ÄKTA design

Monitor UPC-900

User Manual

18-1125-55

Amersham Biosciences

Important user information

All users must read this entire manual to fully understand the safe use of ÄKTA™ monitor UPC-900 Workstation.

Safety symbols

The following Warning symbols highlights instructions that must be strictly followed in order to avoid personal injury. Be sure not to proceed until the instructions are clearly understood and all stated conditions are met.

WARNING! Read the instruction to avoid hazardous conditions.

WARNING! Avoid exposure to hazardous laser radiation.

Caution notices

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

Notes

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

CE Certifying

This product meets all requirements of applicable CE-directives. 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 Amersham Biosciences instruments, or
- connected to other products recommended or described in this manual, and
- used in the same state as it was delivered from Amersham Biosciences except for

  alterations described in this manual.

WARNING!

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

Terms and Conditions of Sale

Unless otherwise agreed in writing, all goods and services are sold subject to the terms and conditions of sale of the company within the Amersham Biosciences group which supplies them. A copy of these terms and conditions is available on request.

Should you have any comments on this product, we will be pleased to receive them at:

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Trademarks

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Short instructions on back page

About this manual

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

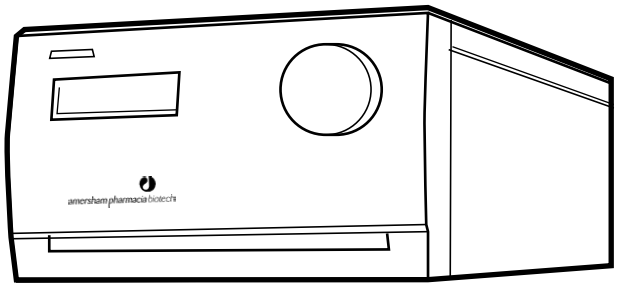
Sections 1 – 5 contain the necessary information for operating the instrument.

1 Introduction

1.1 General

Monitor UPC-900 is a high precision on-line monitor for the combined measurement of UV absorption, pH and conductivity in liquid chromatography. The UPC-900 features:

- Fixed wavelengths of 254, 280 nm (Hg lamp), or 214 nm (Zn lamp).
- Other wavelengths of 313, 365, 405, 436 and 546 nm through filter change.
- Two alternative flow cells with pathlengths of 2 mm and 5 mm.
- Fast response
- High accuracy and reproducibility
- Flow cells with low dead volume
- Accurate and reliable monitoring through self-test and self-calibration
- Flow cells can be connected close together, minimising band broadening and time delay between detectors.



The monitor and the flow cells are separate units. All flow cells are connected to the rear of Monitor UPC-900.

1.2 Safety

- The module is designed for indoor use only.
- Do not use in a dusty atmosphere or close to spraying water.
- Operate in accordance with local safety instructions.

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

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

WARNING! The module must not be opened by the user. It contains high voltage circuits that can deliver a lethal electric shock.

WARNING! When lamp power is on, the lamp socket carries a dangerous voltage. Do not connect/disconnect with the monitor switched on.

WARNING! The module uses high intensity ultra-violet light. Do not remove the UV lamp while the monitor is running.
Before changing a UV lamp, ensure that the lamp cable is disconnected from the monitor to prevent injury to eyes.
If the mercury lamp is broken, make sure that all mercury is removed and disposed of according to national and local

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

WARNING! HCl and NaOH are injurious to health. Avoid spillage.

WARNING! Hg lamps contain small amounts of mercury and must be handled with care and disposed of according to national or local environmental regulations.

2 Installation

2.1 Unpacking

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

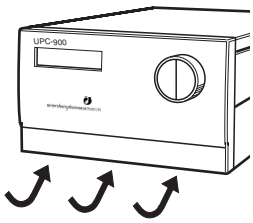
Note: *It is important that the filters, flow cells and lamps are not handled during unpacking. For protection of these items, they should remain in their packing materials until required for use.*

We recommend that all packing materials be retained if onward transport of the module is expected.

Note: *pH measurement is optional in ÄKTA*FPLC.

CAUTION! Read the following information carefully to ensure that the module is installed correctly.

2.2 General precautions

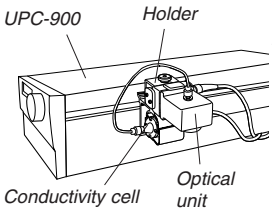


The module should be located in a place of low temperature variations, away from heat sources, draughts and direct sunlight.

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

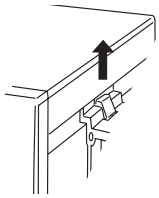
The module should be installed on a stable laboratory bench or in ÄKTA FPLC™ system rack. To ensure correct ventilation, a free space of 0.1 m is required behind and in front of the module. Do not place soft material under the module to ensure that the ventilation inlet below the front is not blocked.

2.3 Installing the optical unit



Installing the holder

1 Hook the holder into the slot on the right hand side of the module. Secure it by pushing up the slide clamp.



Flow cell installation

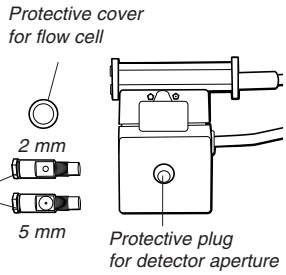
There is one analytical (5 mm) and one preparative (2 mm) flow cell available (stand-alone units only include the preparative flow cell). Both cells are installed in the same way, as described below:

1 Remove the red protective plugs from the detector housing and the flow cell.

2 Insert the flow cell into the detector housing from above.

Note: *The flow cell can only be placed in one correct position.*

3 Secure the flow cell by turning the locking nut until the stop position.

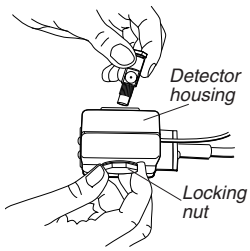


Note: *If the locking nut is not tightened sufficiently, the monitor will function poorly (e.g. drifting base-line).*

4 Place the protective cover around the flow cell to protect the electronics inside the optical unit from liquid spillage.

Note: *Avoid spillage for prolonged monitor lifetime.*

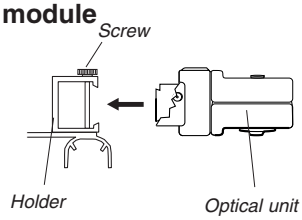
5 To remove the flow cell, reverse the procedure.



Note: *Ensure that the Hg lamp position and the filter is selected according to the wave length to be used. This is described in the Changing lamp assembly section below.*

Connecting the optical unit to the module

1 Place the optical unit in the holder, or in a suitable location as close to the column as possible. The optical unit can be placed up to 1.5 m from the monitor housing.



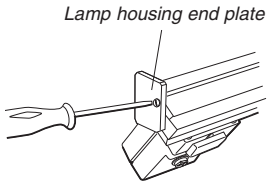
2 Secure it by tightening the screw in the holder.

3 Connect the lamp cable to the socket **Lamp** on the rear panel of the module.

4 Connect the signal cable to the socket **Optical unit** on the rear panel of the module.

Changing lamp assembly

WARNING! The module uses high intensity ultra-violet light. Do not remove the UV lamp while the monitor is running.
Before changing a defective lamp, ensure that the lamp cable is disconnected from the monitor to prevent injury to eyes.
If the mercury lamp is broken, make sure that all mercury is removed and disposed of according to national and local environmental regulations.



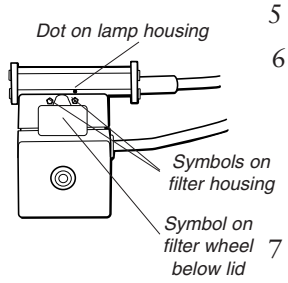
1 Use a Philips screwdriver to detach the end plate by removing one and loosening the other of the two holding screws on the lamp housing to be removed.

2 Slide the lamp housing off the filter housing.

3 Detach the end plate, as in step 1 above, from the lamp housing to be fitted to the optical unit.

4 Slide the lamp housing onto the filter housing. The lamp and signal cables should be on the same side. As you slide the lamp housing into position, depress the two pressure pads on the filter housing in sequence to facilitate the installation.

5 Refit the lamp housing end plate.



6 Slide the lamp housing firmly into place. There will be a faint click when the housing is positioned correctly. The Hg lamp housing can take up two positions, one for 280 nm, marked by ◻ on the filter housing, and the other marked by ◼ for all other wavelengths. The Zn lamp housing has only one position.

7 Set the wavelength to be used by selecting lamp position (indicated by a dot on the lamp housing) in combination with the appropriate filter, i.e. the dot on the lamp housing should be adjacent to the symbol on the filter housing corresponding to the symbol on the filter wheel for the filter to be used. A click will indicate that the filter is in position.

Note: *The wavelength and the flow cell type should also be entered in the Questions menu in UNICORN.*

Filter change

The Hg optics with 254 and 280 nm filters and the Zn optics with the 214 nm filter are delivered with filters installed. If other filters are to be used, install the new filters as described in *2.10 Installing optical filters (optional)*.

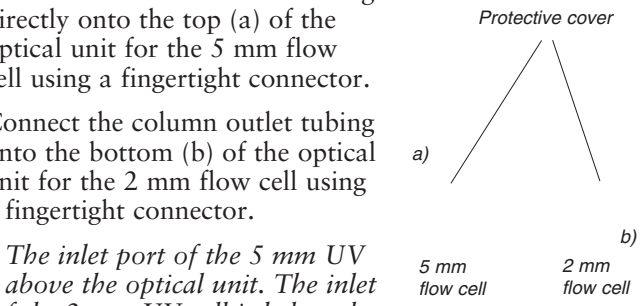
Connection to the column

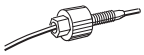
1 Fix the optical unit directly under the column if possible.

Note: *Always position the optical unit with the filter wheel cover facing upwards.*

2a Connect the column outlet tubing directly onto the top (a) of the optical unit for the 5 mm flow cell using a fingertight connector.

2b Connect the column outlet tubing onto the bottom (b) of the optical unit for the 2 mm flow cell using a fingertight connector.





Note: *The inlet port of the 5 mm UV cell is above the optical unit. The inlet port of the 2 mm UV cell is below the optical unit.*

3 Screw to fingertightness.

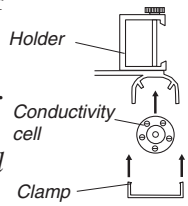
4 Connect the optical unit outlet tubing onto the opposite hole in the flow cell. Use fingertight connectors.

If no outlet tubing exists, cut a piece of PEEK tubing (i.d. 0.5 mm, o.d. 1/16"). The length should be 170 mm when using the 5 mm flow cell, and 230 mm when using the 2 mm flow cell.

5 Connect the other end of the tubing to the conductivity flow cell or to another appropriate unit.

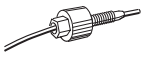
2.4 Installing the conductivity flow cell

1 Place the conductivity cell in its cell holder, or in a suitable location, as close to the optical unit/column as possible. The cell can be placed up to 1.5 m from the monitor housing. Secure the cell with the clamp.



