 mm) which interferes the calculation of the protein concentration in the eluate. To achieve normalized absorbance, the path length in the UV flow cell must be calibrated

It is recommended to calibrate a new UV flow cell before use. An old flow cell can be calibrated whenever you think that a calibration is required.

Equipment required

To perform the calibration you need test solutions, syringes and accessories for desired cell length:

- UV-900 1 mm Calibr. kit, no. 18-6324-01
- UV-900 2 mm Calibr. kit. no. 18-6324-02
- UV-900 5 mm Calibr. kit. no. 18-6324-04
- UV-900 10 mm Calibr. kit, no. 18-6324-05

To calculate the real cell length use the supplied Excel-file:

UV-900 Cell Calibr. Excel-file, no. 18-6324-06

Removing an old calibration setting

If the instrument already has a calibrated cell length set that differs from the nominal length, a warning message appears on the UV monitor display at start-up. Another message, displaying the calibrated cell length, also appears. The display toggles between these messages. The display automatically returns to the main operating menu after about 45 seconds, or if pressing **OK**.

Please Check UV Cell! Press OK/Esc to cont.

Real UV Cell Length (?,??mm, SN??????

1 To remove an old calibration value, press Esc while the display toggles between the two messages. The Set UV Cell Length menu appears. Press OK.

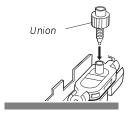
Set UV Cell Length (--,--mm, SN-----)

2 At the **Set Calibr. Length** menu, set the cell length to **0**. Press **OK**.

Set Calibr. Length (--,--mm) --.-<u>-</u>

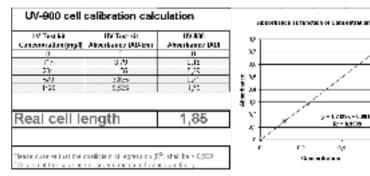
Preparing the calibration

- 1 Remove any old calibration value by setting the cell length to 0 (see above).
- 2 Unpack the UV Test kit.
- 3 Make sure that the Flow restrictor is connected in the flow path after the UV flow cell.
- Mount the Union Luer female/ 1/16" male, included in the test kit, in the upper inlet of the UV flow cell.



y = Logs < - 0.8

Open the UV-900 Cell Calibr. Excel-file on the computer.



The solution bottles are marked with the concentration value and the reference absorbance value for each solution.

Enter the concentration of the solutions in increasing order in the column UV Test kit Concentration (mg/l).

Enter the absorbance value in increasing order in the column

UV Test kit Absorbance (AU/cm).

Performing the absorbance measurements

For each of the four test solutions, the absorbance will be measured and automatically compared to its reference value.



WARNING! 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.



WARNING! Sulfuric acid included in the test soluitons is injurious to health. Avoid spillage and use eye protection.

- Switch on the UV lamp, see 3.4 Setting lamp on/off. 1
- 2 Select a main menu on the monitor display that shows wavelength 254 nm
- Fill one of the supplied syringes with 1.5–2 ml of the 0 AU/cm solution. Make sure that there is no air in the syringe!
- Fit the syringe in the Union Luer connector and inject the solution. Do NOT remove the syringe.
- Wait until the monitored absorbance value is stable. 5
- 6 Perform an **Autozero** operation. See section 3.7 Autozero.

- 7 Remove the syringe.
- 8 Inject the other four test solutions in increasing concentration order. Use a new syringe for each solution.

Note: Air trapped in the UV cell causes inaccurate measurements. To avoid introducing air into the cell, gently fill the Luer union up to the edge using test solution from the syringe that is next. Then insert the syringe into the Luer union.

- 9 After each injection, wait for a stable absorbance value and enter the measured values in the column UV -900 Absorbance (AU) in the UV-900 Cell Calibr. Excel-file.
- 10 When all absorbance values have been entered, the real cell length is shown next to the **Real cell length** square.

Note: The regressions coefficient should be larger than 0.999. If not, one or several measured values are faulty.

Entering the new real length

1 Select menu **Set UV Cell Length** in the UV monitor. Press **OK.** The **Set Calibr. Length** menu appears. Press **OK.**

2 Enter the **Real cell length** value (from the Excel-file) and press **OK**.

The **Set UV Cell Type** menu appears. Check the cell type value.

