人と科学のステキな未来へ
コスモ・バイオ株式会社

Pico200

µl Spectrophotometer

USER MANUAL

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Part no 41 56 1883

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DECLARATION OF CONFORMITY

This is to certify that the following products conform to the requirements of the following Directives -:

Pico200 Micro volume spectrophotometer part numbers:

P200/P200P/P200BT/P200SD/P200E/P200PE/P200BTE/P200SDE

2006/95/EC	Low voltage equipment safety directive
2004/108/EC	EMC directive
2002/96/EC & 2003/108/EC	EC Directive on Waste Electrical and Electronic Equipment (WEEE)
2002/95/EC	ROHS directive

Standards to which conformity is declared, where relevant, are as follows

EN61010-1:2010	Safety requirements for electrical equipment for measurement, control and laboratory use. General requirements
EN61326-1:2006	Electrical equipment for measurement, control and laboratory use -EMC Requirements
EN 12100-1,2:2003 (+A1:2009)	Safety of machinery – Basic concepts, general principles for design
EN 14121-1:2007	Safety of machinery, Risk assessment

P200BT and P200BTE also conform to the requirements of the following Directives

1999/5/EC	Radio and Telecommunications terminal equipment directive (for Bluetooth devices)

This equipment has been tested and found to comply with the limits for a CLASS A digital device, pursuant to part 15 of the FCC Rules.

ESSENTIAL SAFETY NOTES

Caution symbol. 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.

There are no bio-hazardous materials within the unit; however this unit could be used with bio-hazardous samples. Before using the instrument the customer should have in place decontamination procedures designed to protect laboratory workers from occupationally acquired infections. In the event of a spillage the area should be disinfected, rinsed with distilled water and then allowed to dry. The exterior may be wiped with a suitable disinfectant cleaning wipe.

- Decontamination. Equipment returned for repair should include an appropriate decontamination certificate
- It is the responsibility of the customer to ensure that the user of the equipment is trained in the techniques used and is provided with a safe working environment.
- Any chemicals used in Analyses should be used, stored and disposed of in accordance with manufacturer's guidelines and local safety regulations
- Toxic Fumes. Efficient laboratory ventilation must be provided when working with volatile solvents or toxic substances
- Waste disposal. Disposal of some solvents and chemicals may be classed as hazardous waste and must be dealt with in accordance with local regulatory practice.
- Personal protective equipment. This is not required to operate the unit but the samples measured may require PPE. A local risk assessment should be carried out.

The lamp source within the unit is flashed to produce UV and visible spectral energy through the sample. The energy level is low, confined to the optical path and not a hazard in normal use. Do not operate the unit with the optical fibre connections disconnected as prolonged exposure to the beam could be hazardous to eyes and skin.

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 supply unit and 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 on. 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.
- In accordance with good laboratory practice it is recommended that periodic performance validation checks be carried out using traceable standards and that when measuring batches of samples that known standards be included for comparison and verification of result values
- 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.

The  WEEE crossed-out wheeled bin symbol indicates that the product is covered by the Waste Electrical and Electronic Equipment (WEEE) Directive and is not to be disposed of as unsorted municipal waste. Any products marked with this symbol must be collected separately and in accordance with local regulatory practice

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 Picodrop® is a microlitre spectrophotometer that provides the user with the facility to recover their sample after measurements have been taken. It is a full spectrum (220 – 950nm) spectrophotometer which allows for measurements of common laboratory samples, such as DNA, RNA and protein, in small volumes, with a high degree of accuracy and precision.

Samples are contained within patented UVpette™ pipette tips. There is no possibility for cross contamination or carry over on a sample platform. Precious samples can be handled within a sterile environment and are completely recoverable.

A 2.5µl sample is drawn up directly into the UVpette™ using a P10 pipette. The pipette is placed into the holder, which positions the tip through a light beam, emitted from a fibre optic cable connected to the tip holder. The light source is a pulsed xenon lamp and the light path through the tip is 0.9mm. **The default path length is set in all modes to Picotip (0.9mm),** which is designed for use with UVpette tips as supplied by Picodrop Ltd. Resulting data displayed in reports and graphs are calculated to display the equivalent 10mm absorbance values.