Note: *When the conductivity flow cell is used in conjunction with the pH electrode, place the conductivity flow cell and select its flow direction so that the screw head end of the flow cell faces the pH flow cell.*

2 Connect the conductivity cell to the socket **Conductivity Flow Cell** on the rear panel of the module.

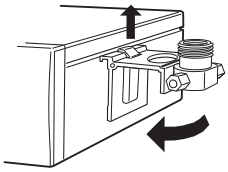


3 Connect the tubing with fingertight connectors.

2.5 Installing the pH flow cell and electrode

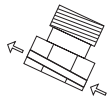
Mounting the flow cell holder

In ÄKTAFPLC systems, the pH electrode is optional.

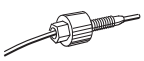


1 Hook the flow cell holder on the right hand side of the housing. Secure it with the slide clamp.

If the flow cell holder is not used, the flow cell must still be installed at an angle of 30° from the vertical with the outlet placed higher than the inlet to prevent air bubbles being trapped in the cell.



The flow direction is marked on the flow cell.



2 Connect the tubing with finger-tight connectors.

Inserting the pH electrode

Note: *Handle the pH electrode with care.*

> **CAUTION!** The tip of the pH electrode consists of a thin glass membrane. Protect it from breakage, contamination and drying out or the electrode will be destroyed. Always store the electrode with the end cover filled with a 1:1 mixture of pH 4 buffer and 2 M KNO_3. **Do NOT store in water only**.

1 Unpack the pH electrode. Ensure that it is not broken or dry.

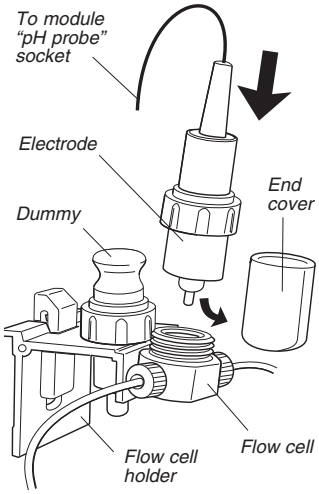
2 Before using the electrode, remove the electrode end cover and immerse the glass bulb in buffer for 30 minutes.

3 Remove the dummy electrode from the flow cell and store it in the flow cell holder.

4 Carefully insert the electrode in the flow cell. Tighten the nut by hand to secure the electrode.

Note: *If the flow cell is full of liquid, it is not possible to insert the electrode. If so, loosen the inlet connection while inserting the electrode to allow the liquid to run out from the flow cell. Remember to re-tighten the connector.*

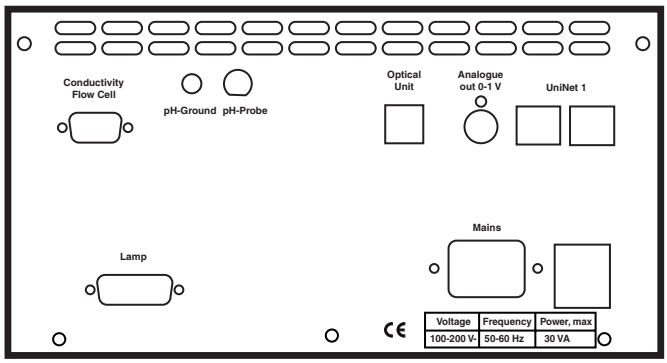
Note: *If the electrode is not fully inserted, the system will leak and a dead volume will occur in the holder.*



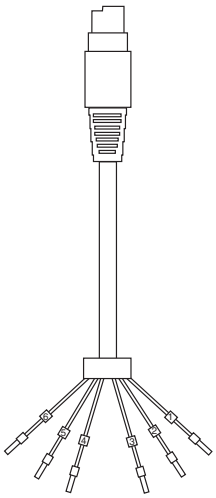
5 Connect the pH electrode cable to the socket **pH Probe** on the rear of the module.

2.6 Connecting electrical signal cables

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



Connecting to chart recorder (if used)



The external chart recorder outputs for UV, pH and conductivity from the monitor are 0–1 V.

1 Connect the chart recorder to the mini-DIN-socket **Analogue out 0-1 V** using the cable supplied. Pin designations for the signals are as follows:

Pin no.	Signal	Range	Value (full scale)
1	UV	0-1 V	0.001-5 AU
2	signal ground	0 V	-
3	conductivity	0-1 V	0.1 mS/cm-999.9 mS/cm
4	signal ground	0 V	-
5	pH	0-1 V	0-14
6	signal ground	0 V	-

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.

2.7 Connecting to a communication link

CAUTION! The mains power to ÄKTAFPLC must be switched OFF before connecting the module to the *UniNet 1* link.

The monitor can be used as a stand-alone module or in ÄKTAFPLC. As a stand-alone module, it is controlled from the front dial and display. In ÄKTAFPLC, the monitor is controlled from a PC running UNICORN™ version 3.0 or higher via *UniNet 1* cables, or manually from the front dial and display.

Connect two *UniNet 1* cables to the *UniNet 1* connectors. The module can be connected in series anywhere between the PC and a termination plug. The *UniNet* 1 link connects, in series, the PC with Pump P-920, Monitor UPC-900 and the Frac-900. The termination plug is connected to the last module in the chain.

2.8 Connecting to supply voltage

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

1 Make sure the on/off switch is in the OFF-position (**0**).

2 Connect the supplied mains cable between the module and a grounded mains socket. Any voltage from 100–240 V AC, 50–60 Hz can be used.

Note: *The module contains no user-replaceable fuse.*

2.9 Preparing the module for use

Before performing the following procedures, we recommend you read sections 3.1–3.3.

- Calibrate the pH electrode, see section *3.6*.
- Set the conductivity cell constant, see section *B.2.3*. (Only required when the flow cell has been replaced.)
- Calibrate the temperature sensor, see section *B.2.6*. (Only required if the monitor is to be used for high accuracy measurements, or if the flow cell has been replaced.)

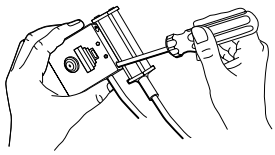
Note: *The conductivity cell constant is shown on the packaging. Retain the packaging in case the conductivity cell constant needs to be re-entered.*

Note: *The measured temperature is the temperature in the conductivity flow cell, which can differ from the ambient temperature in ÄKTAdesign systems.*

Note: *When running chromatography using organic solvents, we recommend that the pH electrode is removed and the dummy electrode inserted in its place as organic solvents will cause pH electrode degeneration.*

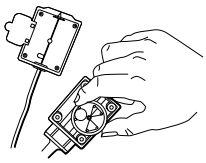
2.10 Installing optical filters (optional)

The Hg optics with 254 and 280 nm filters and the Zn optics with the 214 nm filter are delivered with filters installed. If other filters are to be used, install them as follows:



1 If the Zn lamp is attached, remove the lamp housing as described in section *Changing the UV lamp*.

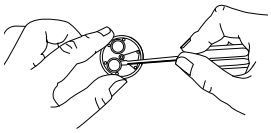
2 Remove the four screws in the filter housing. Separate the filter housing from the detector housing.

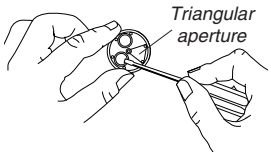


3 Carefully remove the filter wheel from the filter housing.

4 If necessary, remove the filter(s) from the filter wheel by pressing it(them) out, e.g. with a small screwdriver.

Note: *Filters are sensitive optical components. Never touch the optical surfaces or expose them to temperatures above 60 °C. Clean them with dry lens cleaning tissue and store them, when not in use, in the box in which they were supplied. Heavy contamination may be removed by using a lens tissue dipped in ethanol.*

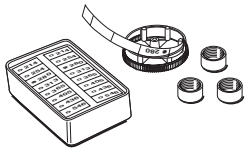




5 Insert the filter(s) of choice into the filter wheel (maximum 3 filters) with the correct orientation (the mirror side facing upwards) and position over one of the three triangular apertures. The filters snap in by pressing them quite firmly. Do not touch the filter surface.

6 Remove the circular plastic band showing the wavelength(s).

7 Remove labels from the band if necessary.



8 Place the correct labels on the band with the label designation facing outwards. Ensure that the label position corresponds to the filter position, i.e. the label should be placed opposite the filter.

9 Reassemble the circular plastic band with the filter wheel peg fitting into the band notch.

10 Check that all filters are clean. Insert the filter wheel back into the filter housing.

Note: *The filter wheel can be placed only in correct position.*

11 Reassemble the filter housing with the detector housing by fastening the four screws.

3 Operation

3.1 On/off

Selftest **Please wait...**

Switch on the module at the mains switch on the rear panel. At switch-on, the module performs a selftest. Several beeps are heard during this process. If an error is detected, an error message is shown.

UPC-900 **<version no.>**

Name and software version number is shown for 2 seconds.

Calibrating

All parameters are set to factory default values.

Calibration ok

AU Cond% pH

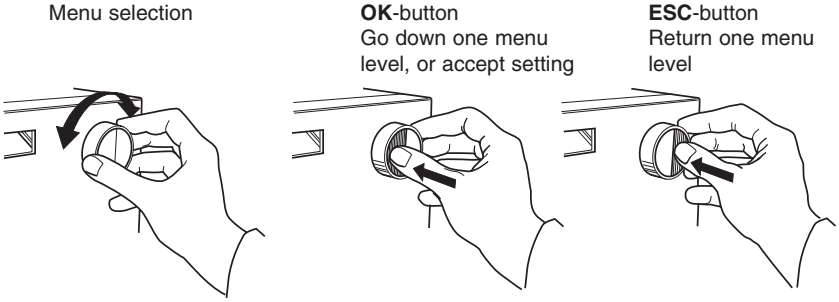
The selftest takes approx. 1 minute. When start-up is completed with no errors, the display shows the main menu 1.

The monitor can be used immediately but the full specifications are not obtained until after 1 hour of lamp warm–up.

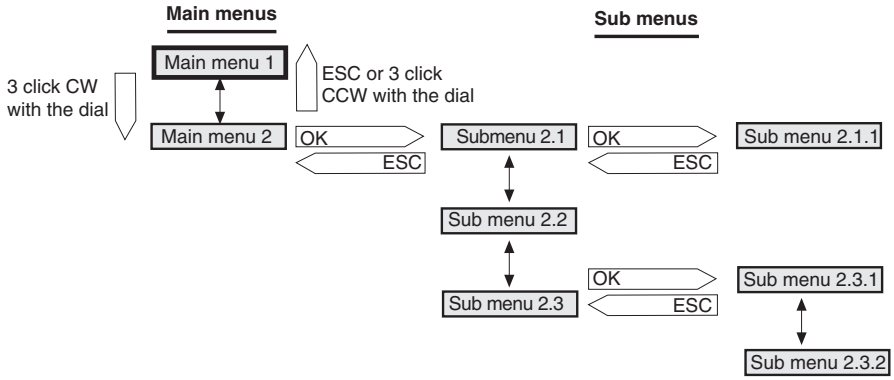
3.2 Menu selection and settings

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 **OK**. Pressing **ESC** moves back one menu level.

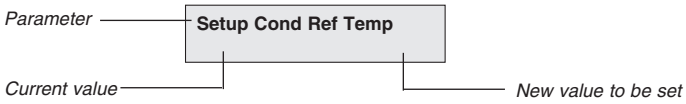


Return to main menu

Pressing **ESC** repeatedly always returns to the **main menu 1,** which is the main operating menu.

