

# LIGHTWAVE II & LIGHTWAVE II<sup>+</sup>

# **USER MANUAL**









# **Declaration of Conformity**

This is to certify that the Lightwave II and Lightwave II<sup>†</sup> UV/Visible Spectrophotometers part numbers: 80-3003-72 / -73 / -74 & 80-3004-60 / -61 / -62 manufactured by Biochrom Ltd. conform to the requirements of the following Directives-: 73/23/EEC & 89/336/EEC & IVD

Standards to which conformity is declared

EN 61010-1: 2001 Safety requirements for electrical equipment for measurement, control and laboratory use.

EN 61326-2.3: 1998 Electromagnetic compatibility - generic emission standard Electrical equipment for measurement, control and laboratory use.

EN 61000-4-6: 1992 Electromagnetic compatibility - generic immunity standard part 1. Residential, commercial and light industry.

BS EN 591:2001 Instruction for use for in vitro diagnostic instruments for professional use.

BS EN 13612:2002 Performance evaluation of in vitro diagnostic medical devices

2002/96/EC This appliance is marked according to the European directive 2002/96/EC on Waste Electrical and Electronic Equipment (WEEE). By ensuring this product is disposed of correctly, you will help prevent potential negative consequences for the environment and human health, which could otherwise be caused by inappropriate waste handling of this product.

The symbol 📈 on the product, or on the documents accompanying the product, indicates that this appliance may not be treated as household waste. Instead it shall be handed over to the applicable collection point for the recycling of electrical and electronic equipment. Disposal must be carried out in accordance with local environmental regulations for waste disposal.

Signed:

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### **ESSENTIAL SAFETY NOTES**

There are a number of warning labels and symbols on your instrument. These are there to inform you where potential danger exists or particular caution is required. Before commencing installation, please take time to familiarise yourself with these symbols and their meaning.



Caution (refer to accompanying documents). Background colour yellow, symbol and outline black.

# Unpacking, Positioning and Installation

- Check the contents of the pack against the packing list. If any shortages are discovered, inform your supplier immediately.
- Inspect the instrument for any signs of damage caused in transit. If any damage is discovered, inform your supplier immediately.
- Ensure your proposed installation site conforms to the environmental conditions for safe operation: Indoor use only.
  - Temperature range 5°C to 35°C. Note that if you use the instrument in a room subjected to extremes of temperature change during the day, it may be necessary to recalibrate (by switching off and then on again) once thermal equilibrium has been established (2-3 hours).
  - Maximum relative humidity of 80% up to 31°C decreasing linearly to 50% at 40°C
- The instrument must be placed on a stable, level bench or table that can take its weight (< 4.5 kg) so that air can circulate freely around the instrument.
- This equipment must be connected to the power supply with the power cord supplied. It can be used on 90 240 V, 50-60 Hz supplies.
- If the instrument has just been unpacked or has been stored in a cold environment, it should be allowed to come to thermal equilibrium for 2-3 hours in the laboratory before switching. This will prevent calibration failure as a result of internal condensation.
- Switch on the instrument via the keypad ( ) after it has been plugged in. The instrument will perform a series of self-diagnostic checks.
- Please read through this user manual prior to use.
- Please contact your original supplier in the first instance if you experience technical or sample handling difficulties.

If this equipment is used in a manner not specified or in environmental conditions not appropriate for safe operation, the protection provided by the equipment may be impaired and instrument warranty withdrawn.

# INTRODUCTION

# Your spectrophotometer

Your spectrophotometer is a simple-to-use UV/Visible instrument with a CCD array detector (1024 pixels). It has no moving parts, which is the basis of the rapid scanning operating system. The look and operation of the Lightwave II and II<sup>+</sup> are identical; the only difference between them is the bandwidth. Throughout the rest of this manual the term Lightwave II will be used to cover both instruments.

The user interface is built around folders which are displayed on the home page when the instrument is switched on. After switch on and calibration, the default home page is "Lightwave II" offering the choice of

Applications General spectroscopic methods

Favourites A folder to store your more frequently used configured methods

Methods Contains nine folders that can store less frequently used configured methods (nine

methods per folder)

Utilities Instrument set up (date, time, language, etc) and games

The instrument is supplied with a program PVC (Print via Computer) on the accompanying CD. When used with a USB cable to connect to a PC onto which the software has been installed, it enables the user to "print through" the PC directly to the printer that is connected to it. The data may also be stored as an Excel spreadsheet, as an EMF graphics file, a comma delimited (csv) data file, a tab delimited (txt) data file or in native PVC format for later access

Alternatively, results may be sent to the PC via a Bluetooth accessory; this can either be supplied pre-installed or is available as an optional accessory if the need for its use arises after installation of the product. PVC works in a similar way.

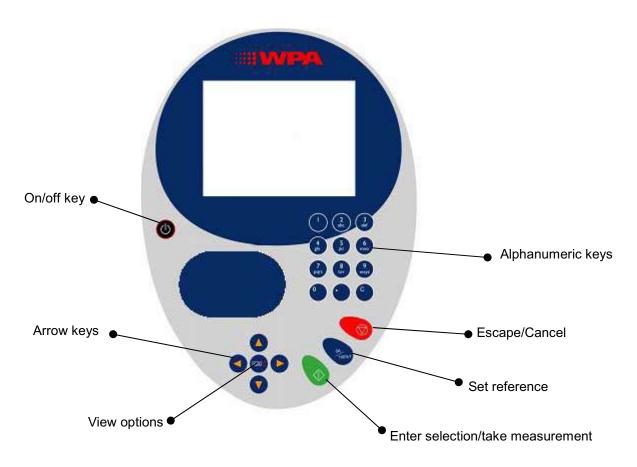
A printer is available for the instrument; this may either be supplied pre-installed or is available as an optional accessory if the need for its use arises after installation of the product.

# Sample handling tips

- Note that the light beam is directed from RIGHT to LEFT through the cell chamber; therefore please ensure the cell is inserted in the correct alignment.
- The cell holder supplied with the instrument accepts standard 10 40mm pathlength quartz, glass or plastic cells.
- The optical height is 15 mm, and the minimum volume that can be used is approx. 70µl in a micro cell.
- 12 mm test tubes may be used (e.g. for cell cultures), however they are not recommended as higher quality
  data is produced by using disposable cuvettes for the analysis. If used, align the indicator line on 12 mm test
  tubes in the same direction to ensure reproducible positioning of the tube. Note that test tubes do not last
  forever, and that the surface becomes scratched and blemished through repetitive use; if this is the case they
  should be replaced.

# Keypad and display

The back-lit liquid crystal display is very easy to navigate around using the alphanumeric entry and navigation arrow keys on the hard wearing, spill proof membrane keypad.