ONLY USE PIPETTE TIPS MANUFACTURED BY PICODROP LIMITED

Consumable re-order details:

Ref# UVTIPB	P10 UVpette pipette 96 boxed tips, UV transparent to 230nm
Ref# UVTIPG	P10 UVpette pipette tips 2,000 loose bagged, UV transparent to 230nm

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 "Pico200" 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
Life Science	Standard Life Science methods such as nucleic acid assays, protein assays and cell counting

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 tip or cell chamber; therefore please ensure the cell is inserted in the correct alignment.
- The optional cell holder accepts standard 10 mm pathlength quartz, glass or plastic cells.
- The optical height is 15 mm, and the minimum volume that can be used is approx. 10µl in a Quartz ultra-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.

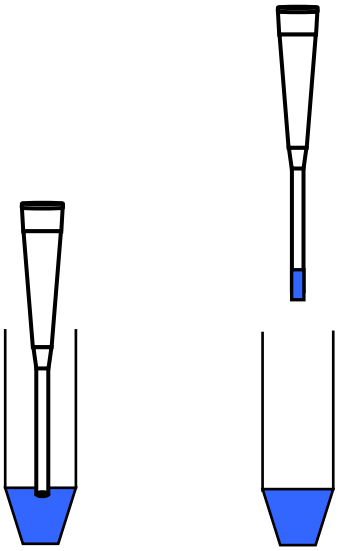
Correct Pipetting Procedure

Vortex sample briefly (5-20secs)
Spin down samples briefly (10-15secs in a microfuge).
Using a P10 pipette and a UVpette™ tip, pipette your sample. A minimum volume of 2.5ul is recommended. To minimise solution on the outside of the tip, avoid submerging the tip too far below the sample meniscus. If necessary, **Wipe off excess liquid from outside of tip with a dry lint-free tissue**, this is particularly important if using viscous protein solutions. Be careful not to touch the bottom of the tip as the sample may be drawn out by the tissue.

Do not place UVpette tips or your sample too close to heaters or the fan of the PC as heating the tips or sample may result in a rapid contraction in volume once the tip is placed the cooler pipette holder. This sample contraction will result in a space or bubble being visible at the bottom of the tip. This space may interfere with sample measurement if allowed to rise more than 2mm up the tip. It is preferable that the sample, tip and pipette holder are allowed to equilibrate to room temperature for 5 minutes before commencing measurement.

The detection point of the tip is 1.5mm from the end of the tip so it is best procedure to try not to submerge the tip more than 1mm into your solutions, otherwise tip wiping may be necessary.

It is optional whether you decide to use the same tip for both blank and sample or a different tip each time. The special ultra-low retention tips will ensure zero carryover of blank to your sample and so using the same tip is a safe operation. Using different tips for blank and sample will also provide results which meet the published specification but does however introduce an additional variable which may become significant with very low concentration samples i.e. <10ul/ng DNA



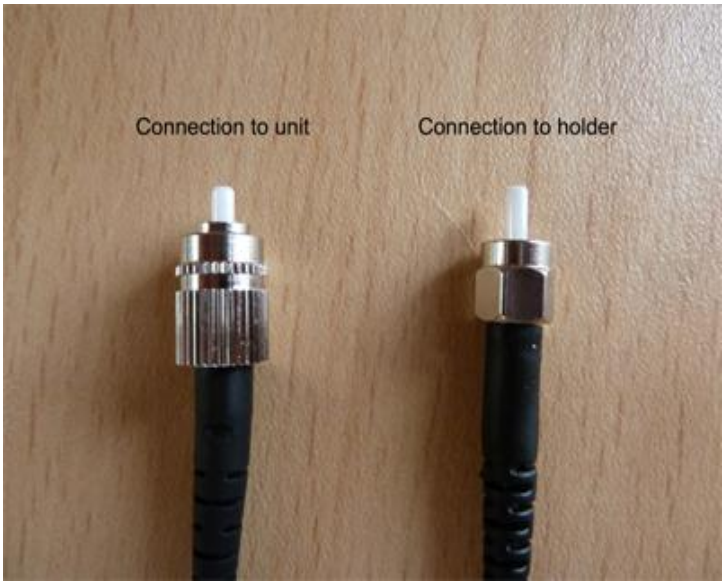
Recommended pipetting procedure

Instrument Set Up

Hardware installation (If not already pre-assembled)