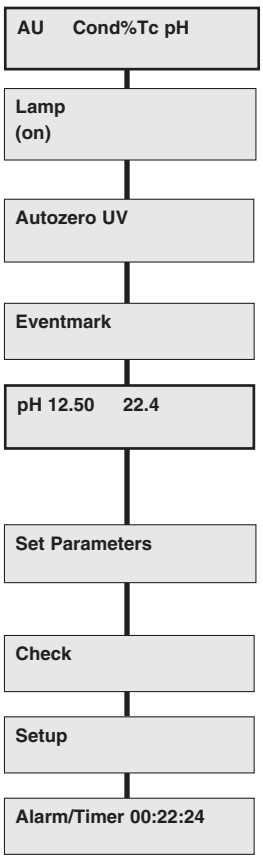
Select value

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



To simplify entering large numerical values, the cursor moves to next digit if the dial is turned quickly in one direction. 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 **OK**. To cancel, press **ESC**.

3.3 Main menu overview



Main menu 1. This menu is accessed from all positions by pressing **ESC**.

Lamp control. This menu is accessed by turning the dial one step counter-clockwise from main menu 1. It is used to manually switch the lamp on/off.

Autozero. Used to zero the UV signal from the monitor.

Eventmark. Used to set eventmarks on the UV curve.

Main menu 2. Used to display pH, temperature (in the conductivity flow cell) and conductivity in mS/cm as well as in percentage of full range.

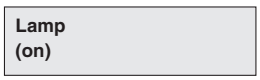
Parameter menus. Used to set measurement parameters for **Cond**, **pH** and **UV**.

Check menus. Checking internal operating values. See *Reference information* section *B.1*.

Setup menus. Setting up language, unit number, etc. See *Reference information* section *B.2*.

Alarm/Timer menus. Setting different timer options. See *Reference information* section *B.3*.

3.4 Controlling the UV lamp

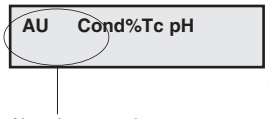


The **Lamp** menu is accessed by turning the dial one step counter-clockwise from main menu 1, and then pressing **OK**. We recommend the lamp be switched off to conserve lamp operating time when no measurement is being made. A warm-up time of 60 minutes is required to achieve full specifications. However, in most cases, a warm-up time of 15 minutes is sufficient.

Switching the lamp on/off

1 Switch the lamp on/off by moving the cursor with the dial, and then pressing **OK**.

3.5 Reading the UV absorbance value



The main menu 1 shows the absorbance value with four digits for the chosen wavelength. This menu is reached from any other menu by pressing **ESC**.

If the lamp is off, **Lamp Off** is displayed instead.

3.6 Autozeroing UV

The autozero function in the UV part of the module sets the detected absorbance to zero when **OK** is pressed. Using autozero is recommended at the start before the sample is injected.

Autozero UV

1 From main menu 1, turn the dial one step clockwise to select **Autozero UV**, then 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**.

3.7 Creating eventmarks

Eventmarks can be set and displayed as spikes on the UV curve, for example, when the sample is injected. The spikes are 10% of the full scale AU setting.

Eventmark

1 From main menu 1, turn the dial two steps clockwise to select **Eventmark**, then press **OK**. A spike on the UV curve is created.

3.8 Setting Cond, pH and UV analogue outputs

This settings menu is used to set measurement parameters (zero and full range values) for **Cond**, **pH** and **UV** on the analogue output channels. The analogue output channels can be connected to a chart recorder, for example.

Set Parameters

From the main menu 1, turn the dial four steps clockwise to enter the menu **Set Parameters**. Click **OK**, select the quantity to set by moving the cursor with the dial, then click **OK** again.

Set Parameters

Note: *The user interface of the monitor must be unlocked if connected to a UNICORN control system.*

Setting the Cond analogue output

Set Cond Analog. Out

1 Current analogue settings are displayed (zero and full scale values).

Set Cond Zero Level	2	Press **OK** to access the settings menu. Press **OK.**
Set Cond Zero Level	3	Set the desired zero value. The range is 0.000 - 999.9 mS/cm. Press **OK** to acknowledge**.**
Set Cond Full Scale	4	Turn the dial clockwise to access the settings menu. Press **OK.**
Set Cond Full Scale	5	Set the desired full scale value. The range is 0.000 - 999.9 mS/cm. Press **OK** to acknowledge**.** Press **ESC** two twice to return to menu **Set Parameters.**

Setting the pH analogue output

After having selected **Set Parameters pH** and pressed **OK**, turn the dial one step clockwise to skip the **Calibrate pH** menu. Then press **OK.**

Note: *The pH values for zero level and full scale must differ by at least 1 pH unit.*

Note: *The zero level and full scale values can be calibrated in any order.*

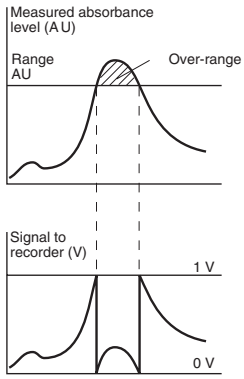
Set pH Analogue Out	1	Current analogue settings are displayed (zero and full scale values).
Set pH Full Scale	2	Press **OK** to access the full scale settings menu. Press **OK.**
Set pH Full Scale	3	Set the desired full scale value. The range is pH -0.50 - 14.30. Press **OK** to acknowledge**.**
Set pH Zero Level	4	Turn the dial clockwise one step to access the zero level settings menu. Press **OK.**
Set pH Zero Level	5	Set the desired zero level value. The range is pH -0.50 - 14.30. Press **OK** to acknowledge**.** Press **ESC** twice to return to menu **Set Parameters.**

Setting the UV analogue output

Set UV Analogue Out	1	Current analogue settings are displayed (zero and full range values). Allowed full range values are 0.0001, 0.0002, 0.0005, 0.001, 0.002, 0.005, 0.01, 0-02, 0.05, 0.1, 0.2, 0.5, 1.0, 2.0 and 5.0. Zero level is set as a percentage of full scale.

Set UV Range	2 Press **OK** to access the settings menu. The current setting is displayed. Press **OK.**
Set UV Range	3 Set the desired full range value. Press **OK** to acknowledge**.**
Set UV Zero Level	4 Turn the dial clockwise to access the settings menu. The current setting is displayed. Press **OK.**
Set UV Zero Level	5 Set the desired zero level value. Press **OK** to acknowledge**.** Press **ESC** twice to return to menu **Set Parameters.**

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



3.9 Calibrating conductivity

The cell constant for the particular flow cell is written on the flow cell packaging. Refer to section *B.2* in *Reference information* for how to enter the cell constant.

Adjustment of the cell constant is only necessary when the monitor is to be used to determine conductivity with high accuracy. The procedure is described in *Reference information* section *B.2*. Calibration can also be performed from UNICORN.
Select **System Control:System:Calibrate**.

3.10 Calibrating the pH electrode

A good laboratory routine is to calibrate the module once a day, when the electrode is replaced and if the ambient temperature changes. The pH electrode is calibrated using standard buffer solutions in a two point calibration. The two buffer solutions can have any pH value as long as the difference between them is at least 1 pH unit. Calibration can also be performed from UNICORN. In UNICORN, select **System Control:System:Calibrate**. Select the pH monitor. The calibration procedure can be done with the pH electrode either fitted in or removed from the flow cell.

Calibrating with the electrode outside the flow cell

When calibrating the electrode out of the flow cell and changing from one buffer to another, rinse the electrode tip with distilled water and dab it carefully with a soft tissue to absorb the remaining water. Do NOT wipe the electrode as this may charge it and give unstable readings.

The steps below describe the procedure used with the electrode removed from the flow cell.

Note: *The user interface of the monitor must be unlocked if connected to a UNICORN control system.*

1 Remove the pH electrode from the flow cell and immerse the electrode in the first standard buffer solution (normally pH 7.0).

Set Parameters

2 From main menu 1, turn the dial four steps clockwise to enter menu **Set Parameters**. Press **OK**.

Set Parameters

3 Select **Set Parameters pH** . Press **OK**.

Calibrate pH (7.00 - 12.00)

4 Select menu **Calibrate pH.** Current calibration values are displayed (buffer 1 - buffer 2).
Buffer 1=fixed lower calibrated pH value. Range=0.00-14.00
Buffer 2=fixed higher calibrated pH value. Range=0.00-14.00

Note: *The pH values for buffer 1 and 2 must differ by at least 1 pH unit.*

Calibr pH Buffer 1

5 Press **OK** to access the settings menu. The order of calibration, buffer 1 or buffer 2, is optional. Press **OK** to start with buffer 1, or turn the dial to start with buffer 2. In this example, we start with buffer 1.

Calibr pH Buffer 1

6 This text disappears when the reading is stable and the following text is then shown:

Calibr pH Buffer 1

7 Adjust the pH value in the display using the dial so that it corresponds to the known pH value of the first buffer solution and press **OK**.

Calibr pH Buffer 2

8 Turn the dial clockwise to access the buffer 2 calibrating menu. Rinse the electrode tip with distilled water and then immerse the electrode in the second buffer solution (e.g. pH 4.0 or 9.0) and press **OK**.

Calibr pH Buffer 2

9 This text disappears when the reading is stable and the following text is then shown:

Calibr pH Buffer 2

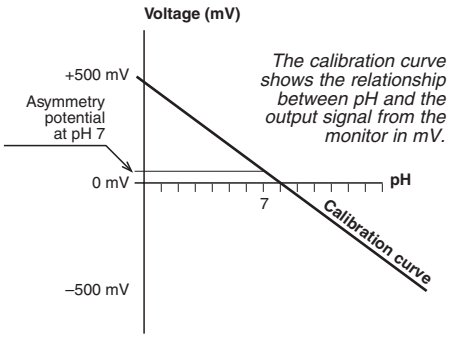
10 Adjust the pH value in the display using the dial so that it corresponds to the known pH value of the second buffer solution and press **OK.**

11 When the two point calibration is performed successfully, the following text is shown for one second:

Calibration OK

Calibrated Electrode

12 From the **Calibrate pH Buffer 2** menu, turn the dial one step clockwise to access sub menu **Calibrated Electrode Slope.** This menu shows the slope of the calibration curve, where 100% corresponds to 59.16 mV per pH step at 25 °C. The asymmetry potential at pH 7 is shown as a mV value. Press **ESC** to return to the **Set Parameters** menu.



13 Before use, rinse the electrode using distilled water.

A new electrode typically has a slope of 95 - 102% and an asymmetry potential within ±30 mV. As the electrode ages, the slope decreases and the asymmetry potential increases.

As a rule, when an electrode has an asymmetry potential outside of ±60 mV and a slope lower than 80%, and no improvement can be achieved by cleaning, it should be replaced.

An electrode is still usable at lower slopes and higher asymmetry potentials but the response will be slower and the accuracy diminished.

Calibrating with the electrode in the flow cell

When calibrating with the electrode fitted in the flow cell in ÄKTAFPLC, follow the above procedure but let at least 30–35 ml (with 2 ml mixer) of standard buffer solution be pumped through the system to stabilize pH. Leave the pump running while calibrating. Switch to the second standard buffer solution and repeat the procedure. For a description of calibration from UNICORN with the electrode fitted in the flow cell, see Chapter 6 in the *UNICORN User Manual.*

3.11 Filtering noise in the UV signal

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 according to the table below may be distorted. Because of this, the averaging time should be as short as possible. On delivery, the averaging time is set to 1.3 s.