If the value is correct, turn the selection dial clockwise to go to next menu. If the value has to be changed, press **OK** to change it.

4 At the Set Cell Serial No. menu press OK.

5 Enter the serial number of the cell (see the label on the flow cell). Press **OK**.

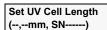
3.12 Changing flow cell

The flow cell can be changed when required, for example from 2 mm to 10 mm when the sensitivity of the measurement must change due to a small amount of sample being applied, or from a 10 mm to 2 mm when a lower sensitivity is desired, due to output signal limitation.

See section 2.3 Installing the flow cell.

3.13 Restart after power failure

If the power supply to the instrument is interrupted, the instrument automatically restarts itself and displays the main operating window. All set values are retained in the instrument but the instrument starts with the lamp switched-off





Set UV Cell Type (2 mm)

Set Cell Serial No. (SN -----) ---

4 Maintenance



WARNING! Always disconnect the power supply before attempting to replace any item on the system during maintenance.



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

4.1 Periodic maintenance

Interval	Action (see procedures below)
Every 3 month	Check the instrument
Every 6 month or more often if required	Clean the flow cell and optical fiber connectors



WARNING! Remove liquid or dirt from the system surface using a cloth and, if necessary, a mild cleaning agent.



WARNING! 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.



WARNING! When using hazardous chemicals, make sure that the entire system has been flushed thoroughly with bacteriostatic solution, e.g. NaOH, and distilled water before service and maintenance.

4.2 Cleaning-in-place

Pump a cleaning or sanitizing agent through the flow cell. The standard recommendation is to pump 1 M NaOH at 1 ml/min for 30 minutes and then wash out with buffer.



WARNING! NaOH is injurious to health. Avoid spillage.

4.3 Checking the instrument

4.3.1 Lamp intensity

Check Lamp Intensity 210nm 85% 300nm 95%

- 1 Select menu Check, press OK.
- 2 Select menu Check Lamp Intensity.

If the lamp intensity is < 20%, contact GE Healthcare for lamp replacement or change of internal optical fiber.

4.3.2 Lamp on-time

Check Lamp Run Time 200h

- 1 Select menu **Check**, press **OK**.
- 2 Select menu Check Lamp Run Time.

If the lamp ontime is > 4000 hours, contact GE Healthcare for lamp replacement.

4.3.3 Flip time

The flip time is the time the monochromator stepper motor has been in operation.

1 Select menu Check, press OK.

Check Flip Time 50h

2 Select menu Check Flip Time.

If the flip time is > 2000 hours, contact GE Healthcare for maintenance.

4.3.4 UV flow cell

This menu shows the set path length and the serial number of the UV flow cell.

1 Select menu **Check** and press **OK**.

UV Flow Cell (--,--mm, SN -----)

2 Select menu UV Flow Cell.

4.4 Cleaning the flow cell and optical fiber connectors

A clean flow cell and optical connectors are essential for ensuring the correct operation of the UV-monitor.

CAUTION! Do not allow solutions containing dissolved salts, proteins or other solid solutes to dry out in the cell. Do not allow particles to enter the flow cell as damage to the flow cell may occur.

4.4.1 Cleaning the flow cell

- 1 Connect a syringe to the inlet of the flow cell and squirt distilled water through the cell in small amounts. Then fill the syringe with a 10% surface active detergent solution like *Decon 90*, *Deconex 11*, *RBS 25* or equivalent, and squirt five times.
- 2 After five squirts, leave the detergent solution in the flow cell for at least 20 minutes.
- 3 Pump the remaining detergent solution through the flow cell.
- 4 Rinse the syringe and flush the cell with distilled water (10 ml).

4.4.2 Cleaning the optical fiber connectors

Wipe with 30% isopropanol on lens paper.

4.4.3 Instrument housing



WARNING! When using hazardous chemicals, make sure that the entire system has been flushed thoroughly with bacteriostatic solution, e.g. NaOH, and distilled water before service and maintenance.

4.5 Cleaning the system

- Wipe the surface regularly with a damp cloth. Do not allow spilt liquid to dry on the instrument.
- Remove dirt from the surface using a cloth and a mild cleaning agent.
- Let the system dry completely before using it.

Wipe the instrument housing regularly with a damp cloth. Let the instrument dry completely before use.