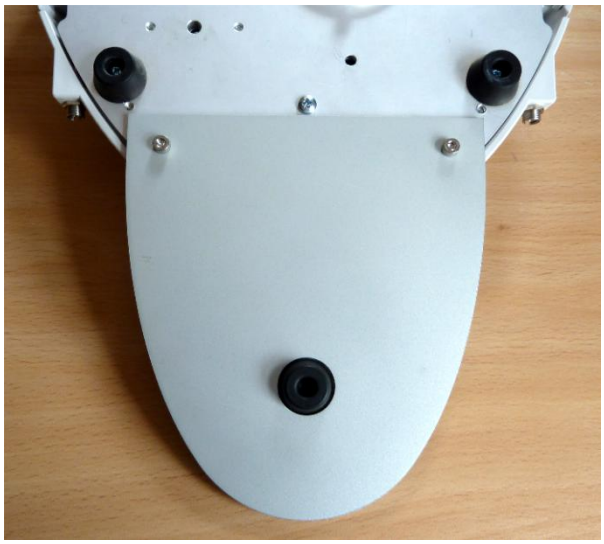
The pipette holder has been dismantled and disconnected for shipment. Fix the pipette holder to its round base by simply screwing it on.

Locate the 2 Silver fibre optic cables. The fibres have a different connector at each end.



Connect the cables to the base of the pipette holder. Push the connector into the socket and hand-tighten the nut to hold the connector in place. ***Do not use tools to tighten the fixing nut.***

Connect the base plate to the underside of the unit with the 2 hex head screws provided



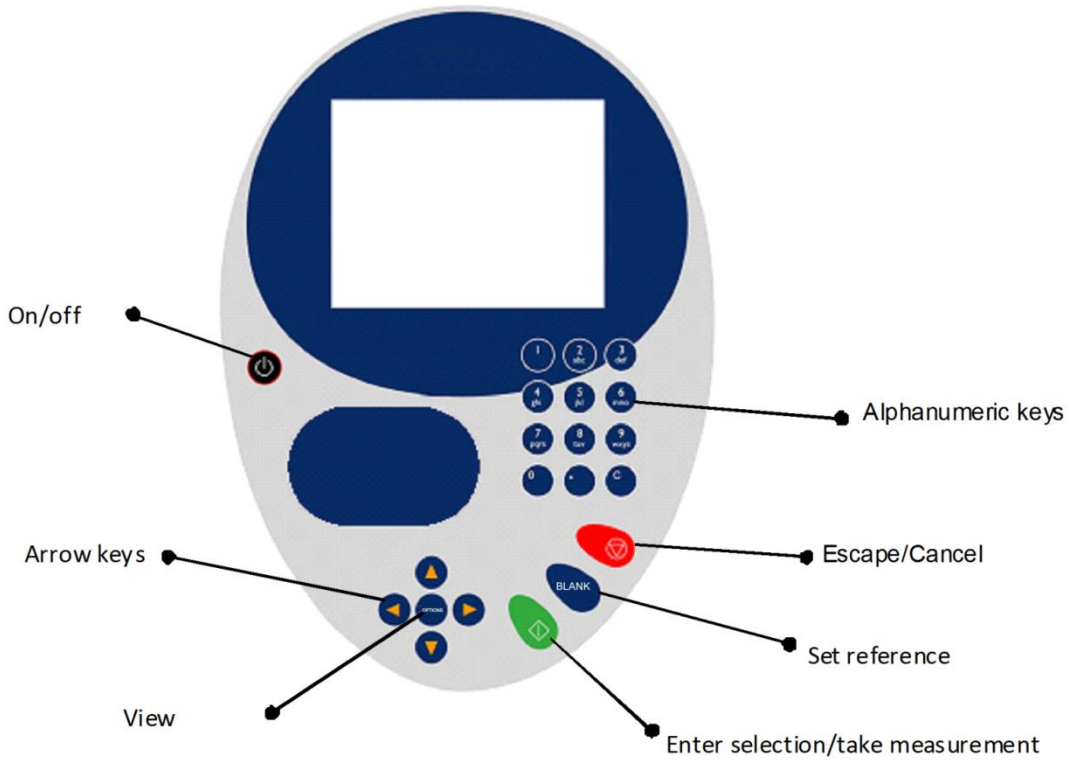
Place the holder on the base plate and connect fibre cables to the unit connectors. Note the groove in connector to unit, ensure correct alignment before screwing in connector.