1 From main menu 1, turn the dial six steps clockwise and press **OK** twice.

2 Select **Setup UV** and press **OK**.

3 The menu **Set Averaging** is displayed, showing the current set averaging time. Press **OK**.

4 Set the desired value and press **OK**. Values allowed are 0.02, 0.04, 0.08, 0.16, 0.32, 0.64, 1.3, 2.6, 5 and 10 s.

Averaging time (s)	*Corresponding time constant (s) (approximately)*	*Min. peak width at half height (s)*
10.0	5	50
5.1	2	32
2.6	1	16
1.3	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
0.04	0.01	0.2
0.02	0.01	0.1

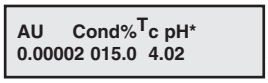
In UNICORN, the averaging time is set with the instruction **AveragingTime** in **System Control:Manual:Alarm&Mon**

3.12 Changing UV flow cell

The flow cell can be changed when required, for example, from 2 mm to 5 mm when the sensitivity of the measurement must be increased due to a small amount of sample being applied, or from 5 mm to 2 mm when a lower sensitivity is desired, due to output signal limitation.

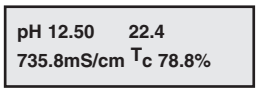
See section *2.3 Installing the optical unit*, sub-section *Flow cell installation*.

3.13 Reading pH, temperature and conductivity values



The main operating menu 1 shows UV absorption, conductivity in percentage of full range and pH. If the pH value is not stable or is changing, an asterisk is displayed, i.e. **pH***. This menu is reached from any other menu by pressing **ESC**.

If temperature compensation is switched on, the display will show **Cond% Tc** instead of **Cond%**, see section *B.2.4 Setup conductivity temperature compensation* in *Reference information*.



By turning the dial three steps clockwise, main operating menu 2 is shown instead. This display shows pH, temperature (in the conductivity flow cell) and the actual conductivity in mS/cm together with the percentage value. If temperature compensation is switched on, **Tc** is shown in the display.

The display of pH, temperature and conductivity can be disabled, see Section *B.2* of *Reference information*.

3.14 Storage and shut-down

Storage of the UV flow cell

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

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.

Storage of the conductivity flow cell

Overnight: The conductivity cell can be left filled with a buffer.

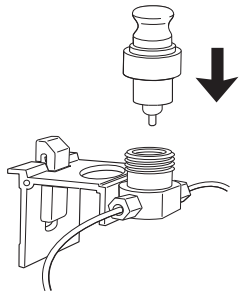
Weekend or long term storage: Flush the conductivity cell with water and fill with 20% ethanol.

Storage of the pH electrode

CAUTION! Never leave the pH electrode in the flow cell for any period of time when the system is not used, since this may cause the glass membrane of the electrode to dry out. Dismount the pH electrode from the flow cell and fit the end cover filled with a 1:1 mixture of pH 4 buffer and 2 M KNO_3.

Do NOT store in water only.

The pH electrode should **always** be stored in a 1:1 mixture of pH 4 buffer and 2 M KNO_3 when not in use. When the pH electrode is removed from the flow cell, the dummy electrode (supplied) can be inserted in the flow path.



Electrode regeneration: If the electrode has dried out, immerse the lower end of the electrode in buffer with a 1:1 mixture of pH 4 buffer and 2 M KNO_3 overnight.

3.15 Restart after power failure

If the power supply to the module is interrupted, it automatically restarts when power is restored and displays the main operating menu. All set values are retained in the module and the lamp is switched on.

4 Maintenance

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

CAUTION! Only spare parts approved or supplied by Amersham Biosciences may be used for maintaining and servicing the module.

4.1 Periodic maintenance

Interval	**Action** (see procedures below)
Every 3 months	Check the monitor
Every 6 months or more often if required	Change the pH electrode Clean the UV flow cell
When required	Clean the conductivity cell Clean the pH electrode

4.2 Cleaning and checking the module

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

Lamp intensity

Check Lamp Intensity

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

2 Select menu **Check Lamp Intensity**.

If:
R <300 mV for 254 nm
R <150 mV for 280 nm
R <150 mV for 214 nm,
replace the lamp according to Section *4.10 Changing the UV lamp*, or contact Amersham Pharmacia Biotech for lamp replacement.

Lamp run time

Check Lamp Run Time
Hg 2300h Zn 340h

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

2 Select menu **Check Lamp Run Time**.

- The lifetime of a Hg lamp at 254 nm, in room temperature is typically 7000 hours (in coldroom, typically 2000h).
- The lifetime of a Hg lamp at 280 nm, in room temperature is typically 3500 hours.
- The lifetime of a Zn lamp is typically 2000 hours in room temperature.

When necessary, replace the lamp according to Section *4.10 Changing the UV lamp*, or contact Amersham Biosciences for lamp replacement.

Autozero

The module internal absorbance value for autozero can be checked to test the consistency of buffers.

Check Autozero

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

2 Select menu **Check Autozero**. The autozero absorbance value for the wavelength used is shown.

4.3 Cleaning the UV flow cell in-place

WARNING! NaOH is injurious to health. Avoid spillage.

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

4.4 Cleaning the UV flow cell off-line

A clean flow cell is essential for ensuring the correct operation of the UV-monitor.

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

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 continue to squirt five more 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 then flush the flow cell with distilled water (10 ml).

4.5 Cleaning the conductivity flow cell in-place

WARNING! NaOH is injurious to health. Avoid spillage.

Remove the pH electrode and install the dummy electrode in the pH flow cell.

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

4.6 Cleaning the conductivity flow cell off-line

WARNING! NaOH is injurious to health. Avoid spillage.

If the conductivity measurements are not comparable to previous results, the electrodes in the flow cell may be contaminated and require cleaning. To clean the flow cell:

1 Pump 15 ml of 1 M NaOH at 1 ml/min through the flow cell either by using a pump or a syringe.

2 Leave it for 15 minutes.

3 Rinse thoroughly with 50 ml distilled water.

Note: *If the flow cell is totally blocked, the blockage can be removed using a needle or a wire with a diameter less than 0.8 mm.*

4.7 Changing the conductivity flow cell

The conductivity flow cell can be changed when required. Make sure the monitor is switched off before disconnecting/connecting the cell from the rear of the monitor housing.

If the cell is replaced with a new flow cell, the monitor must be calibrated with the new cell constant value written on the flow cell package. See section *B.2* in *Reference information*. If the cell constant is not known, it can be determined (see also section *B.2* in *Reference information*).

4.8 Cleaning the pH electrode

Note: *The pH electrode has a limited lifetime and should be replaced every six months, or when the response time is slow or the slope is out of range (<80%).*

WARNING! HCl and NaOH are injurious to health.
Avoid spillage.

Use one of the following procedures to clean the electrode to improve the response:

- **Salt deposits**: Dissolve the deposit by immersing the electrode first in 0.1 M HCl, then in 0.1 M NaOH, and again in 0.1 M HCl. Each immersion is for a 5 minute period. Rinse the electrode tip in distilled water.
- **Oil or grease films**: Wash the electrode tip in a liquid detergent and water. If the film is known to be soluble in a particular organic solvent, wash with this solvent. Rinse the electrode tip in distilled water.
- **Protein deposits:** Dissolve the deposit by immersing the electrode in a 1% pepsin solution in 0.1 M HCl for five minutes, followed by thorough rinsing with distilled water.

If these procedures fail to rejuvenate the electrode, the problem is most likely a clogged liquid junction.

1 Heat a 1 M KNO_3 solution to 60 - 80 °C.

2 Place the electrode tip in the heated KNO_3 solution.

3 Allow the electrode to cool while immersed in the KNO_3 solution before re-testing.

If these steps fail to improve the electrode response, replace the electrode.

4.9 Changing the pH electrode

See section 2.4 *Installing the pH flow cell and electrode*.

5 Trouble-shooting

5.1 General

UPC-900
Version 1.00

When contacting Amersham Biosciences for support, state the program version of the module, which is shown for 2 seconds during start-up.

WARNING! The module must not be opened by the user. It contains high voltage circuits that can deliver a lethal electric shock.

5.2 Faults and actions

If the suggested actions do not correct the fault, call Amersham Biosciences.

UV measurement

No text on the front display	1	Check that the mains cable is connected and the power switch is in **ON-position I**.
Noisy UV-signal drift or instability	1	Select menu **Check Autozero** to check the autozero absorbance value. If AZ is between 1.5 and 2, there may be air bubbles in the flow cell, or the wrong buffer system in use.
	2	Wrong filter for the lamp used. Check that the lamp is in proper position and that the correct filter is used.
	3	The buffer may be impure. Check if the signal is still noisy with water.
	4	There may be air in the flow cell. Check that the flow restrictor gives a back-pressure of 0.2 MPa.
	5	If there is a lot of air in the water, degas the buffer before use.
	6	Check the connections of the optical unit.
	7	Clean the UV-cell, see sections 4.3 and 4.4.
	8	Locking nut in optical unit not properly tightened. Turn the locking nut to the stop position.
Ghost peaks	1	Check that there is no air in the eluents. Degas if necessary.
	2	Clean the system in accordance with the *ÄKTA*^{FPLC} *System Manual.*
	3	Clean the column in accordance with the column instructions.
	4	Check that the mixer is functioning correctly and that the correct chamber volume is being used.

Fault	*Action*
Low sensitivity	1 Aging lamp. Check the lamp and replace if necessary 2 Wrong lamp position. Check that the lamp position and the wavelength used (filter position) fit together. 3 Dirty on-line filter. Clean or replace the filter.
Error in external chart recorder	1 Check the chart recorder in accordance with its manual. 2 Test the recorder function by selecting recorder test according to section *B.1* in *Reference Information*.
pH measurement (optional in ÄKTAFPLC systems)	
No response to pH changes	1 Check that the electrode cable is connected properly to the rear of the module. 2 The electrode glass membrane may be cracked. If so, replace the electrode.
Small response to pH changes	1 Clean the pH electrode according to section *4.8* and recalibrate. 2 If the problem remains, replace the pH electrode.
Slow pH response or calibration impossible	1 Check the electrode glass membrane. If it is contaminated, clean the electrode following the instructions in section *4.8 Cleaning the pH electrode*. 2 If the membrane has dried out, the electrode may be restored by soaking it in buffer overnight. 3 Clogged liquid junction. Refer to section *4.8 Cleaning the pH electrode*.
Incorrect/unstable pH reading	1 Check that the electrode cable is connected properly to the rear of the module. 2 Check that the pump operates correctly. 3 Check that the electrode is correctly inserted in the flow cell and, if necessary, hand-tighten the nut. 4 If air in the flow cell is suspected, tap the flow cell carefully or tilt it to remove the air. Alternatively, flush the flow cell with buffer at 20 ml/min for 1/2 min. Use a flow restrictor after the pH electrode. 5 Check that the pH electrode is not broken. 6 Check that the pH electrode is calibrated. 7 Check the slope (see section *3.10 Calibrating the pH electrode*). If it is outside the range 80–105% or if the asymmetry potential deviates more than ±60 mV from 0 mV, clean the pH electrode. Recalibrate. If the problem persists, replace the pH electrode. 8 Clean the pH electrode if required, see section *4.8 Cleaning the pH electrode.* 9 Compare the response of the pH electrode with that of another pH electrode. If the response differs greatly, the electrode may require cleaning or replacement.