4.6 Recycling

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

5 Trouble-shooting

5.1 General

Monitor UV-900 V1.06 When contacting GE Healthcare for support, state the program version of the instrument, which is shown for 2 seconds after the self-test, during switch-on, or in the menu **Check Service Mode**, see *page 35*.



WARNING! The instruments must not be opened by the user. They contain high voltage circuits which can give a lethal electric shock.

5.2 Faults and actions

If the suggested actions do not correct the fault, call GE Healthcare.

Fault	Possible cause	Corrective action
No text on the front display	No power to the monitor	Check that the mains cable is connected and the power switch is in ON-position I
Noisy UV-signal, signal drift	The buffer may be impure	Check if the signal is still noisy with water
or instability	There may be air in the flow cell	Check that the flow restrictor gives a back pressure of 0.2 MPa in ÄKTAexplorer, and 3-5 MPa in ÄKTApurifier
		If there is a lot of air in the water, degas the buffer continuously (we recommend helium sparging)
		Check the connections of the UV cell optical fibers
		Clean the UV cell, see section 4.4
Ghost peaks	Air in the eluents	Degas if necessary (we recommend helium sparging)
	Dirt or residues in the flow path from previous runs	Clean the system in accordance with ÄKTAexplorer or ÄKTApurifier System Manual
	Residues in the column from previous runs	Clean the column in accordance with the column instructions
	Bad mixing of the eluents	Check that the mixer is functioning correctly and that the correct chamber volume is being used
Error in external chart recorder	The recorder not properly set up	Check the chart recorder in accordance with its manual
	Monitor UV-900 not properly set up	Test the recorder function by selecting recorder test according to section <i>Check Analogue Out</i> on <i>page 35</i>

5.3 Error messages

If the suggested actions do not correct the fault, call GE Healthcare.

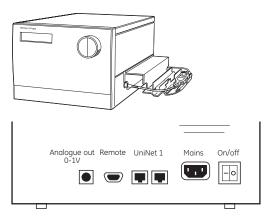
Message	Action
Sample fiber failure	There may be too high absorption in the UV cell.
	1. Check the liquid.
	2. Ensure that there are no air bubbles in the system.
	3. Check the connections of the UV cell optical fibers.
	 Clean the UV cell, see section Cleaning the flow cell and optical fiber connectors.
Not calibrated Calibration failed	Recalibrate by switching the instrument off and then back to on, or choose Recalibrate in the Check menu
Change lamp	Call GE Healthcare for Xe-lamp exchange
Mode fiber failure Reference fiber failure Lamp module failure Low light intensity Block filter is defect	Call GE Healthcare
High intensity in Reference fiber	Recalibrate by switching the instrument off and on
High intensity	1. Disconnect the optical fiber connectors.
in Sample fiber	2. Recalibrate by switching the instrument off and on.
ERROR Number 10	1. Switch off the instrument.
ERROR Number 16	2. Check all connections.
ERROR key(OK) ERROR key(Esc) ERROR key(OK+Esc) ERROR 100 ERROR 109-113 ERROR 120-121	3. Switch on the instrument.
ERROR 106-108	1. Switch off the instrument.
ERROR 118	2. Check all UniNet connections.
	3. Switch on the instrument.

Reference information

A Description

A.1 Instrument

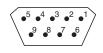
The Monitor UV-900 is a UV-absorption monitor with variable wavelength. The instrument contains no internal user replaceable items.



Connector/switch	Function
Analogue out 0-1 V	Recorder output, 3 channels 0-1 V
Remote	Digital signal inputs, lamp on/off, autozero, event mark
UniNet 1	Computer network
Mains	Supply voltage, grounded
On/off	Instrument on/off switch

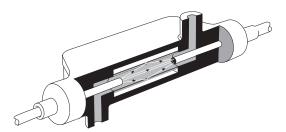
Connect any auxiliary equipment to the 9-pole D–Sub female REMOTE connector (5 V TTL signals only).

Pin	Signal	Function		
		Active status = low or closed terminal to pin 5 (0V)		
1	Remote on/off	Active = lamp on, inactive = lamp off		
2	Autozero	From inactive to active > 100 ms = Autozero		
3	Event mark	From inactive to active > 100 ms = Event mark		
4	-			
5	0 V			
6-9	_			



A.2 Flow cell

The optical path length of the flow cell is 2 mm and 10 mm. The flow cell is made of quartz with a titanium housing.