Slide the provided ‘PIPETTE GUIDE’ collar onto the pipette. The easiest way to attach the collar is to place the collar in the tall pipette holder and then insert the pipette until the collar has located on the pipette. Remove the pipette and the collar will remain attached to the pipette. The collar can be easily removed for cleaning if necessary. Please note that tip ejector on the Gilson manual pipette will NOT function once the adapter is in position. Tips should be removed from the pipette by hand.

Key	Action
On/off key:	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: Options	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: Blank	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:	Enter, or confirm, a selection. Take a measurement.

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.



Options (select using key pad numbers)

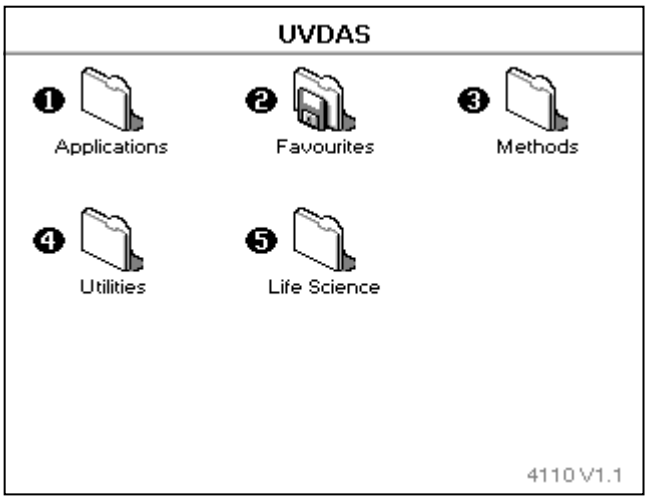
1.	View parameters for the experiments.
2.	Print the results.
3,4,5,6	Described in the application.
7.	Define the sample number you wish to start from.
8.	Save the parameters as a method to a defined folder name with a defined method name.
9.	Toggle auto-print on/off. Default is off.

Exit options by pressing , or wait.

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

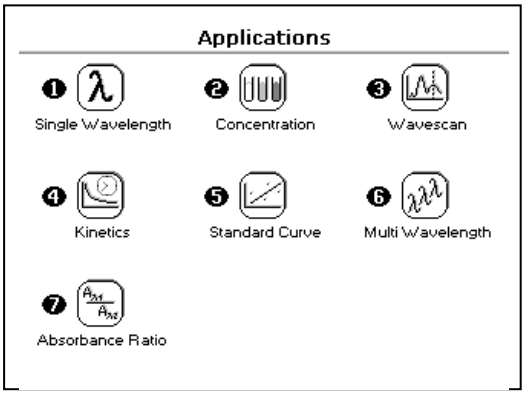
Software style

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 number	Description
1 Applications	1	Single wavelength, Concentration, Wavelength scan, Kinetics, Standard Curve, Multiple wavelengths and Ratio
2 Favourites	2	Saved User selected and configured methods
3 Methods	3	Sub folder selection for User selected and configured methods
4 Utilities	4	Instrument set up (date, time, language, etc) and Games
5 Life Science	5	Nucleic acids, Proteins and Cell counting

THE APPLICATIONS FOLDER



SUMMARY:

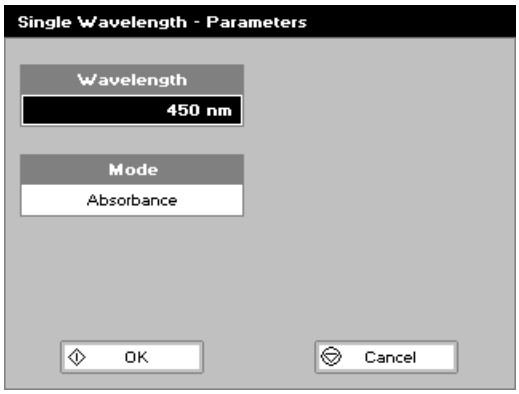
Function	Key pad number	Description
Single Wavelength	1	Absorbance or %T (transmission) at a single user defined wavelength.
Concentration	2	Concentration measurement at a single wavelength based on a simple Factor entered or calculated from a single standard.
Wavescan	3	Wavelength scan between two user defined wavelengths. Range 220-950 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 its 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).