Fault	*Action*
	10 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.
	11 Check that the pH electrode has been calibrated at the correct temperature.
	12 In organic solvents such as ethanol, methanol and acetonitrile, stable pH measurements are not possible since dehydration of the membrane will occur. We recommend that the pH electrode is not used in applications using organic solvents. Mount the dummy electrode instead.
	13 Clogged liquid junction. Refer to section *4.8 Cleaning the pH electrode*.
pH values vary with varied back-pressure	1 Replace the pH electrode.
Conductivity measurement	
Incorrect or unstable reading	1 Check that the conductivity flow cell cable is connected properly to the rear of the module.
	2 Check that the pump operates correctly.
	3 If temperature compensation is being used, check that the temperature sensor is calibrated, and that the correct temperature compensation factor is in use.
	4 Check that the column is equilibrated. If necessary, clean the column.
	5 Check the operation of the mixer.
Baseline drift or noisy signal	1 There may be air in the flow cell. Use a flow restrictor after the flow cell and check that the flow restrictor gives a back-pressure of 0.2 MPa.
	2 Check for leaking tubing connections.
	3 Check that the column is equilibrated. If necessary, clean the column.
	4 Check the operation of the mixer and the pump.
	5 Clean the flow cell according to the procedure in section *4.5* or *4.6*.
Conductivity measurement with the same buffer appears to change over time	1 Clean the flow cell according to the procedure in section *4.5* or *4.6*.
	2 The ambient temperature may have changed. Use a temperature compensation factor, see section *B 2.4 Setting up conductivity temperature compensation*, in *Reference information*.
Absolute conductivity value wrong	1 Turn the flow cell so the end with the screws is facing the pH flow cell.
	2 Recalibrate the conductivity cell.
	3 Calibration solution, 1.00 M NaCl, not correctly prepared. Prepare a new calibration solution and recalibrate the conductivity cell.

Fault	*Action*
Ghost peaks appear in the gradient profile	1 A charged sample has been detected (e.g. protein). 2 Air bubbles are passing through the flow cell. Check for loose tubing connections. If necessary, use a flow restrictor after the conductivity flow cell.
Other problems	
Error in external pH or Cond. chart recorder	1 Check the chart recorder in accordance with its manual. 2 Test the recorder function and input voltage, which should be 1 V full scale. 3 Check the conductivity scaling and pH scaling, see section *3.8 Setting the Cond analogue output* and section *B.1.4 Checking recorder*.
No text on the front display	1 Check that the mains cable is connected and the power switch is in ON-position.

5.3 Error messages

If the suggested actions do not correct the fault, call Amersham Biosciences.

Message	*Action*
34 Start up failed Retry/Call service	1 Perform a new start-up. The preceding message may tell more about the cause. 2 If not, call service.
35 WARNING wrong averaging time set	1 Wrong value for averaging time set. See section 3.11 on how to set the averaging time relative to peak width.
50 Electrical error Call for service ↓ **57 Electrical error Call for service** **75 Electrical error Call for service**	1 Call for Service.
70 Lamp disconnected If not call service	1 Connect the lamp, or call for service.
71 WARNING low light intensity	1 Check connection between optical unit and monitor. 2 Check that lamp and filter position correspond. 3 Change lamp. If warning persists, call for service.
72 Change lamp or call for service **76 Change lamp or call for service**	1 If used in cold room, additional warm up might be needed. 2 If problem remains, change the lamp. 3 If problem remains, call for service.
73 WARNING Too much straylight leaks in	1 Check that filter wheel cover is closed. 2 Check that non-transparent tubings are used at UV flow cell inlet and outlet. 3 Check that optical unit is not exposed to direct sunlight. 4 If problem remains, call service
77 WARNING Autozero Out of range	1 Auto is not allowed from a level above 2 AU. 2 Check buffers. 3 Clean UV cell.

83 WARNING temp_cal will be changed	1 Press OK to accept the change. 2 Press ESC to skip the change.
84 WARNING cond_cal will be changed	
85 WARNING condscale (0-100%)<0.1mS	1 The difference between 0 and 100% must be at least 0.1 mS/cm. 2 Increase the span between zero and full scale setting. See section 3.8.
86 WARNING cond_cell bad/not connected	1 Check that the conductivity cell is connected. 2 Recalibrate temperature. 3 If problem remains, replace the conductivity cell.
87 WARNING pH-probe bad/not connected	1 Check the pH electrode connection. 2 Clean the pH electrode. 3 If problem remains, change the pH electrode.
88 Electrical error Call for service	1 Factory calibration for pH is lost. The monitor can still be used but may not meet specifications for pH measurement. 2 Call for service.
89 Electrical error Call for service	1 Factory calibration for conductivity is lost. The monitor can still be used but may not meet specifications for conductivity measurement. 2 Call for service.
90 ATTENTION set<=0mV first	1 Önly visible to service personnel.
91 WARNING bad pH ad value	
92 WARNING electrode slope <70 or >110%	1 Electrode slope is out of range. Check buffers and recalibrate. 2 Clean the pH electrode and recalibrate. 3 If the message remains, replace the pH electrode.
93 pH_cal failed check electrode	
94 WARNING<1pH unit between cal_buff 1&2	1 The difference between the pH of the buffers used during calibration must be at least 1 pH unit.
95 Temp cal failed check cond cell	1 Check that the conductivity cell is connected. Recalibrate. 2 The measured temperature value differs from the reference value by more than ±5 °C, or actual temperature is lower than -8 °C. Recalibrate. 3 If the message remains, replace the conductivity cell.
97 WARNING pH scale (0-100%)<1pH unit	1 The difference between the zero level and full scale must be at least 1 pH unit. Increase the span between zero and full scale settings. See section 3.8.

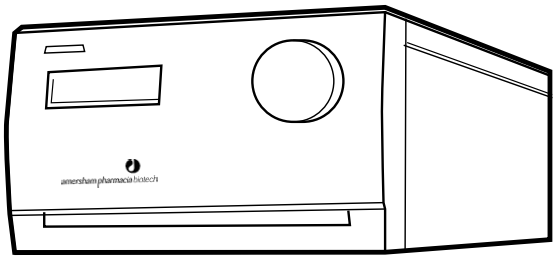
98 Cal failed. Cell constant not 0.1-300	1 Conductivity cell constant is out of range. 2 Wrong solution used during calibration. Use 1.00 M NaCl and recalibrate. 3 Air in conductivity cell during calibration. Flush the flow cell with calibration solution and recalibrate. 4 Dirty conductivity cell. Clean the flow cell and recalibrate. 5 If the problem remains, change the conductivity cell.
ERROR key (OK) **ERROR key (Esc)** **ERROR key (OK+Esc)**	1 The key was pressed during self-test, or is faulty. 2 Switch off the module. 3 Switch on the module.
ERROR number 100 **ERROR number 109-113** **ERROR number 120-121**	1 Switch off the module. 2 Check all connections. 3 Switch on the module.
ERROR number 106-108 **ERROR number 118**	1 Switch off the module. 2 Check all UniNet 1 and UniNet 2 connections. 3 Switch on the module.
Exc x/y in ab.c **Exc DIV/O in ab.c** **Exc instr in ab.c** **Exc address in ab.c**	1 Switch off the module. 2 Check all connections. 3 Switch on the module.

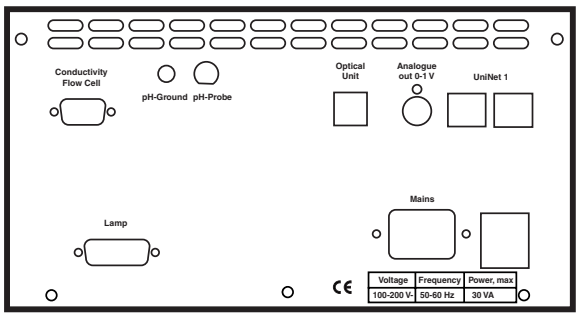
Reference information

A Description

A.1 Module

Monitor UPC-900 is an on-line monitor for measurement of UV-absorption, pH (optional in ÄKTAFPLC) and conductivity. The monitor can work with standard glass pH electrodes with a built in liquid-filled reference electrode and a BNC connector.





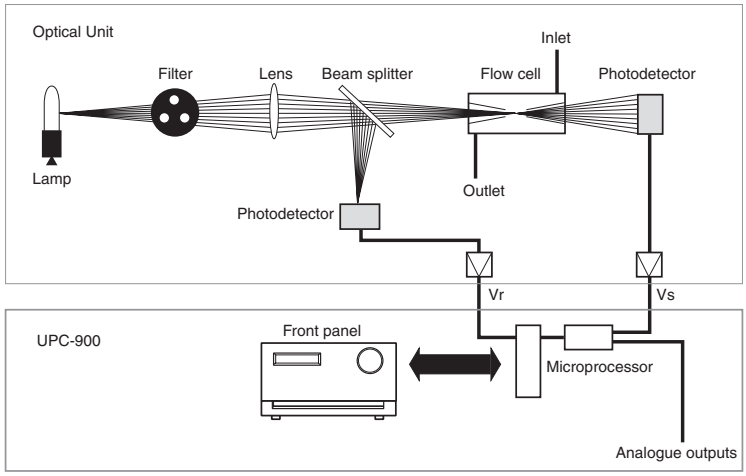
Conductivity measurement has a dynamic range from 1 µS to 999.9 mS/cm and is suitable for a wide range of applications.

Connector/switch	*Function*
UniNet 1	Computer network
Analogue out 0-1 V	Chart recorder outputs, 3 channels 0–1.0 V
Optical Unit	UV signal from optical unit
pH-Probe	Connection to pH electrode, standard BNC socket
pH-Ground	Reference ground for pH measurement
Conductivity Flow Cell	Connection to conductivity flow cell, 9 pole D-sub connector
Lamp	UV lamp connection
Mains	Supply voltage, grounded
0/1	Mains supply on/off switch

The module contains no internal user-replaceable items.

A.2 UV optical unit

The UV optical unit houses the lamp (Zn or Hg), the wavelength filter and the UV flow cell. The light beam is directed through a double conical or straight flow-through cuvette (6 ml or 2 ml illuminated volume) to a photodetector. The photodetector current is fed to the signal processing circuitry in the module.



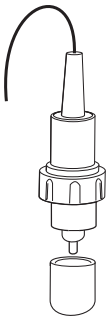
The reference signal comes from the same point in the lamp as the signal measuring the sample, thus assuring a stable baseline by eliminating the effects of variations in lamp intensity.

The Hg lamp emits light only at certain wavelengths. It does not emit light at 280 nm, so for this wavelength, the light is converted at a fluorescent surface before it passes the filter. On the lamp housing, there is a special exit for 280 nm light, which means that the lamp position needs to be changed when working with this wavelength.

For 214 nm wavelength, a Zn lamp is used. This lamp is larger than the Hg lamp and is therefore mounted in a larger lamp housing.