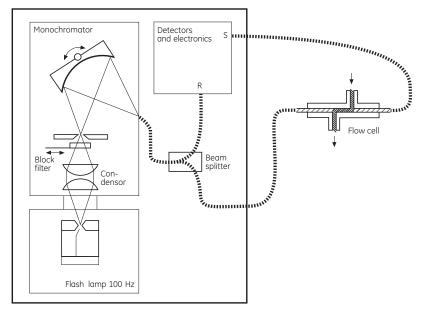
The unique design of the flow cell prevents the formation of distinct interfaces between eluent components with different refractive indices and eliminates the negative influence these would cause. The precision of monitoring is enhanced by the construction of the flow cell, which ensures total reflectance of light. This maintains a high intensity of light to the detector. The long path length combined with a small cell volume increases sensitivity.

A.3 Monitor principle

A Xenon flash lamp gives a high intensity, continuous spectrum throughout the range 160–2000 nm. The light enters a monochromator which includes a condensing system, blocking filter, entrance slit and a concave aberration-corrected holographic grating. Monochromatic light from the grating is directed to an optical fibre. The grating is turned by a stepping motor for wavelength selection between 190–700 nm, in steps of 1 nm. Up to 3 wavelengths can be monitored simultaneously.

For wavelengths between 360–700 nm, a blocking filter is moved into the light path to filter out unwanted light, of a half wavelength from the second order spectrum, before entering the monochromator.

The light from the monochromator to the flow cell and from the cell to the detector electronics is guided by optical fibres which focus its full intensity on the liquid flow path, maximising the sensitivity of the monitoring. Before entering the flow cell, the monochromatic light is split in a beam splitter, with 50% of the light passing through the sample fibre (S) and the flow cell, and 50% being directed through the reference fibre (R). Two photodiodes with identical characteristics monitor the intensities of the measuring and reference beams.



The long path length and small volume of the flow cell ensure very high sensitivities and high signal-to-noise ratios. The detection system is also very stable and because the optical unit is located away from the lamp and electronics, noise and drift caused by temperature variations is avoided.

At calibration the instrument automatically finds 2 persistent lines in the spectrum of Xenon. The wavelengths of these known lines are used to calibrate the stepper motor that turns the grating.

B Menus

B.1 Check menu

Checking autozero level

The instrument internal absorbance value for autozero can be checked to test the consistency in buffers.

Check Autozero AZ1 (215) 0.23456 AU Select main menu **Check**, press **OK**. The autozero absorbance value for wavelength **1** is shown.

AZ2 (254) 0.23775 AU AZ3 (280) 0.12326 AU 2 Turn the selection dial clockwise to display wavelengths **2** and **3**.

Lamp intensity, Lamp Ontime, Fliptime, UV Flow cell

See Chapter 4 Maintenance.

Checking the wavelength

If there is any doubt that the instrument is showing the correct values, the wavelength calibration can be checked.

1 Select main menu **Check**, press **OK**.

Checking Wavelength Please wait...

Wavelengths OK 230+0 459+0 542+0 OK

or

Checking Wavelength Please wait...

Wavelengths not OK 230-4 459+3 542+4 OK

Wavelengths not OK Recalibrate? OK

2 Select sub menu Check Wavelength, press OK. The check starts and after approx. 20 seconds the message Wavelength OK or Wavelength not OK is shown together with the deviation values. If not OK, recalibration can be selected.

If a deviation in wavelength occurs repeatedly when checked, contact Amersham Biosciences.

Recalibrate

If the instrument is left switched on for a long period (> 10 days), it may be necessary to recalibrate it. This calibration is identical to that done when the instrument is switched on.

1 Select main menu Check, press OK.

Recalibrating Please wait...

Recalibration finished OK

Select sub menu Recalibrate, press OK. The recalibration starts and after approx. 60 seconds the message Recalibration finished is shown.

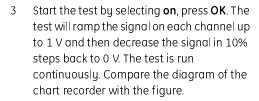
Check Analogue Out

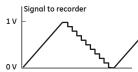
The function of the connected chart recorder can be tested.