<b>Key</b> On/off key	Action Turns the instrument on/off
Arrow keys	Use the four arrow keys to navigate around the display and select the required setting from the active (highlighted) option.
View Options: 図보던	View options for that application mode. Some of these are common to all applications and described below. Options unique to an application are described in the relevant section.
Alphanumeric keys	Use these to enter parameters and to write text descriptions where appropriate, or required. Use repeated key presses to cycle through lower case, number and upper case. Leave for 1 second before entering next character. Use C button to backspace and 1 to enter a space.
Escape/Cancel: 🕏	Escape from a selection and return to the previous folder. Stop making measurements.
Set Reference: 0A/100%T	Set reference to 0.000 A or 100%T on a reference solution at the current wavelength in the mode selected. When in scan mode, do a reference scan.
Enter: <b>Φ</b>	Enter, or confirm, a selection. Take a measurement.



# **Options** (select using key pad numbers)

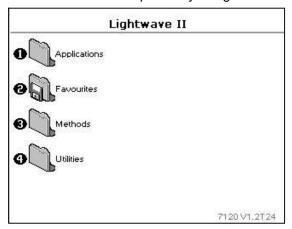
- 1. View parameters for the experiments.
- 2. Print the results.
- 3,4,5,6 Described in the application.
- 7. Define the sample number you wish to start from.
- 8. Save the parameters as a method to a defined folder name with a defined method name.
- 9. Toggle auto-print on/off. Default is off.

Exit options by pressing **②**, or wait.

Experienced operators can use the numeric keys as a shortcut to the option required without needing to enter the Options menu.

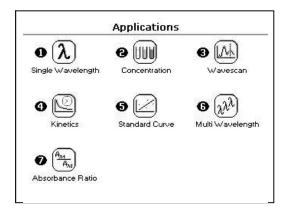
# Software style

The user interface is built around having folders of files which are displayed on the home page when the instrument is switched on. Different folders are numbered and opened by using the associated number key on the keypad.



Summary Function	Keypad numbe	r Description
<b>O</b> Applications	1	Single wavelength, Concentration, Wavelength scan, Kinetics, Standard Curve, Multiple wavelengths and Ratio
<b>@</b> Favourites	2	Saved User selected and configured methods
<b>3</b> ↓ Methods	3	Sub folder selection for User selected and configured methods
<b>4</b> Utilities	4	Instrument set up (date, time, language, etc) and Games

# THE APPLICATIONS FOLDER



# **SUMMARY:**

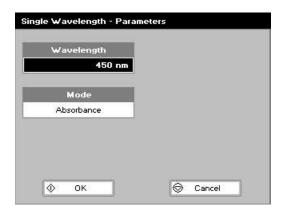
Function	Key pad nur	mber Description
<b>Φ</b> (λ) Single Wavelength	1	Absorbance or %T (transmission) at a single user defined wavelength.
Concentration	2	Concentration measurement at a single wavelength based on a simple Factor entered or calculated from a single standard.
<b>3</b> My wavescan	3	Wavelength scan between two user defined wavelengths. Range 200-950 nm, with user configurable peak finding function.
Kinetics	4	Absorbance versus time measurements either rate or end value based.
Standard Curve	5	Generation of calibration curve by measuring standards at a single wavelength.
Multi Wavelength	6	Absorbance or %T (transmission) at up to 5 user defined wavelengths.
Absorbance Ratio	7	Ratio of absorbance values at two user specified wavelengths.

# **OPTIONS**

Within each application the user has the possibility to select various options that define the way results are treated. If not using a stored method, it is advisable to check that these Options have been appropriately set for your experiment when coming to the instrument. Note that setting the "History" parameter to on (see Preferences later) will cause the instrument to store it's last settings. If the "History" parameter is turned off, all parameters and options will return to their default settings when you leave that application. (Unless it has been saved as a method).

# 1: Single Wavelength - Abs and %T

This makes simple absorbance (A) and % transmission (%T) measurements on samples, measuring the amount of light that has passed through a sample relative to a reference (this can be air). The procedure is as follows:



# Step 1

Set wavelength by using keypad numbers or left and right arrows.

Press the down arrow key.

# Step 2

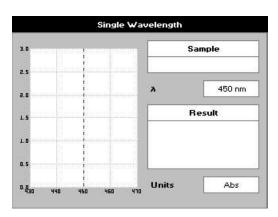
Select the mode, *Absorbance* or *%T*, using the left and right arrows.

# Step 3

To enter the results screen with the selected parameters press OK  $oldsymbol{\Phi}$ 

OR

Cancel the selections and return to the Applications Folder by pressing Cancel  $\mathbf{\hat{Q}}$ .



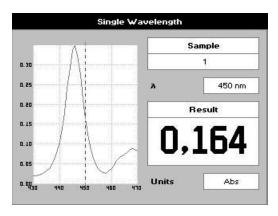
# Step 4

Insert the reference. Press 0A/100% key. This will be used for all subsequent samples until changed.

# Step 5

Insert sample and press **①**.

Repeat step 5 for all samples.



# Results

The result at the selected wavelength is displayed on screen. Use the left and right arrows to move the cursor and display the value at the cursor position (+/- 15nm from set wavelength).

Press Cancel **t** to return to the Applications Folder.

Press ☒☒☑ to display available Options which are described below.



# **Options** (select using key pad numbers)

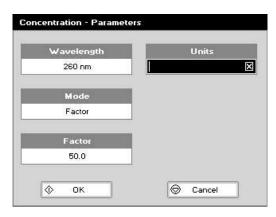
- 1. Return to parameters screen (step 1 above).
- 2. Print result via selected method.
- 3. Toggle between Absorbance and %T mode.
- 4. Print graph greyed out if no data are available.
- 7. Sample number add a prefix to the sample number and reset the incrementing number to the desired value.
- 8. Save method use the left and right arrows to select a folder to store in (Favourites/Methods 1-9), press the down arrow and enter name.
- 9. Auto-print toggles auto-print on/off.

Exit options by pressing **Q**, or wait.

# 2: Concentration

This makes simple concentration measurements on samples, by measuring the amount of light that has passed through a sample relative to a reference (this can be air). Concentration is obtained by multiplying the measured absorbance at a specific wavelength by a factor. The factor may be known in advance, or may be calculated by the instrument by measuring a standard of known concentration.

The procedure is as follows:



# Step 1

Set wavelength by using keypad numbers or left and right arrows.

Press the down arrow key.

# Step 2

Select the mode, Factor (user entered) or Standard (factor is calculated from a calibration sample), using the left and right

Press the down arrow key.

**Step 3** (if Factor is selected)

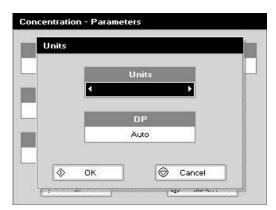
Enter the Factor using the keypad numbers. Range 0.001 to 9999. Use the C button to delete the last digit entered.

Press the down arrow key.

Step 3 (if Standard is selected)

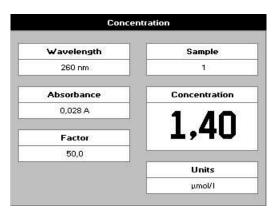
Enter the concentration using keypad numbers. Range 0.01-9999. Use the C button to delete the last digit entered.