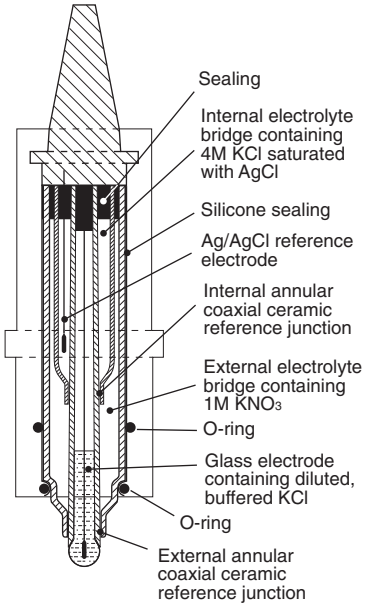
The lamp connectors are keyed to inform the monitor software of which lamp type is connected.

A.3 pH electrode

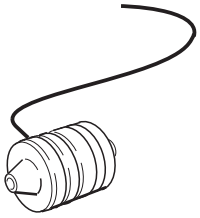


The pH electrode is of the sealed combination double junction type. It contains a sealed Ag/AgCl reference which cannot be refilled, an internal electrolyte bridge of 4 M KCl saturated with Ag/AgCl, an outer electrolyte bridge of 1 M KNO_3, an annular ceramic reference junction and a low profile pH membrane. The pH electrode is delivered with a transparent cover.

The flow cell should not be used with any other pH electrode.

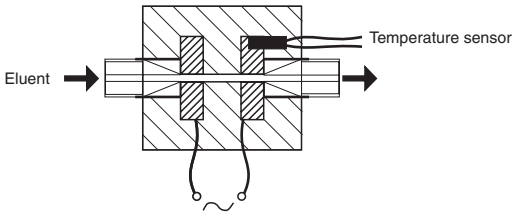


A.4 Conductivity cell



The flow cell has two cylindrical titanium electrodes positioned in the flow path of the cell. An alternating voltage is applied between the electrodes and the resulting current is measured and used to calculate the conductivity of the eluent. The monitor controls the AC frequency and increases it with increasing conductivity between 50 Hz and 50 kHz giving maximum linearity and true conductivity values.

The conductivity is automatically calculated by multiplying the measured conductance by the flow cell's cell constant. The cell constant is pre-calibrated on delivery but can be measured with a separate calibration procedure. This procedure is described in *Reference information* section *B.2*.



One of the electrodes has a small temperature sensor for measuring the temperature of the eluent in the flow cell. Temperature variations influence the conductivity and, in some applications when highly precise conductivity values are required, it is possible to program a temperature compensation factor that recalculates the conductivity to a set reference temperature.

B Menus

B.1 Check menus

B.1.1 Checking autozero level

The module internal absorbance value for autozero can be checked to test the consistency of buffers.

Check Autozero
AZ 0.00006 AU

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

2 Select sub menu **Check Autozero**. The autozero absorbance value for the used wavelength is shown.

B.1.2 Checking lamp run time

The lamp run time can be checked to determine the need for lamp replacement. Run times for both Hg and Zn lamps are monitored.

Check Lamp Run Time
Hg 2300h Zn 340h

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

2 Select sub menu **Check Lamp Run Time**.

B.1.3 Checking lamp intensity

The lamp intensity can be checked to determine the status of the lamp used.

Check Lamp Intensity

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

2 Select sub menu **Check Lamp Intensity**.

B.1.4 Checking recorder

The function of a connected chart recorder can be tested.

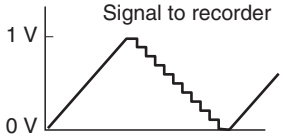
1 Select menu **Check** and press **OK**.

2 Select sub menu **Check Recorder** and press **OK**.

Check Recorder

3 Start the test by selecting **on** and press **OK**.

Check Recorder
Please wait!



The test will ramp the signal on each channel up to 1 V and then decrease the signal in 10% steps back to 0 V. The test is run continuously. Compare the diagram of the chart recorder with the figure.

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

B.1.5 Checking service mode

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

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

Check Service Mode

2 Select sub menu **Check Service Mode** and press **OK**.

Telephone Service: 012345678901	3 The service telephone number is displayed, press **OK.**
Contract Number: 012345678901	4 The service contract number is displayed, press **OK**.
Serial Number: 01234567 YM 012345	5 The module serial number is displayed, press **OK**.
UPC-900 V1.00	6 The module name and software version are displayed, press **OK**.
Date of Maintenance: ?	7 The date of the last service is displayed, press **OK**.
Buzzer Test	8 A test of the module's buzzer is performed, press **OK**.

B.2 Setup menus

B.2.1 Selecting setup sub menus

There are setup sub menus for temperature, conductivity, pH and UV absorbance.

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

Setup
Temp Cond pH UV

2 Select sub menus **Temp, Cond, pH** or **UV** with the dial and press **OK**.

B.2.2 Setup adjust temperature

Calibration of the temperature sensor in the conductivity flow cell is only necessary if the monitor is used in high accuracy measurement or if the conductivity flow cell is replaced.

1 Place the flow cell together with a precision thermometer inside a box or empty beaker to ensure that they are not exposed to draught. Leave them for 15 minutes to let the temperature stabilise.

2 Read the temperature on the thermometer.

Setup
Temp Cond pH UV

3 Select sub menu **Setup Temp** and press **OK**.

Setup Adjust Temp
(22.3°C)

4 Select sub menu **Setup Adjust Temp** and press **OK.** The current temperature is shown.

Warning! This will
change temp calib

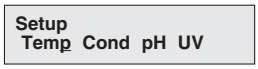
5 A warning message is shown until confirmed by pressing **OK**.

Setup Adjust Temp
(22.3°C) 22.3

6 The current adjustment value is displayed as default. Enter the temperature shown on the thermometer and press **OK**.

B.2.3 Setup show temperature

The display of the temperature in the conductivity flow cell, shown in the main operating menu 2, can be enabled or disabled.



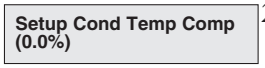
1 Select sub menu **Setup Temp** and press **OK**.

2 Select sub menu **Setup Show Temp** and press **OK.** The current status for showing temperature is displayed. If **on** is shown, current temperature is displayed in main menu 1 and 2. If **off** is shown, no temperature is displayed in the main menus.

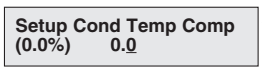
3 Change the setting if desired and press **OK**.

B.2.4 Setup conductivity temperature compensation

1 Select sub menu **Setup Cond** and press **OK**.

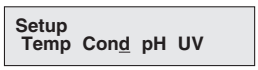


2 Select sub menu **Setup Cond Temp Comp** and press **OK**. The current temperature compensation factor is shown. 0.0% means that the compensation is off (default setting). The range is 0.0-9.9%.

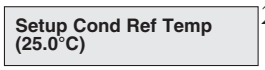


3 The current compensation factor is displayed as default. Adjust the compensation factor setting as necessary and press **OK**.

B.2.5 Setup conductivity reference temperature



1 Select sub menu **Setup Cond** and press **OK**.



2 Select sub menu **Setup Cond Ref Temp** and press **OK**. The current reference temperature value is shown. 25.0 °C is the default setting. The range is 0.00-99.9 °C.

3 The current reference temperature value is displayed as default. Adjust the reference temperature value setting as necessary and press **OK**.

B.2.6 Setup adjust conductivity

Normally it is not necessary to adjust the cell constant as the flow cell is pre-calibrated on delivery. Adjustment is only necessary when replacing the conductivity flow cell with a flow cell whose cell constant is unknown. We recommend that the conductivity flow cell is recalibrated after cleaning. When adjusting the cell constant from UNICORN, select **System Control:System:Calibrate** and then select **CondCalib**.

Note: *The conductivity temperature compensation must not be used when adjusting the cell constant. Set the* ***Setup Cond Temp Comp*** *to 0 (see section B.2.4). The temperature sensor must be calibrated*

before adjusting the cell constant (see section B.2.2).

1 Prepare a calibration solution of 1.00 M NaCl, 58.44 g/l. Let the solution stand until it is at room temperature. This is important for exact measurements.

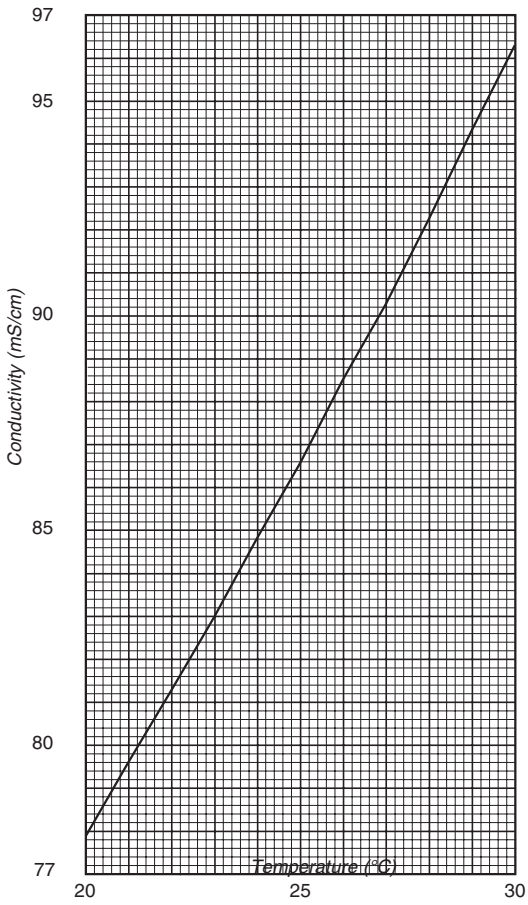
2 Fill the flow cell completely with the calibration solution by pumping at least 15 ml through the cell with a syringe.

3 Stop the flow and wait 15 minutes until the temperature is constant in the range 20–30 °C.

4 Read the conductivity value displayed and compare it with the theoretical value from the graph below at the temperature of the calibration solution. If the displayed value and the theoretical value correspond, no further action is required.

If the values differ, proceed with actions 5–8

Conductivity of 1.00 M NaCl at 20–30 °C

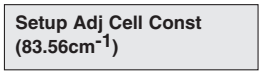


Setup **Temp Cond pH UV**	5 Select sub menu **Setup Cond** and press **OK**.
Setup Adjust Cond **(1.000mS/cm)**	6 Select sub menu **Setup Adjust Cond** and press **OK.** The current conductivity value is shown.
Warning! This will **change cell calib**	7 A warning message is shown until confirmed by pressing **OK**.
Setup Adjust Cond **(83.53mS/cm) 86.6**	8 The current value is displayed as default. Enter the theoretical conductivity value according to the graph and press **OK**. The new cell constant is automatically calculated. The range is 1.000-999.9 mS/cm.

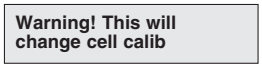
B.2.7 Setup adjust cell constant

After replacing the flow cell, the cell constant has to be set. (The cell constant is shown on the packaging).

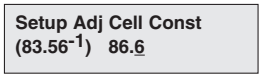
1 Select sub menu **Setup Cond** and press **OK**.



2 Select sub menu **Setup Adjust Cell Const** and press **OK.** The current cell constant is shown.



3 A warning message is shown until confirmed by pressing **OK**.



4 The current cell constant value is displayed as default. Enter the new cell constant as read from the packaging and press **OK**. The range is 0.1-300.0 cm^{-1}.

When entering the cell constant from UNICORN, select **System Control:System:Calibrate** and select **Cond_Cell**.