1 Select main menu **Check**, press **OK**.

Check Analogue Out (off) on

2 Select sub menu **Check Analogue Out**, press **OK**.





4 Stop the test by pressing **OK** or **ESC**.

Check Service Mode

Service information relevant to the instrument can be checked. Information may not be available in all menus.

1 Select main menu **Check**, turn the selection dial clockwise for next display.

Check Service Mode

2 Select sub menu Check Service Mode by pressing OK.

Telephone Service 012345678901

3 The service telephone number is displayed. Turn the selection dial clockwise to view the next display.

Contract Number 012345678901

4 The service contract number is displayed. Turn the selection dial clockwise to view the next display.

Serial Number 01234567 YM 012345

5 The instrument serial number is displayed. Turn the selection dial clockwise to view the next display.

Monitor UV-900 V1.06

6 The instrument name and software version are displayed. Turn the selection dial clockwise to view the next display.

Date of Maintenance ?

7 The date of the last service is displayed. Turn the selection dial clockwise to view the next display.

Buzzer Test.

8 Press **OK** to test the instrument buzzer.

Press Esc to return to the Check Service Mode menu.

B.2 Setup menu

Setup language

Sets the language used in the display.

Setup Language (GB) <u>GB</u> D F E I

- 1 Select main menu **Setup**, press **OK**.
- 2 Select sub menu **Setup Language**, press **OK**.
- 3 Select the desired language.

GB = British English

D = German

 $\mathbf{F} = \text{French}$

 $\mathbf{E} = Spanish$

I = Italian

Setup unit number

The unit number is the identification the UV monitor has on the UniNet-bus. It should correspond to the number set in UNICORN for the UV monitor. The number should be set to **0** if one UV monitor is used. If more than one UV monitor is used, they must all have different identification numbers.

1 Select main menu **Setup**, press **OK**.

Setup Unit Number (0)

- 2 Select sub menu **Setup Unit Number,** press **OK**.
- 3 Select unit number (0-25).

Setup display angle

The display angle can be set to compensate for different viewing heights.

1 Select main menu **Setup**, press **OK**.

Setup Display Angle (->I) ->\ ->I ->/

- 2 Select sub menu **Set Display Angle**, press **OK**.
- 3 Select viewing angle (->\ Up, -> I Mid or -> I Down).

B.3 Setting and using the alarm timer

You can set the alarm function to either a fixed alarm time or using a count-down timer. The UV monitor can be started or stopped automatically, or an alarm can sound, at the set time. It is not possible to set both an alarm time and a count-down timer. Current values are shown in parentheses.

Alarm/Timer 12:30:52

1 Select main menu **Alarm/Timer**, press **OK**.

Alarm/ Timer Action (Bzz) Buzzer Run End

2 Set the action to take place. Press OK to select action. Buzzer will generate an audible alarm for 15 s and a message. Run will start the pump at the set flow rate, End will stop the pump, each generating one beep and a message.

Set Alarm 12:32:22 (0) 0<u>0</u>.00.00

3 Use the sub menu Set Alarm if you want to set an alarm at a fixed time. Press OK to enter the time value in the form HH.MM.SS, pressing the OK button after entering each time unit.

Set Timer (0) 0<u>0</u>.00.00

4 If you want to set a count-down timer, turn the dial to select sub menu **Set Timer**. Press **OK** to enter the countdown value in the form **HH.MM.SS**,
pressing the **OK** button after entering each time unit.

Alarm/Timer 12:30:52 (Bzz 12:33:00)

5 Press **ESC** button to return to the **Alarm/Timer** menu which now shows the set alarm time or count-down time as **BzzHH:MM:SS**.

Bzz12:41:29 12:41:49 !! Alarm time!!

6 When the alarm time is due or the count-down timer reaches 00:00:00, an alert display is shown and the instrument beeps, until the **OK** button is pressed.

Set Clock 0<u>0</u>:32:22 (0<u>0</u>:36:53)

The alarm timer is based on the internal instrument clock which can be set in the **Set Clock** menu placed after the **Alarm/Timer** menu. The clock will be reset when power is turned **OFF**.