Press the down arrow key.



# Step 4

Units: The user can enter a text string up to 8 characters long. To access a list of pre-defined units press the Options key ☒☒☒ and then use the left/right arrows (µg/ml, µg/µl, pmol/µl, mg/dl, mmol/l, µmol/l, g/l, mg/l, µg/l, U/l, %, ppm, ppb, conc or none). These units can also be edited once OK is pressed. This screen also allows the number of displayed decimal points (DP) to be selected, from 0 to 2 Note that the result will always be fixed to 5 significant figures regardless of how many decimal points are selected (so 98768.2 will display as 98768 even with 1 decimal point selected). Press OK  $\Phi$  to store the chosen parameters or Cancel .



To enter the results screen with the selected parameters press

ok 🛇

OR

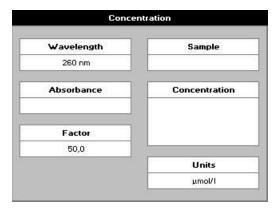
Cancel the selections and return to the Applications Folder by pressing Cancel .

# Step 6 (if using a Factor)

Insert the reference. Press 0A/100% key. This will be used for all subsequent samples until changed.

# Step 7

Insert sample and press  $\mathbf{\Phi}$ .



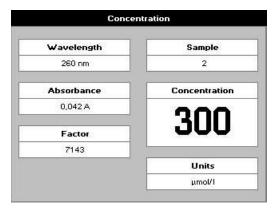
# Step 6 (if using standard mode)

Insert the reference. Press 0A/100% key. This will be used for all subsequent samples until changed.

Press to display the Run Standard screen.

Run the standard by pressing **O**R

Press cancel to return to the measure screen.



# Step 7

Insert the sample and press **O**.

The concentration of the sample is displayed. Results shown as ---- indicate the concentration is out of range.

Repeat step 7 for all samples.

Press **O** to return to the Applications Folder.

Press 🗷 🗷 to display available Options which are described below.



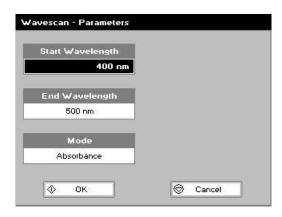
**Options** (select using key pad numbers)

- 1. Return to parameters screen (step 1 above).
- 2. Print result via selected method.
- 3. Toggles on/off, displaying a graph of wavescan +/- 20 nm from selected wavelength.
- 4. Return to Run Standard screen.
- 7. Sample number add a prefix to the sample number and reset the incrementing number to the desired value.
- 8. Save method use the left and right arrows to select a folder to store in (Favourites/Methods 1-9), press the down arrow and enter name.
- 9. Auto-print toggles auto-print on/off.

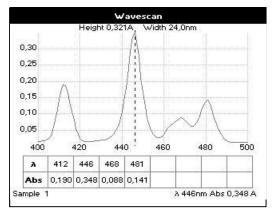
Exit options by pressing **②**, or wait.

# 3: Wavescan

An absorption spectrum can be obtained from your instrument, enabling simple identification of peak height and position. The procedure is as follows:



# 2,5 2,0 1.5 1,0 0,5 0,0 400 420 440 460 480 500 λ Abs λ 450nm mole



# Step 1

Set start wavelength by using keypad numbers or left and right arrows.

Press the down arrow key.

# Step 2

Set end wavelength by using keypad numbers or left and right arrows.

Press the down arrow key.

# Step 3

Select the mode, *Absorbance* or %T, using the left and right arrows.

# Step 4

To enter the measurements screen with the selected parameters press OK  $\Phi$ 

ΟR

Cancel the selections and return to the Applications Folder by pressing Cancel  $\mathbf{\Omega}$ .

# Step 5

Insert the reference. Press 0A/100% key. This will be used for all subsequent samples until changed.

# Step 6

Insert sample and press **①**.

Repeat step 6 for all samples.

# **Results**

A graph of the wavescan is displayed, along with a table of Absorbance/%T at each peak. Use the left and right arrows to move the cursor along the graph. When it reaches a peak the peak height and width of the peak is displayed at the top of the screen.

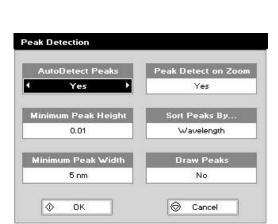
To zoom in on the wavelength scale, use the up arrow. This auto-scales on the Absorbance/%T scale (dependent on the Graph Scale option) and this is retained for subsequent measurements.

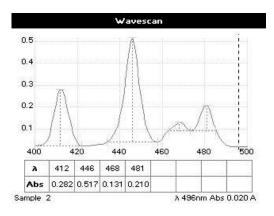
To zoom out again, use the down arrow.

Press to return to the Applications Folder.

Press ☒☒☑ to display available Options which are described next.







**Options** (select using key pad numbers)

- 1. Return to parameters screen (step 1 above).
- 2. Print result via selected method.
- 3. Toggle between Absorbance and %T mode.
- 4. Displays Peak Detection Parameter Screen. See description below.
- Manually adds a peak position to the peak table in the results screen at the position set by the cursor. If the cursor is returned to this position the legend "User Defined Peak" is displayed at the top of the scan and this option changes to Delete Peak...
- Displays Graph Scale Parameter Screen. See description below.
- 7. Sample number add a prefix to the sample number and reset the incrementing number to the desired value.
- 8. Save method use the left and right arrows to select a folder to store in (Favourites/Methods 1-9), press the down arrow and enter name.
- 9. Auto-print toggles auto-print on/off.

Exit options by pressing **②**, or wait.

# Peak Detection (Shortcut button 4)

**AutoDetect Peaks:** Turns on and off the automatic peak detection. The following options determine how peaks are detected:

**Minimum peak height:** Minimum height the peak has to be above the higher of the two adjacent minima for the peak to be detected

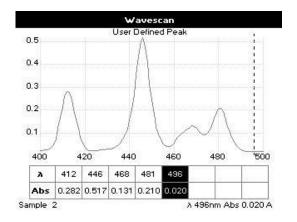
**Minimum peak width:** Minimum width of the peak as determined by the difference in wavelength between the higher of the two adjacent minima and the opposing intersection of that higher minimum level and the peak profile. (See the screen displayed below).

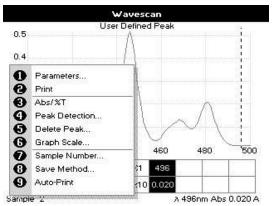
**Peak Detect on Zoom:** Determines whether peaks are reassessed and tabulated when the user zooms into a region of the wavescan. If off leaves the peak detection as determined on the un-zoomed display

**Sort peaks by...:** Determines the sequence that peaks are reported by. Can be wavelength, peak height or peak width.

**Draw Peaks:** Switches display of peak cursors on and off. These show vertical dashed lines displaying the measured peak height and horizontal dashed lines showing the peak width

Pressing Cancel  $\bigcirc$  ignores the selection, pressing  $\diamondsuit$  accepts them.