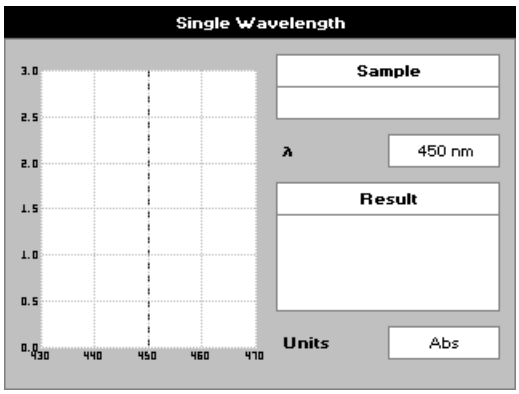
1: Single Wavelength – Abs and %T

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:



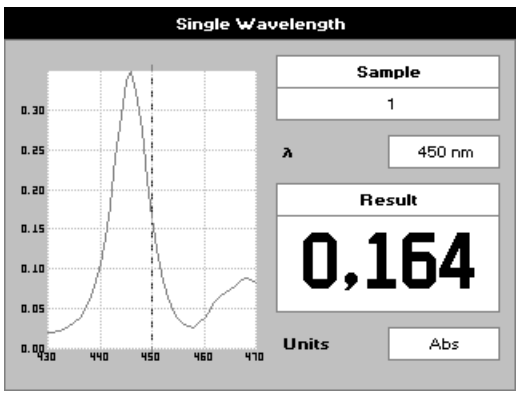
Step 1
Set wavelength by using keypad numbers or left and right arrows.
Press the down arrow key.
Step 2
Select the mode, *Absorbance* or *%T*, using the left and right arrows.
Step 3
To enter the results screen with the selected parameters press OK

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



Step 4
Insert the reference. Press Blank key. This will be used for all subsequent samples until changed.
Step 5
Insert sample and press .

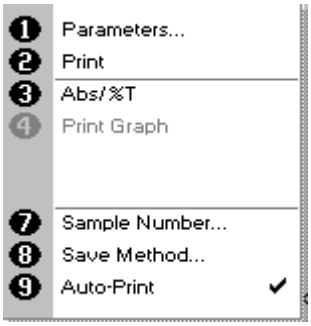
Repeat step 5 for all samples.



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 to return to the Applications Folder.

Press Options to display available Options which are described below.



Options (select using key pad numbers)

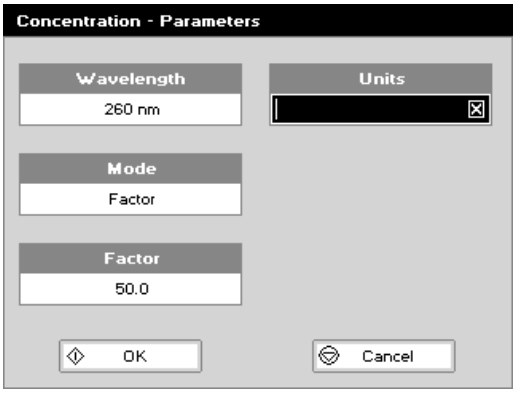
1. Return to parameters screen (step 1 above).
2. Print result via selected method.
3. Toggle between Absorbance and %T mode.
4. Print graph – greyed out if no data are available.
7. Sample number – add a prefix to the sample number and reset the incrementing number to the desired value.
8. Save method – 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  , or wait.

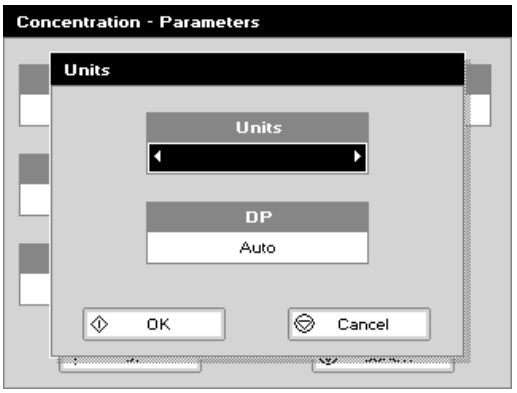
2: Concentration

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.

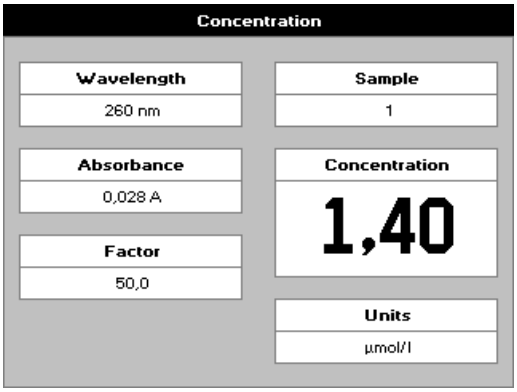
The procedure is as follows:



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 arrows.
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 Options 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 .