B.2.8 Setup show conductivity

1 Select sub menu **Setup Cond** and press **OK**.

2 Select sub menu **Setup Show Cond** and press **OK.** The current status for showing conductivity is displayed. If **on** is shown, current conductivity is displayed in main menus 1 and 2. If **off** is shown, no conductivity is displayed in the main menus.

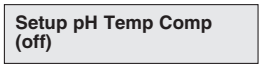
3 Change the setting if desired and press **OK**.

B.2.9 Setup pH temperature compensation

The relationship between pH and the output signal from the pH electrode is temperature dependent. For accurate measurements during temperature changes, the pH measurement can be temperature compensated. In normal applications, when the temperatures of the buffers and calibration buffers are identical, temperature compensation is not necessary.

When using temperature compensation, it is important that the temperature of the pH electrode is the same as that of the conductivity flow cell since that is where the temperature is measured.

1 Select sub menu **Setup pH** and press **OK**.



2 Select sub menu **Setup pH Temp Comp** and press **OK.** The current status for showing pH is displayed. If **on** is shown, T_C is displayed in main menus 1 and 2. If **off** is shown (default), T_C is not displayed in the main menus.

3 Change the setting if desired and press **OK**.

B.2.10 Setup show pH

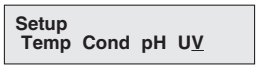
Normally the pH is displayed in the main operating menus (see section *3.13 Reading pH, temperature and conductivity values*). If not required, the pH display can be set to off.

1 Select sub menu **Setup pH** and press **OK**.

2 Select sub menu **Setup Show pH** and press **OK.** The current status for showing pH is displayed. If **on** is shown, current pH is displayed in main menus 1 and 2. If **off** is shown, no pH is displayed in the main menus.

3 Change the setting if desired and press **OK**.

B.2.11 Setup UV averaging filter constant



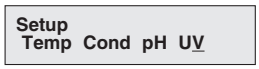
1 Select sub menu **Setup UV** and press **OK**.

2 Select sub menu **Set Averaging** and press **OK.** The current filter constant is shown. Valid values are 0.02, 0.04, 0.08, 0.16, 0.32, 0.64, 1.3, 2.6, 5 and 10 seconds.

3 Change the setting if desired and press **OK**.

B.2.12 Setup lamp run time

When the UV lamp is replaced, the **Lamp Run Time** counter should be reset.



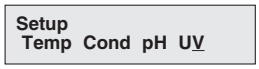
1 Select sub menu **Setup UV** and press **OK**.

2 Select sub menu **Setup Lamp Run Time** and press **OK**.

3 Set the **Lamp Run Time** counter to zero with the dial. Press **OK** to acknowledge.

B.2.13 Setup show UV

Normally UV absorbance is displayed in main menu 1. If not required, the UV absorbance display can be set to off.



1 Select sub menu **Setup UV** and press **OK**.

2 Select sub menu **Setup show UV** and press **OK.** The current status for showing UV is displayed. If **on** is shown, the current UV value is displayed in main menu 1. If **off** is shown, no UV value is displayed in main menu 1.

3 Change the setting if desired and press **OK**.

B.2.14 Setup language

The language used on the display can be changed.

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

Setup Language
(GB) GB D E F I

2 Select sub menu **Setup Language** and press **OK**.

3 Select the desired language.
GB = English
D = German
F = French
E = Spanish
I = Italian

B.2.13 Setup unit number

The unit number is the identification the Monitor UPC-900 has on the UniNet-bus. It should correspond to the number set in UNICORN for the Monitor UPC-900. The number should be set to 0 if one UPC-900 is used. If more than one UPC-900 monitor is used, they must all have different numbers.

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

Setup Unit Number

2 Select sub menu **Setup Unit Number** and press **OK**.

3 Select unit number (0–25) and press **OK**.

B.2.14 Setup display angle

Sets the display angle to compensate for different viewing heights.

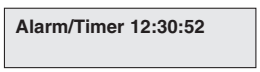
1 Select main menu **Setup** and press **OK**.

2 Select sub menu **Set Display Angle** and press **OK**.

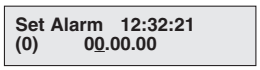
3 Select viewing angle (->\ Up, ->| Mid or ->/ Down).

B.3 Setting and using the alarm timer

You can set the alarm function to either a fixed alarm time or use a count-down timer. You cannot set both an alarm time and the count-down timer. The default or current value is shown in parentheses.

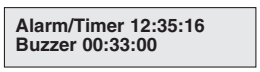


1 Select main menu **Alarm/Timer**. The display shows current time.

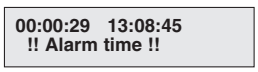


2 Press **OK**. Turn the dial one step clockwise to select sub menu **Set Alarm** if you want to set an alarm at a fixed time. Press **OK** to enter the time in the form **HH.MM.SS** and press **OK** again after entering each time unit.

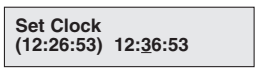
3 If you want to set a count-down time, turn the dial one step further to select sub menu **Set Timer**. Press **OK** to enter the count-down value in the form **HH.MM.SS** and press **OK** again after entering each time unit.



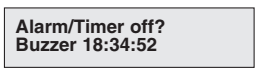
4 Press **ESC** to return to the **Alarm/Timer** menu, which now shows the set alarm time or count-down time as **BzzHH.MM.SS**.



5 When the alarm time is due or the count-down timer reaches 00:00:00, an alert display is shown and the module beeps for 30 s, or until **OK** is pressed. The display shows the time elapsed since the alarm and the current time. A second alert display is shown until **OK** is pressed.



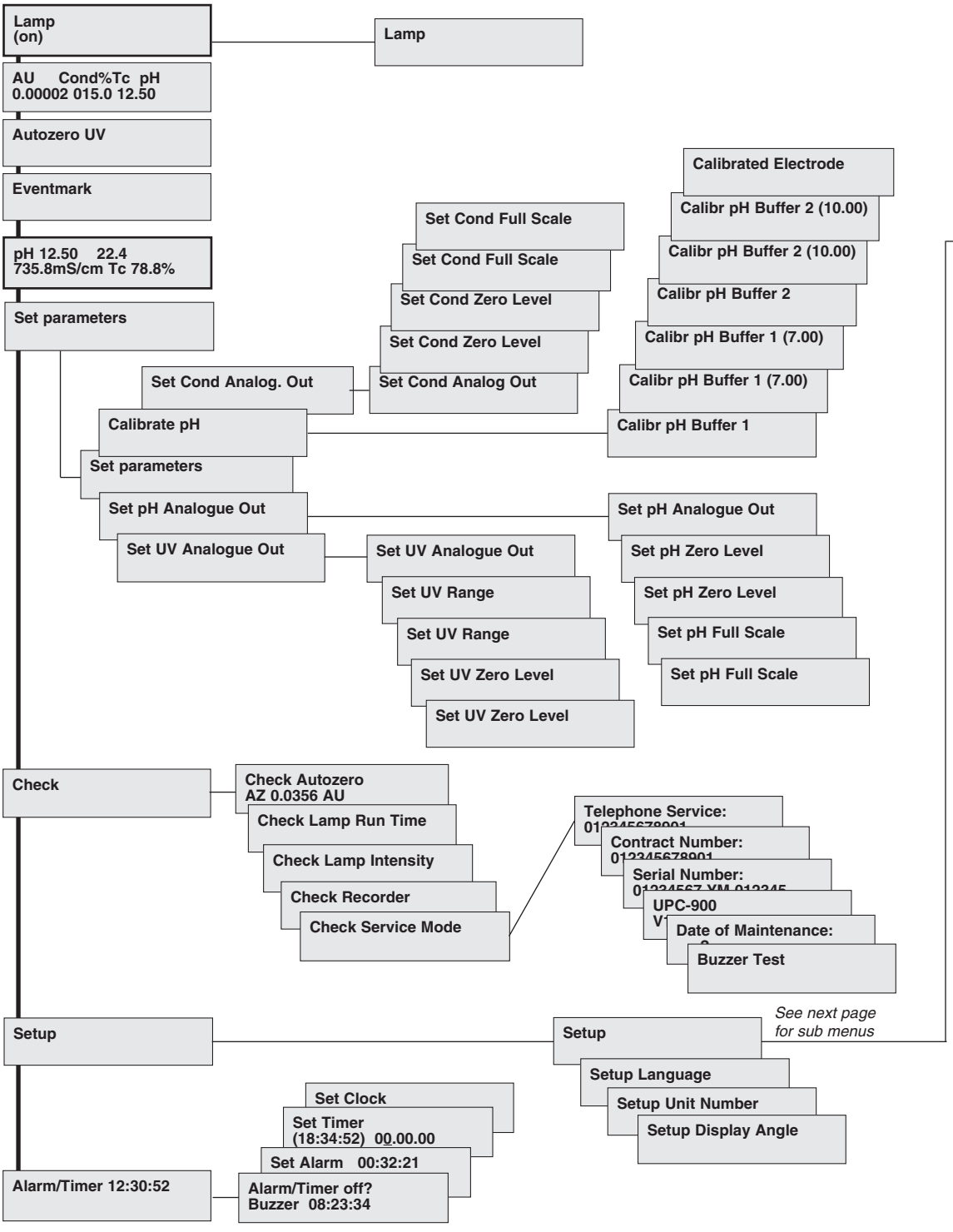
The alarm timer is based on the internal module clock, which can be set in the **Set Clock** menu located after the **Set Timer** menu. The clock will be reset when power is turned off.