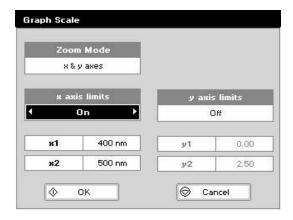




# Add Peak... (Shortcut button 5)

Adds a used defined peak at the current cursor position. The entry is then display in inverse colouring to discriminate between user defined peaks and auto-detect peaks. When the cursor is positioned over the user defined peak a legend "User Defined Peak" appears at the top of the graph. The option then changes to Delete Peak to enable the user to remove the peak.

Note Storing a method at this stage will save these user defined wavelengths, each time method is run Absorbance value at these wavelengths is reported



# Graph Scale...

This enables the user to set up a defined graph by defining the limits in either or both of the x and y axes.

# Zoom mode:

This sets up the operation of the Zoom keys (up and down arrows). "x & y axes" expands the display around the cursor measurement point, whilst the other options select the absorbance or wavelength axes respectively. With x or y axis limits set to on, zooming out will only be permitted to the set limits.

# x/y axis limits:

Setting "x (or y) axis limits" to "On" activates the start and finish points of the desired graph to user defined specific wavelengths and/or absorbance values.

Pressing Cancel  $\bigcirc$  ignores the selection; pressing  $\bigcirc$  accepts them and displays the required graph.

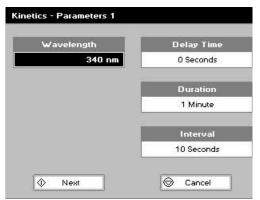
# 4: Simple Kinetics

Kinetics studies, where the change in absorbance needs to be followed as a function of time at a fixed wavelength, can be readily performed.

Reagent test kits are routinely used for the enzymatic determination of compounds in food, beverage and clinical laboratories by measuring NAD / NADH conversion at 340 nm. The change in absorbance over a specified time period can be used to provide useful information when an appropriate factor, defined in the reagent kit protocol, is applied. Reaction rate and enzyme activity can be calculated if the factor used takes account of the absorbance difference per unit time, as opposed to the absorbance difference per se.

For this reason, the change in absorbance per minute (ΔA/min), concentration (ΔA/min x factor) and correlation coefficient (calculated from a best fit of the data points) are displayed. They may not be relevant for simple kinetics experiments.

The procedure to define a new method is as follows:



# **Kinetics Parameter 1 Screen**

Step 1 (Wavelength)

Enter all numerical values using the keypad numbers or the left and right arrows. Use the up and down arrow keys to move between boxes.

Step 2 (Delay time)

Enter the delay time in seconds before measurements are taken. This can be a maximum of 600 seconds (10 minutes).

Step 3 (Duration)

Enter the time in minutes over which measurements are taken. This can be a maximum of 60 minutes.

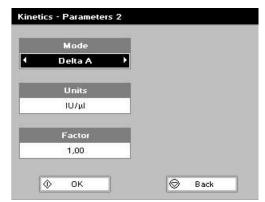
Step 4 (Interval)

Enter the interval time in seconds between measurements using the left and right arrows. Options are: 5, 10, 20, 30 or 60 seconds.

Step 5

Press Next **o** to go to the next parameters screen OR

Press Cancel **t** to return to the Applications Folder.

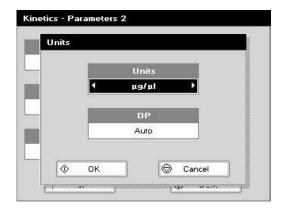


# **Kinetics Parameters 2 Screen Step 6**

Select the measurement mode using the left and right arrows. Delta A: change in absorbance over the measurement duration (or selected period).

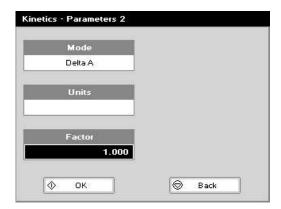
Final A: absorbance at the end of the measurement duration (or selected time).

Slope: rate of change of absorbance over the measurement duration or selected period.



# Step 7 Units:

Units: The user can enter a text string up to 8 characters long. To access a list of pre-defined units press the Options key ΣΣ and then use the left/right arrows (μg/ml, μg/μl, pmol/μl, mg/dl, mmol/l, μmol/l, g/l, mg/l, μg/l, U/l, %, ppm, ppb, conc or none). These units can also be edited once OK is pressed. This screen also allows the number of displayed decimal points (DP) to be selected, from 0 to 2 Note that the result will always be fixed to 5 significant figures regardless of how many decimal points are selected (so 98768.2 will display as 98768 even with 1 decimal point selected). Press OK to store the chosen parameters or Cancel .



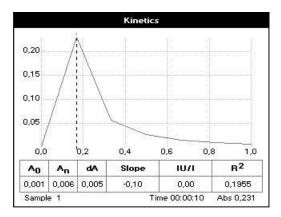
# Step 8

Set the Factor by which the result is multiplied to give the amount in the chosen range using the left and right arrows. Range of 0.01 to 9999.

# Step 9

Press Next **1** to enter the Results screen OR

Press Cancel **1** to return to the Parameters 1 screen.



# **Results**

Insert the reference and press the 0A/100%T key.

Insert the sample and press  $\Phi$  to start the run.

Time (min) is displayed at the bottom of the screen, and absorbance data are plotted on the graph as testing proceeds. The table below the graph gives: absorbance values at  $T_0$  (start of calculation),  $T_n$  (finish of calculation, change in absorbance, slope, regression parameter ( $R^2$ ) of the calculated slope and the result calculated from the selected parameter (dA, final A or slope).

Use the left and right arrows to move the cursor and display the time and absorbance value at measured data points.

Use the up and down arrows to zoom in or out.

Press Cancel **to** return to the Applications Folder.



**Options** (select using key pad numbers)

- 1. Return to parameter 1 screen (step 1 above).
- 2. Print data on the results screen via selected method.
- 3. Print all the data.
- 4. Set the  $t_0$  position (starting point for the slope and dA calculation) at the current cursor position. Value is retained for subsequent samples.
- 5. Set the t<sub>n</sub> position (finishing point for the slope and dA calculation) at the current cursor position. Value is retained for subsequent samples.
- Toggle the calculated slope line on and off.
   Note: if any data points enclosed by t<sub>0</sub> and t<sub>n</sub> are beyond the
   range of the instrument (>2.5A or <0.3A) then this option is
   greyed out.</li>
- 7. Sample number add a prefix to the sample number and reset the incrementing number to the desired value.
- 8. Save method use the left and right arrows to select a folder to store in (Favourites/Methods 1-9), press the down arrow and enter name.
- 9. Auto-print toggles auto-print on/off.

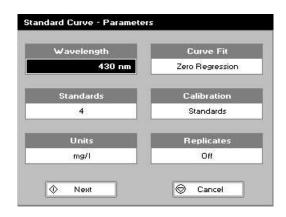
Exit options by pressing **②**, or wait.