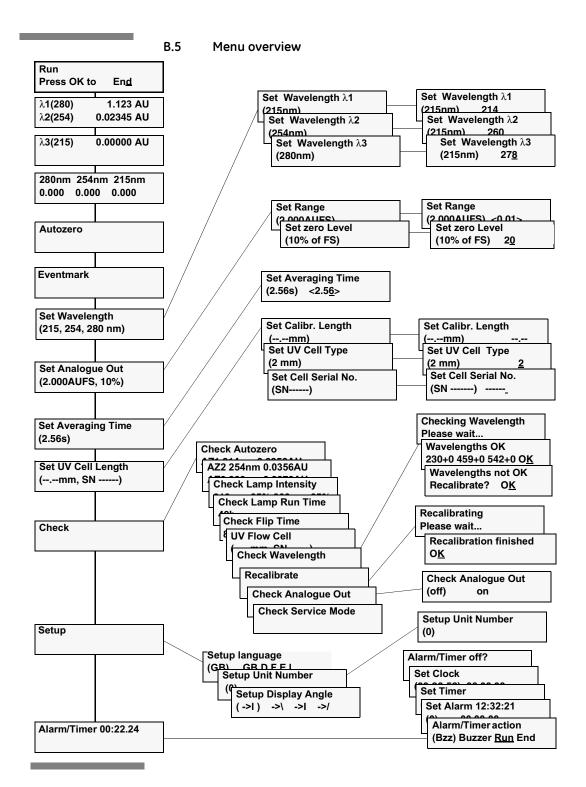
Alarm/Timer off? (Bzz 05:33:00)

An already set alarm/timer function can be reset by pressing **OK** in the menu **Alarm/Timer off?**

B.4 Service displays

Enter Access Code!

The instrument has service displays for use by authorised service personnel. If the service display **Enter Access Code:** is accidentally selected, press the **ESC** button to exit to the normal operation display.



C Technical specifications

Operating data

Wavelength range 190-700 nm in steps of 1 nm, 3 wavelengths simultaneously

Bandwidth 4 nm
Wavelength accuracy ±2 nm
Wavelength reproducibility ±0.01 nm

Wavelength switch time < 500 ms (one cycle from 214 nm to

254 nm and back to 214 nm)

Linearity < 2% deviation up to 2 AU at 260 nm with

Uracil at pH 2

Noise (specified for 10 mm

flow cell)1.2

Single wavelength

short term (0.5–1 min) < 6x10⁻⁵ AU at 230 nm long term (1–10 min) < 6x10⁻⁵ AU at 230 nm

Dual wavelengths

short term (0.5–1 min) < 2×10⁻⁴ AU at 230 and 254 nm long term (1–10 min) < 2×10⁻⁴ AU at 230 and 254 nm Orift² < 2×10⁻⁴ AU/h at 254 nm

Environment +4 to +40 °C

20–95% relative humidity 84–106 kPa (840–1060 mbar)

atmospheric pressure

Flow cell

Max. flow rate 100 ml/min

Max. pressure 2 MPa (20 bar, 290 psi)

Backpressure Max. 0.5 bar at 2 ml/min with water at

25 ℃

Liquid temperature range +4 to +40 °C

Optical path length

2 mm cell 2 mm 10 mm cell 10 mm

Cell volume

2 mm cell 2 μ l 10 mm cell 8 μ l Degree of protection IP 21

Wetted materials PTFE (polytetrafluoroethylene)

PEEK (polyetheretherketone) Titanium (palladium alloy) Quartz (synthetic fused silica)

pH stability range 1–13, 1–14 (<1 days exposure)

 $^{^{\}scriptscriptstyle \perp}$ Measured with water at 1 ml/min, time constant 1 s.

²¹ Typical values at room temperature after varm-up.

Chemical resistance

The wetted parts are resistant to organic solvents and salt buffers commonly used in chromatography of biomolecules, except 100% Ethyl acetate, 100% Hexane, and 100% Tetrahydrofuran (THF) UNF 10-32 "fingertights" for capillary tubing with 1/16" outer diameter

Tubing connections

Physical data

Light source Lamp lifetime Control

Power consumption Power requirement Analogue output Digital inputs

Display
Dimensions (H xW xD)
Weight
Degree of protection

Compliance with standards

Safety Standards

Xenon flash lamp > 4000 hours

Stand alone or from a computer running UNICORN 3.21 or higher through UniNet-1 connection

65 VA

100-240 V AC, 50-60 Hz

3 signals, 0–1 V full scale, overrange function 5 V, 1 mA current sinking, lamp on/off,

autozero, event mark

2 rows with 20 characters each

200 x 260 x 370 mm

8.5 kg IP 20

The declaration of conformity is valid for the instrument only if it is:

- used in laboratory locations
- used in the same state as it was delivered from GE Healthcare except for alterations described in the User Manual
- connected to other CE labelled GE Healthcare modules or other products as recommended.