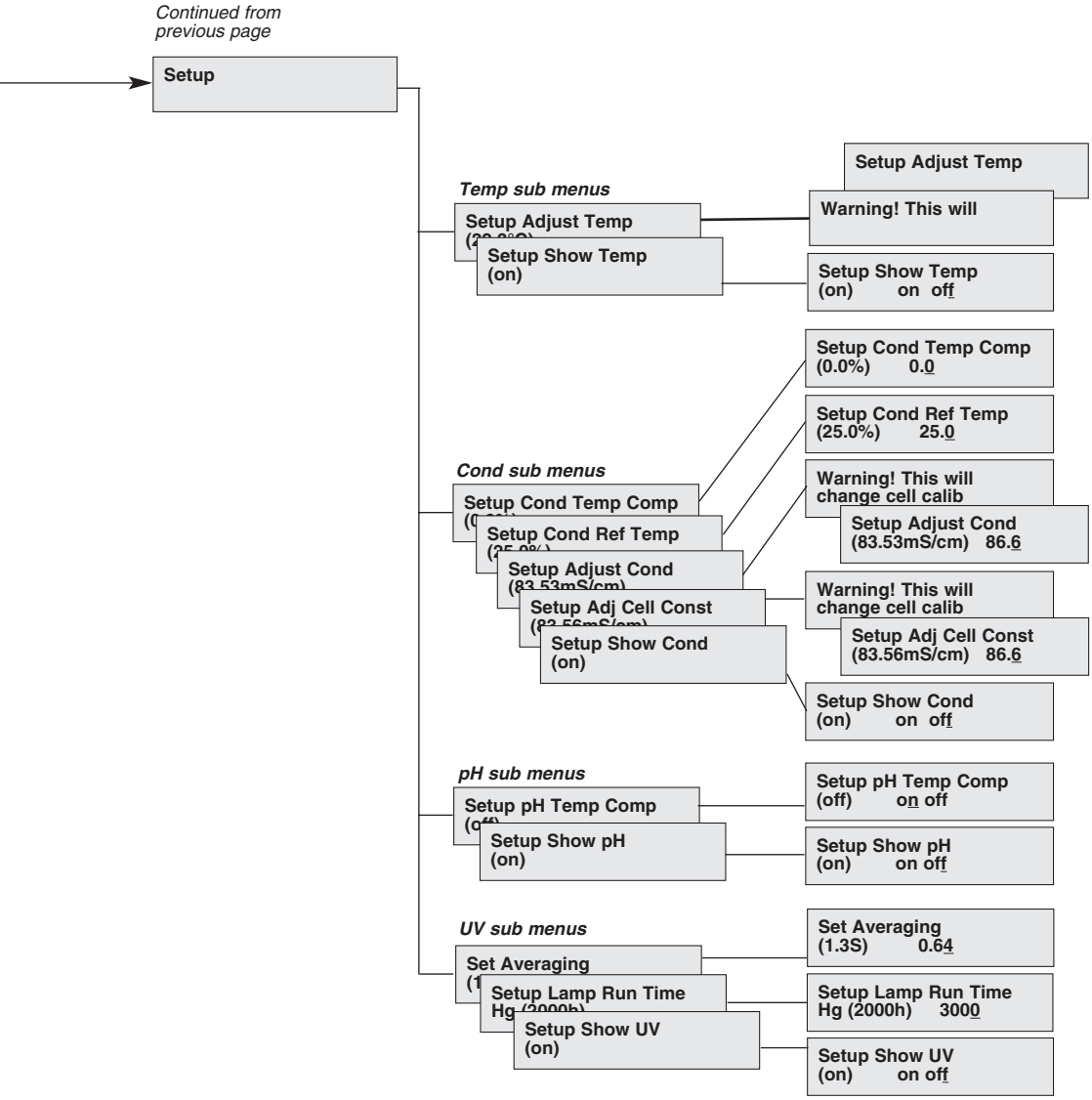


A set alarm/timer function can be reset by pressing **OK** in the menu **Alarm/Timer off?**

B.4 Service displays

The module has service displays for use by authorised service personnel. If the service display **Insert Access Code:** is accidentally selected, press **ESC** to return to the normal operation display.


Lamp (on)
Lamp
AU Cond%Tc pH 0.00002 015.0 12.50
Autozero UV
Eventmark
pH 12.50 22.4 735.8mS/cm Tc 78.8%
Set parameters
Set Cond Full Scale
Set Cond Full Scale
Set Cond Zero Level
Set Cond Zero Level
Set Cond Analog. Out
Set Cond Analog Out
Calibrated Electrode
Calibr pH Buffer 2 (10.00)
Calibr pH Buffer 2 (10.00)
Calibr pH Buffer 2
Calibr pH Buffer 1 (7.00)
Calibr pH Buffer 1 (7.00)
Calibrate pH
Calibr pH Buffer 1
Set parameters
Set pH Analogue Out
Set pH Analogue Out
Set UV Analogue Out
Set UV Analogue Out
Set pH Zero Level
Set UV Range
Set pH Zero Level
Set UV Range
Set pH Full Scale
Set UV Zero Level
Set pH Full Scale
Set UV Zero Level
Check
Check Autozero AZ 0.0356 AU
Check Lamp Run Time
Check Lamp Intensity
Check Recorder
Check Service Mode
Telephone Service:
Contract Number:
Serial Number:
UPC-900
Date of Maintenance:
Buzzer Test
Setup
Setup
See next page for sub menus
Setup Language
Setup Unit Number
Setup Display Angle
Set Clock
Set Timer (18:34:52) 00.00.00
Set Alarm 00:32:21
Alarm/Timer 12:30:52
Alarm/Timer off? Buzzer 08:23:34


Continued from previous page
Setup
Temp sub menus
Setup Adjust Temp
Setup Show Temp (on)
Setup Adjust Temp
Warning! This will
Setup Show Temp (on) on off
Cond sub menus
Setup Cond Temp Comp
Setup Cond Ref Temp
Setup Adjust Cond (83.53mS/cm)
Setup Adj Cell Const
Setup Show Cond (on)
Setup Cond Temp Comp (0.0%) 0.0
Setup Cond Ref Temp (25.0%) 25.0
Warning! This will change cell calib
Setup Adjust Cond (83.53mS/cm) 86.6
Warning! This will change cell calib
Setup Adj Cell Const (83.56mS/cm) 86.6
Setup Show Cond (on) on off
pH sub menus
Setup pH Temp Comp
Setup Show pH (on)
Setup pH Temp Comp (off) on off
Setup Show pH (on) on off
UV sub menus
Set Averaging
Setup Lamp Run Time Hg (2000h)
Setup Show UV (on)
Set Averaging (1.3S) 0.64
Setup Lamp Run Time Hg (2000h) 3000
Setup Show UV (on) on off

B.5 Menu overview

B.5 Menu overview (continued)

C Technical specifications

The full specifications apply only after at least 1 hour warm-up.

C.1 Operating data

UV measurement

Absorbance range	0.01 - 5 AU (full scale)
Autozero range	-0.2 - 2.0 AU
Baseline adjust	Adjustable 0-100% of full scale
Wavelengths	
Hg lamp, fixed	254 and 280
by changing filter	313, 365, 405, 436 and 546 nm
Zn lamp	214 nm
Linearity, 5 mm cell	<3% up to 2 AU at 254 nm <5% up to 1 AU at 280 nm
Static noise	
short term	40×10^{-6} AU at 254 nm (typically 6×10^{-6} AU at 254 nm)
long term	40×10^{-6} AU at 254 nm
Static drift	$\pm 100 \times 10^{-6}$ AU/h at 254 nm
Flow sensitivity	2×10^{-4} AU min/ml

UV flow cell, 2 mm

Flow rate	0 - 100 ml/min
Max. pressure	4 MPa (40 bar, 580 psi)
Back-pressure	Max. 0.05 MPa at 100 ml/min
Liquid temperature range	+4 to +60 °C
Optical pathlength	2 mm
Cell volume	2 µl (30 µl detector volume)

UV flow cell, 5 mm

Flow rate	0 - 20 ml/min
Max. pressure	4 MPa (40 bar, 580 psi)
Back-pressure	Max. 0.02 MPa at 20 ml/min
Liquid temperature range	+4 to +60 °C
Optical pathlength	5 mm
Cell volume	6 µl (10 µl detector volume)

Conductivity measurement

Conductivity range	1 µS/cm to 999.9 mS/cm
Reproducibility	
short term	Max. ±1% or ±5 µS/cm whichever is greater
long term	Max. ±3% or ±15 µS/cm whichever is greater
Noise	Max. ±0.5% of full scale calibrated range
Response time	<3 s (0 - 95% of step)

Temperature sensor	
accuracy	±2.0 °C
drift	±0.5 °C per 10 h
Flow rate sensitivity	±1% within 0–100 ml/min

Conductivity flow cell

Flow rate	0 - 100 ml/min
Max. pressure	5 MPa (50 bar, 725 psi)
Generated backpressure	Max. 0.01 MPa at 100 ml/min

pH measurement

pH range	0 to 14 (spec. valid between 2 and 12)
Accuracy	
temperature compensated	±0.1 pH within +4 to +40 °C
not temperature	

compensated	±0.2 pH within +15 to +25 °C ±0.5 pH within +4 to +15 °C and +25 to +40 °C
Response time	<10 s (0 - 95% of step)
Long term stability	Dev. max. 0.02 pH/h (measured at pH 4.0)
Flow rate sensitivity	Dev. max. 0.1 pH units
pH flow cell	
Flow rate	0.1 - 100 ml/min
Max. pressure	0.5 MPa (5 bar, 72 psi)
Generated back-pressure	Max. 0.02 MPa (0.2 bar, 2.9 psi) at 100 ml/min

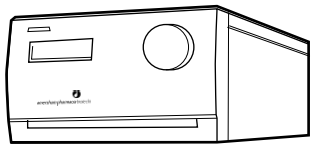
C.2 Physical data

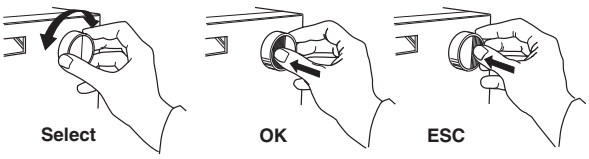
Control	Stand-alone or from a PC with UNICORN version 3.0 or higher, via UniNet 1 connection.
Degree of protection	
housing	IP 20, IP 21
flow cells	IP 44
Power requirements	100–240 V AC, 50–60 Hz
Power consumption	25 VA
Functions	Languages available; English, German, Spanish, French, Italian
UV lamp cable length	1.5 m, AMP 5+2 pole connector
Optical unit, cable length	1.5 m, RJ-45 connector
pH electrode cable length	1.5 m, BNC connector
Cond. cell cable length	1.5 m, D-sub 9 pole connector
Tubing connections	Fingertight connector, 1/16”
Analogue outputs	0–1 V full scale
Display	2 rows with 20 characters each
Dimensions, H x W x D	100 x 260 x 370 mm
Weight	8.5 kg
Wetted materials	
UV flow cells	Quartz, ETFE, titanium
cond. flow cell	Titanium, CTFE
pH electrode	Glass, FFKM (perfluororubber)
flow cell	titanium
dummy electrode	PTFE (polytetrafluoroethylene)
pH stability range	1–13 (1–14, <1 day exposure)
Typical lamp lifetime	
Hg, 254 nm, room temperatur	7000 hours
coold room	2000 hours
Hg, 280 nm, room temperature	3500 hours
Zn, 214 nm, room temperature	2000 hours
Chemical resistance	The wetted parts are resistant to organic solvents and salt buffers commonly used in chromatography of biomolecules, except 100% ethylacetate, 100% hexane, and 100 % tetrahydrofuran (THF).

December 2002

Short instructions

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





1 **Switch on the module** with the mains switch on the rear panel.

2 Switch the lamp **on/off**

3 The **main operating menu** displays measured parameters.

4 **Autozero** the module by pressing **OK**.

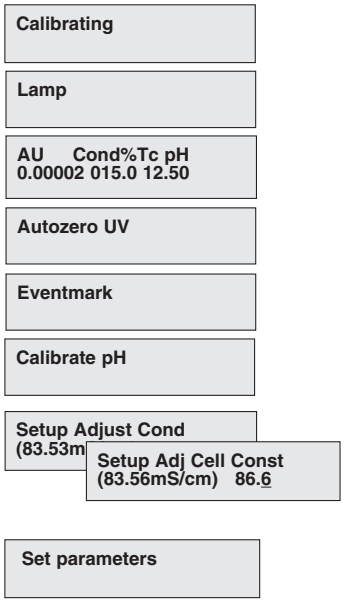
5 Set **Eventmark** by pressing **OK**.

6 **Calibrate** the pH electrode before use and/or daily by using 2 buffers with known pH values.

7 The conductivity flow cell does not normally need to be calibrated.

8 To connect a chart recorder, set the **Analogue output scaling**, 0 - 100%, for measured parameters.

9 Always store the pH electrode in a 1:1 mixture of pH 4 buffer and 2 M KNO_3 when not in use.



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TC information ab, Uppsala. Printed in Sweden by TK i Uppsala AB