# 5: Standard Curve

The construction of a multi-point calibration curve from standards of known concentration to quantify unknown samples is a fundamental use of a spectrophotometer; this instrument has the advantage of being able to store this curve as a method, using up to 9 standards.

To include a zero concentration standard, include this in the number of standards to be entered and enter 0.00 for concentration; use a reagent blank when required to enter the zero standard.

The procedure is as follows:



# Step 1

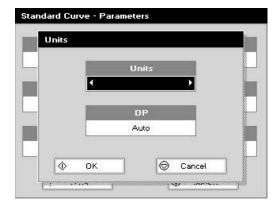
Select the wavelength using the keypad numbers or left and right arrows.

Press the down arrow.

# Step 2

Enter the number of standard concentration points to be used in the curve (1-9).

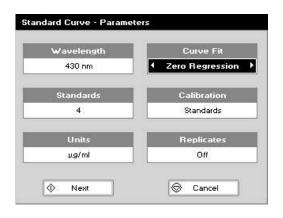
Press the down arrow.



# Step 3

Units: The user can enter a text string up to 8 characters long. To access a list of pre-defined units press the Options key 🗵 🗵 and then use the left/right arrows (μg/ml, μg/μl, pmol/μl, mg/dl, mmol/l, μmol/l, g/l, mg/l, μg/l, U/l, %, ppm, ppb, conc or none). These units can also be edited once OK is pressed.

This screen also allows the number of displayed decimal points (DP) to be selected, from 0 to 2 Note that the result will always be fixed to 5 significant figures regardless of how many decimal points are selected (so 98768.2 will display as 98768 even with 1 decimal point selected). Press OK  $\bullet$  to store the chosen parameters or Cancel  $\bullet$ .



# Step 4

Select the type of curve fit using the left and right arrows. Options: straight line regression, a zero regression (this forces the straight line through the origin), interpolated or cubic spline.

# Step 5

Select the calibration mode: either Standards (measure prepared standards) or Manual (keypad data entry).

Press the down arrow.

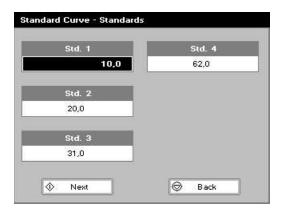
Step 6 (if standards has been selected in step 5)

Select the number of standards to be measured and averaged at each standard concentration point. Can be OFF (1), 2 or 3.

# Step 7

Press Next **t**o enter the Standards screen OR

Press Cancel **to** cancel selections and return to the Applications Folder.



# Standards screen

# Step 8

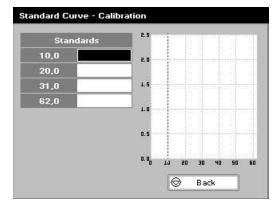
Enter the concentration values by using the keypad numbers and the up and down arrows to move between the different standard boxes. Range 0.001 to 9999.

# Step 9

Press Next igodot to enter the Calibration screen. If any duplicate or non-monotonic (increasing entries) are present the unit will beep and highlight the incorrect entry

OR

Press Back to return to the Parameter screen.



# Calibration Screen (replicates off)

This shows the calibration values and allows standards to be measured.

# Step 10

Insert the reference. Press 0A/100% key.

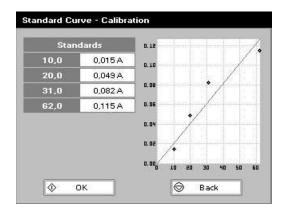
This will be used for all subsequent samples until changed.

# Step 11

Insert the standard (use C to clear previously stored results before measuring).

Press **O** to measure the standard and store the result.

Repeat for all standards.



A graph will display the results and the fitted curve as the measurements are input.

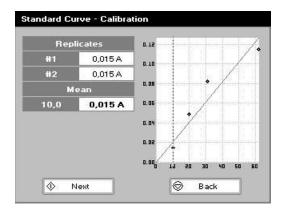
Use the up and down arrows to select a standard to be repeated if a poor reading has been obtained. Use C to clear the previous reading.

# Step 12

Press OK igodot to accept the calibration and go to the Results screen (see below)

OR

Press Back to return to the Standards screen.



# Calibration Screen (replicates on)

This shows the calibration values and allows standards to be measured.

# Step 10

Insert the reference. Press 0A/100% key.

This will be used for all subsequent samples until changed.

#### Step 11

Press  $\bullet$  to display the replicate entry boxes. Use C to clear previously stored results before measuring.

Insert the standard and press Enter to measure the standard and store the result.

Repeat for all replicates and standards.

A graph will display the results and the fitted curve as the measurements are input.

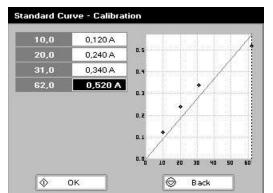
Use the up and down arrows to select a standard to be repeated if a poor reading has been obtained. Use C to clear the previous reading.

# Step 12

Press igodot to accept the calibration and go to the Results screen (see below)

ÒR

Press Back to return to the Standards screen.



# Calibration (Manual entry)

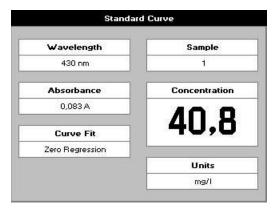
Shows previously entered calibration values and allows values to be entered via the keypad.

The highlighted box can be edited in order to enter an absorbance value corresponding to a given concentration value using the keypad numbers. Range 0.001 to 9999. Use C to backspace and clear the last digit entered and the up and down arrows to move between boxes.

Press OK igodot to accept the calibration and go to the Results screen (see below)

OR

Press Back to return to the Standards screen.



# Results screen

# Step 13

Insert the reference and press the 0A/100%T key. This will be used for all subsequent samples until changed.

# Step 14

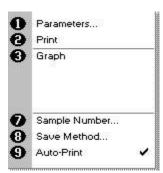
Insert the sample and press  $\Phi$ .

The concentration of the sample is taken and displayed.

Repeat step 14 for all samples.

Press to return to the Applications Folder.

Press ☒☒☑ to display available Options which are described below.



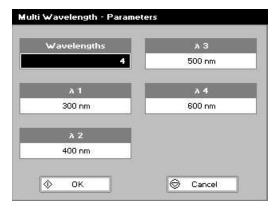
**Options** (select using key pad numbers)

- 1. Return to parameters screen (step 1 above).
- 2. Print result via selected method.
- 3. Toggle graph on/off. Displays calibration graph, cursors give values for last measured sample.
- 7. Sample number add a prefix to the sample number and reset the incrementing number to the desired value.
- 8. Save method use the left and right arrows to select a folder to store in (Favourites/Methods 1-9), press the down arrow and enter name.
- 9. Auto-print toggles auto-print on/off.