This product meets the requirement of the Low Voltage Directive (LVD) 73/23/EEC through the following harmonized standards:

- EN61010-1
- IEC 61010-1
- CAN/CSA-C22.2 No. 61010-1
- UL61010-1

EMC standards

This device meets the requirements of the EMC Directive 89/336/EEC through the following harmonized standards:

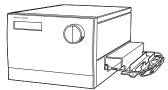
- EN 61326 (emission and immunity)
- EN 55011, GR 2, Class A (emission)
- This device complies with part 15 of the FCC rules (emission). Operation is subject to the following two conditions:
 - 1 This device may not cause harmful interference.
 - 2 This device must accept any interference received, including interference that may cause undesired operation.

D Accessories and spare parts

Item	Quantity per pack	Code no.
Monitor UV-900 complete but without	1	18-1108-35
flow cells		
Flow cell UV-900/2 (2 mm)	1	18-1111-10
Flow cell UV-900/10 (10 mm)	1	18-1111-11
Signal cable	1	18-1110-64
Fibre detachment tool	1	18-1111-16
Teflon tubing, i.d. 1/8", o.d. 3/16"	3 m	18-1112-47
Tubing connector for 3/16" o.d. tubing	10	18-1112-49
Ferrule for 3/16" tubing	10	18-1112-48
Stop plug, 5/16"	5	18-1112-50
Stop plug, 1/16"	5	18-1112-52
Union Luer female/1/16" male	2	18-1112-51
Union 1/16" female/M6 male	6	18-1112-57
Union M6 female/1/16" male	8	18-1112-58
Union 1/16" male/ 1/16" male, i.d. 0.25 mm	2	18-1120-92
Union 1/16" male/ 1/16" male, i.d. 0.50 mm	2	18-1120-93
PEEK tubing, i.d. 0.15 mm, o.d. 1/16"	2 m	18-1156-59
PEEK tubing, i.d. 0.25 mm, o.d. 1/16"	2 m	18-1120-95
PEEK tubing, i.d. 0.50 mm, o.d. 1/16"	2 m	18-1113-68
PEEK tubing, i.d. 0.75 mm, o.d. 1/16"	2 m	18-1112-53
PEEK tubing, i.d. 1.0 mm, o.d. 1/16"	2 m	18-1115-83
Tefzel tubing, i.d. 0.25 mm, o.d. 1/16"	2 m	18-1121-36
Teflon tubing, i.d. 0.75 mm, o.d. 1/16"	2 m	18-1112-54
Fingertight connector 1/16"	10	18-1112-55
UV-900 1 mm Calibr. kit	1	18-6324-01
UV-900 2 mm Calibr. kit	1	18-6324-02
UV-900 5 mm Calibr. kit	1	18-6324-04
UV-900 10 mm Calibr. kit	1	18-6324-05
UV-900 Cell calibr. Excel-file	1	18-6324-06

Short instructions

The following short instructions are intended as a guide for users who are fully familiar with the safety precautions and operating instructions described in this manual. The instructions assume that the unit is installed according to the installation instructions.





- 1 **Switch on the module** with the mains switch on the rear panel.
- 2 **Switch on the UV-lamp** by selecting **Run**.
- 3 **Select wavelength** for one, two or three wavelengths.

- 4 Autozero the module by pressing OK.
- 5 **The main operating menu** (RUN-menu) is shown.

Calibrating
Please wait

End	
Press OK to	Run

Set Wavelength (215, 254, 280 nm)

Set Wavelength λ 1 (215nm)

Set Wavelength λ 2 (254nm)

Set Wavelength $\lambda 3$ (280nm)

Autozero

λ1(215)	1.123 AU
λ2(254)	0.02345 AU
λ3(280)	1.123 AU

www.chromatography. amershambiosciences.com

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