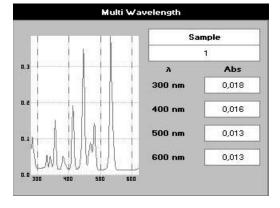
Exit options by pressing  $\mathbf{\Theta}$ , or wait.

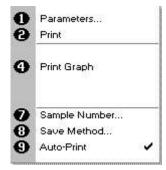
# 6: Multiple Wavelength

This makes up to 5 absorbance measurements on the same sample. The procedure is as follows:



# Multi Wavelength Sample S.S A Abs 300 nm 1.5 400 nm 0.5 0.0 300 400 500 600





# Step 1

Select the number of wavelengths.

Press the down arrow.

# Step 2

Enter the first wavelength using either the number keys or the left and right arrows.

Press the down arrow.

Enter the second wavelength as above and repeat for the number of wavelengths selected (up to 5).

# Step 3

Press OK igodapha to enter the results screen OR

Press Cancel **t** to return to the Applications Folder.

# Step 4

Insert the reference. Press 0A/100% key. This will be used for all subsequent samples until changed.

# Step 5

Insert sample and press �.

Repeat step 5 for all samples.

# Results

A scan plot covering the range of wavelengths selected (with cursors at the relevant wavelengths) and a table of values is displayed.

Press to return to the Applications Folder.

Press ☒☒☑ to display available Options which are described below.

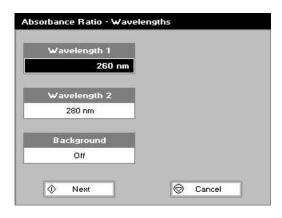
**Options** (select using key pad numbers)

- 1. Return to parameters screen (step 1 above).
- 2. Print result via selected method.
- 4. Print graph using selected method. Grayed out if no data are available.
- 7. Sample number add a prefix to the sample number and reset the incrementing number to the desired value.
- 8. Save method use the left and right arrows to select a folder to store in (Favourites/Methods 1-9), press the down arrow and enter name.
- 9. Auto-print toggles auto-print on/off.

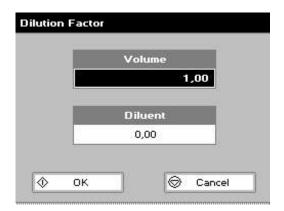
Exit options by pressing **②**, or wait.

# 7: Absorbance Ratio

This makes simple absorbance ratio measurements on samples, measuring the amount of light that has passed through a sample relative to a blank (this can be air) at two wavelengths. The procedure is as follows:



# Absorbance Ratio - Parameters Pathlength 10 mm 1,00 Dilution Factor 1,00 Units μg/ml Φ OK Back



# Step 1

Enter the first wavelength by using the keypad numbers or the left and right arrows.

Press the down arrow.

# Step 2

Enter the second wavelength as above.

Press the down arrow.

# Step 3

Select whether a background correction is applied to both wavelengths 1 and 2 using the left and right arrows.

**Step 4** (If background correction is On)

Enter the third wavelength, from which the background correction will be obtained).

# Step 5

Press Next � to enter the Parameters screen OR

Press Cancel **to** return to the Applications Folder.

# Absorbance Ratio – Parameters Screen

# Step 6

Select the pathlength (5 or 10 mm) using the left and right arrows.

Press the down arrow.

Step 7 (Dilution Factor known)

Enter a dilution factor by using the keypad numbers within the range 1.00 – 9999.

OR

Step 7 (Calculate Dilution Factor)

Press the options key: ☒☒☒.

Enter the volume of the sample (range 0.01 – 9999), using the keypad numbers.

Press the down arrow.

Enter the volume of diluent (range 0.01-9999) by using the keypad numbers.

Press OK  $\bullet$  to calculate the dilution factor and return to the Parameters screen (or press Back  $\bullet$  to cancel selections).

# Step 8

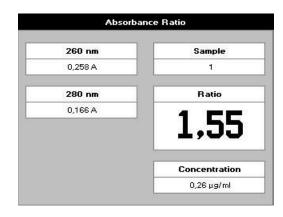
Select units of measurement, using left and right arrows. Options are:  $\mu g/\mu l$ ,  $\mu g/\mu l$ .

Press the down arrow.

# Step 9

Enter the factor using the keypad numbers (Range 0.001 to 9999).

Press OK igodot to enter the results screen or Cancel igodot to return to the Applications Folder.



# **Results Screen**

# Step 10

Insert the reference. Press 0A/100% key. This will be used for all subsequent samples until changed.

#### Step 11

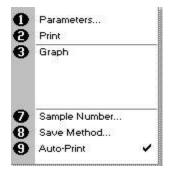
Insert sample and press **①**.

Repeat step 11 for all samples.

The absorbance at selected wavelengths is measured and the ratio between wavelengths 1 and 2 is calculated (both corrected by the background wavelength value if this was selected).

Press **t** to return to the Applications Folder.

Press ☒☒☑ to display available Options which are described below.



**Options** (select using key pad numbers)

- 1. Return to parameters screen (step 1 above).
- 2. Print result via selected method.
- 3. Toggle graph on/off. Graph shows a wavescan plot across the selected wavelengths in place of the individual wavelength.
- 7. Sample number add a prefix to the sample number and reset the incrementing number to the desired value.
- 8. Save method use the left and right arrows to select a folder to store in (Favourites/Methods 1-9), press the down arrow and enter name.
- 9. Auto-print toggles auto-print on/off.

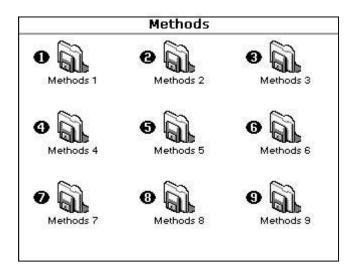
Exit options by pressing **②**, or wait.

# **FAVOURITES AND METHODS FOLDERS**

These folders are the storage locations for any user modified Applications (Methods) that are saved in the Options menu. Both are accessible from the home folders page.

# **Favourites:**

This folder enables the user to quickly select any frequently used Methods. Up to 9 Methods may be stored in the folder.



# Methods:

These are further storage folders enclosed in the top level Methods folder. Up to 9 Methods may be stored in each folder.

Operation is identical to the Favourites Folder.

Saved methods can be locked, unlocked and deleted using the Options menu. Select the method by pressing the relevant key pad number and then press the ☒☒☑ key.

# Methods - Methods 1 ① 入 Single Wavelength ① Delete Method... ② Lock Method... ③ Unlock Method...

# **Delete Method**

Press 1 to select delete method.

Select the method to be deleted using the left and right arrows.

Press **o** to delete the method

OR  $\Phi$  cancel to return to Favourites/Methods folder.

# **Lock Method**

Press 2 to select lock method.

Select the method to be locked using the left and right arrows.

Press the down arrow.

Select a pass code using the keypad numbers or left and right arrows.

Press **t** to lock the method

OR cancel to return to the Favourites/Methods folder.

# **Unlock Method**

Press 3 to select unlock method.

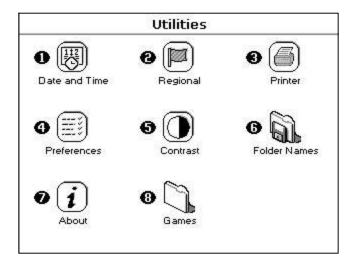
Select the method to be unlocked using the left and right arrows. Press the down arrow.

Enter the pass code using the keypad numbers or left and right arrows.

Press **t**o unlock the method

OR cancel to return to the Favourites/Methods folder.

# **UTILITIES FOLDER**

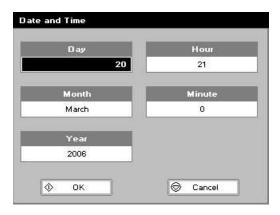


Summary Function	Keypad	number Description
Date and Time	1	Set correct time and date
@ Pagional	2	Select preferred language and number format
3 Printer	3	Printer/output options
<b>④</b> ∰ Preferences	4	Select screen layout (themes) and history
<b>6</b> Ontrast	5	Adjust screen contrast & brightness
6 Folder Names	6	Re-name folders
<b>O i</b> About	7	Serial number and software version
<b>③</b> ☐ Games	8	Spectro Blocks/Sudoku

# **Utilities**

# 1: Date and Time

The procedure is as follows:



Enter the day using the keypad numbers or left and right arrows. Press the down arrow.

Enter the month as above.

Press the down arrow.

Enter the year.

Press the down arrow.

Enter the hour.

Press the down arrow

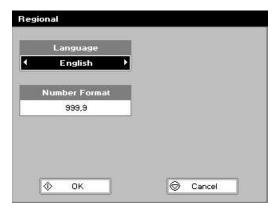
Enter the minute. Seconds are zeroed when OK is pressed.

Press OK igodath to store the settings and return to the Utilities folder OR

Press Cancel **1** to return to the Utilities folder without storing the time.

# 2: Regional

Sets Language and Number Format The procedure is as follows:



Select a language. Options are French, English, or Spanish. (German and Italian will be released in the near future). Press the down arrow.

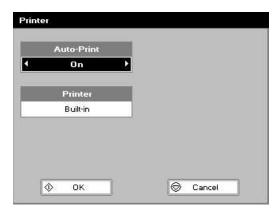
Set the decimal point style. Options are "," or ".".

Press OK igodaphi to store the settings and return to the Utilities folder OR

Press Cancel  $\mathbf{\hat{Q}}$  to return to the Utilities folder without storing the settings.

# 3: Printer

Sets up printing options The procedure is as follows:



Select whether auto-print is on or off using the left and right arrows. When auto-print is on the results are automatically printed after a measurement is taken. When it is off printing has to be initiated manually. This can also be set using the Options key (国国团) in each application or method. The default is OFF. Press the down arrow.

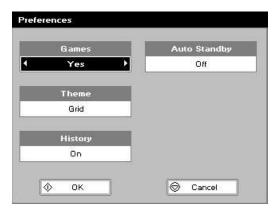
Select how the data are sent. Options are Built in (internal printer), or to a computer via USB port or Bluetooth.

Press OK igodath to store the settings and return to the Utilities folder OR

Press Cancel **to** return to the Utilities folder without storing the settings.

# 4: Preferences

Sets user preferences The procedure is as follows:



Select games function. This determines whether the games folder is displayed or not. Options are yes or no.

Press the down arrow.

Define the screen layout of folders. Options are either a grid format (default) or a list.

Press the down arrow.

Select whether to use previously entered parameters on switch on or use defaults.

Press the down arrow.

Select whether to use a standby mode after defined periods.

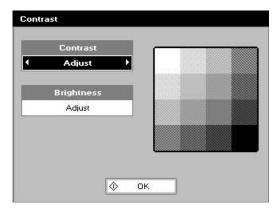
Options are 1 hour, 2 hours, at night or off.

Press OK igodath to store the settings and return to the Utilities folder OR

Press Cancel **to** to return to the Utilities folder without storing the settings.

# 5: Contrast

Ambient temperature can affect the display. This function can optimise the display for local conditions The procedure is as follows:



Adjust the contrast using the left and right arrows.

Press the down arrow.

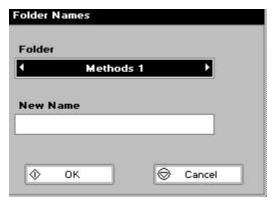
Adjust the brightness using the left and right arrows.

Press the down arrow.

Press OK  $\Phi$  to store the settings and return to the Utilities folder

# 6: Folder Names

This folder allows you to rename the method or favourite folders



Select the folder you wish to rename using the left and right arrows.

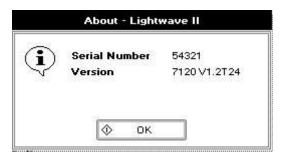
Press the down arrow.

Input the new name for the folder.

Press OK igodaphi to store the settings and return to the Utilities folder OR

Press Cancel **to** to return to the Utilities folder without storing the settings.

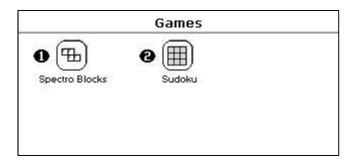
# 7: About



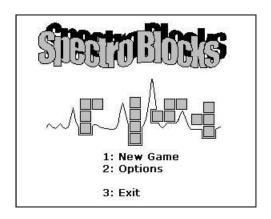
Displays the instrument serial number and software version.

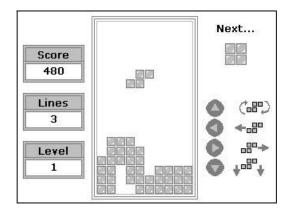
Press OK  $\Phi$  to close the window and return to the Utilities folder

# 8: Games



# 1: Spectroblocks





Classic block dropping game. Follow the instructions!

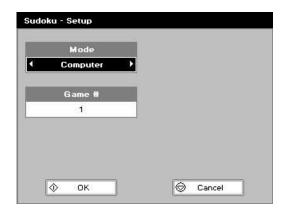
Press Cancel to return to the Utilities folder without storing the settings.

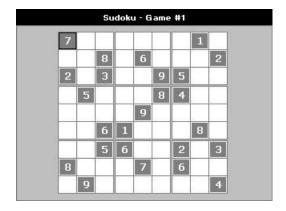
# 2: Su Doku

Can be set up as Computer mode (50 preset games) or User mode (enter your own pattern)

Use the cursors to select the square and the key pad to enter a number. Invalid numbers cannot be entered. Cells can be locked (or unlocked) by using the decimal point. Unlocked cells can be cleared using the C key (see also option key below)

The user mode starts with a blank grid.







# **Options**

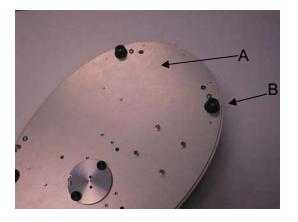
Press ☒☒☑ to display the options menu

- 1. Return to the set-up screen.
- 3. The instrument solves the game for you!
- 4. Clear all entries.
- 8. Save the game. Use the left and right arrows to select a folder to store the game in (Favourites, Methods 1-9), press the down arrow and enter name.

Press Cancel to return to the Utilities folder.

# **ACCESSORIES INSTALLATION**

# Printer installation



1. REMOVE THE POWER CABLE FROM THE INSTRUMENT. Turn the instrument over and remove cap head screws from positions A and B using the Allen key provided.



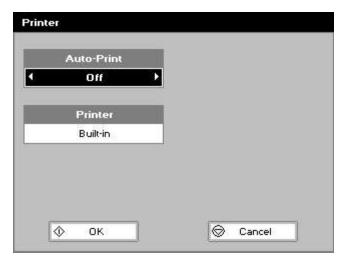
- 2. Turn the instrument back over and lift the accessory cover vertically upwards to remove. Remove the tie-wrap from the cable.
- 3. Invert the instrument and replace the cap head screws at A and B.



4. Plug the accessory cable into the printer.



5. Lower the printer onto the locating bosses and push down firmly.



Switch the instrument on and go to utilities/instrument/preferences and select the Built-in printer.

# Loading / changing the printer paper



1. Lift off the paper cover.

Lock the platen and turn the knob to feed the paper



2. Feed in the paper.

Sometimes it helps if the platen lock is released.

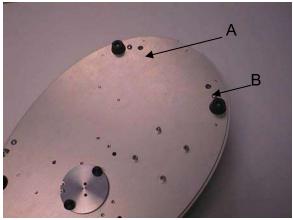


3. Paper gripped.



4. Cover replaced.

# Bluetooth accessory installation



 REMOVE THE POWER CABLE FROM THE INSTRUMENT. Turn the instrument over and remove the cap head screws from positions A and B using the Allen key provided.



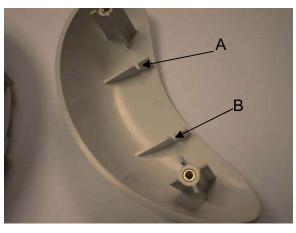
2. Turn the instrument back over and lift the accessory cover vertically upwards to remove. Remove the tie-wrap from the cable



3. Plug the accessory cable into the Bluetooth module.



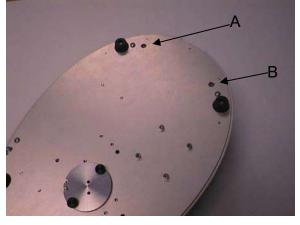
5. Note the slots in the base of the case. The two lugs on the Bluetooth module plug into these



5. Note the slots in the accessory cover, designed to engage with the Bluetooth accessory PCB



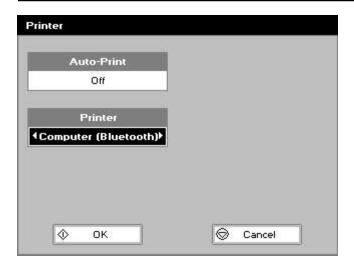
6. Lower the accessory cover vertically downwards onto the instrument, engaging the PCB in the slots.



7. Invert the instrument and replace the cap head screws at A and B.



8. Attach the license label as shown.



9. Switch the instrument on and go to the preferences page under utilities/instrument, and select the Bluetooth option.

# **PRINT VIA COMPUTER**

- PVC (Print Via Computer) is a small application running under Windows 2000™ or Windows XP™ to
  enable a Biowave II or Lightwave II to transfer data into a PC environment. From there the user has a
  selection of choices, the data can be both printed or saved (in a variety of formats). PVC is capable of
  supporting several instruments simultaneously, limited only by hardware and the speed of the host system.
- PVC can operate via USB and Bluetooth simultaneously
- PVC can store data either to a common directory or be configured to save to independent directories by both file format and connection.
- PVC can save data in graphics format, text format or as an Excel™ file

# Installation

See the manual included on the PVC CDROM for installation and operating instructions.

# **ACCESSORIES**

USB cable source locally Built-in printer accessory 80-3003-84 Bluetooth accessory 80-3003-96

# **MAINTENANCE**

# After Sales Support

Support agreements that help you to fulfil the demands of regulatory guidelines concerning GLP/GMP are available.

- Calibration, certification using filters traceable to international standards
- Certificated engineers and calibrated test equipment
- Approved to ISO 9001 standard

Choice of agreement apart from break down coverage can include

- Preventative maintenance
- Certification

When using calibration standard filters, insert such that the flat surface is facing away from the spring end of the cell holder.

Observe all necessary precautions if dealing with hazardous samples or solvents.

# Lamp Replacement

The xenon lamp should not need replacement until after several years of use. In the unlikely event that it does need replacing, this should be undertaken by a service engineer from your supplier.

# Cleaning and general care of the instrument

# **External cleaning**

Switch off the instrument and disconnect the power cord.

Use a soft damp cloth.

Clean all external surfaces.

A mild liquid detergent may be used to remove stubborn marks.

# Changing cell holder or removal for cleaning

This can be removed by undoing the appropriate screws on the bottom of the instrument.

# SPECIFICATION AND WARRANTY

Wavelength range 190 - 1100 nm
Monochromator Flat grating

Wavelength calibration Automatic upon switch on Spectral bandwidth 5 nm (3nm Lightwave II<sup>+)</sup>

Wavelength accuracy ±2 nm
Wavelength reproducibility ±1 nm

Light sourcesPulsed xenon lampDetector1024 element CCD arrayPhotometric range- 0.300 to 2.500A, 0 to 199%T

Photometric linearity ±0.005 Abs or 1% of the reading, whichever is the greater

@ 546 nm

Photometric reproducibility ±0.003 Abs (0 to 0.5 Abs), ±0.007 Abs (0.5-1.0 Abs)

Stray light <0.5% at 220 nm and 340 nm using NaNO<sub>2</sub>

Zero stability ±0.01 Abs/hour after 20 min warm up @ 340 nm

*Noise* 0.005 pk to pk 0.002 rms

Digital output USB port standard, Bluetooth option

*Dimensions* 260 x 390 x 100 mm

Weight <4.5 kg

Power input 90-250 V, 50/60 Hz, Max 30 VA

Specifications are measured after the instrument has warmed up at a constant ambient temperature and are typical of a production unit. As part of our policy of continuous development, we reserve the right to alter specifications without notice.

# Warranty

- Your supplier guarantees that the product supplied has been thoroughly tested to ensure that it meets its
  published specification. The warranty included in the conditions of supply is valid for 12 months only if the
  product has been used according to the instructions supplied. The supplier can accept no liability for loss or
  damage, however caused, arising from the faulty or incorrect use of this product.
- This product has been designed and manufactured by Biochrom Ltd, 22 Cambridge Science Park, Milton Road, Cambridge CB4 0FJ, UK. However, please contact your original supplier in the first instance if you experience technical or sample handling or sample handling difficulties.