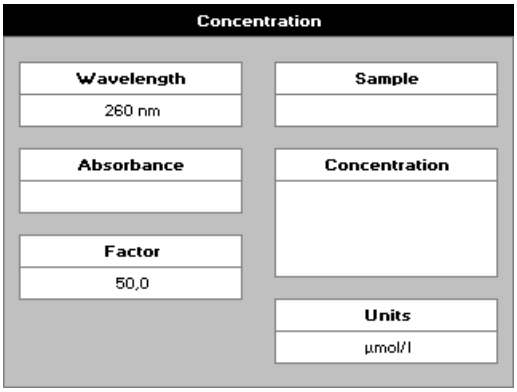


Step 5
To enter the results screen with the selected parameters press OK
OR
Cancel the selections and return to the Applications Folder by pressing Cancel .

Step 6 (if using a Factor)
Insert the reference. Press Blank key. This will be used for all subsequent samples until changed.

STEP 7 Insert sample and press .
The concentration of the sample is displayed. Results shown as ---- indicate the concentration is out of range.
Repeat step 7 for all samples.

Press to return to the Applications folder.
Press Options to display available Options which are described below.



Step 6 (if using standard mode)

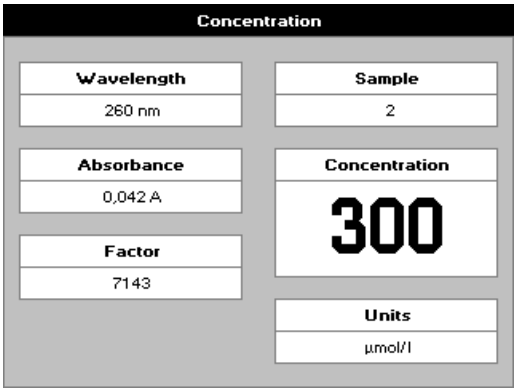
Insert the reference. Press Blank key. This will be used for all subsequent samples until changed.

Press to display the Run Standard screen.

Run the standard by pressing

OR

Press cancel to return to the measure screen.



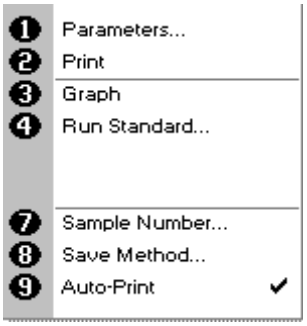
STEP 7 Insert the sample and press .

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

Repeat step 7 for all samples.

Press to return to the Applications Folder.

Press Options to display available Options which are described below.



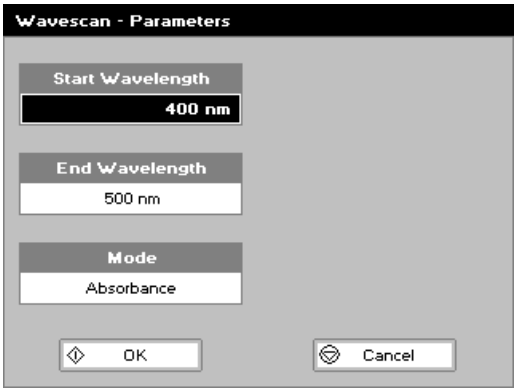
Options (select using key pad numbers)

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.

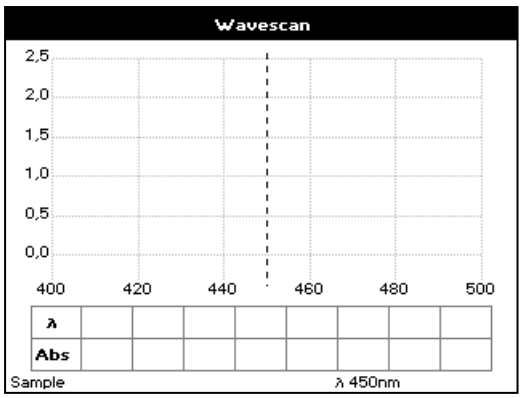
Exit options by pressing , or wait.

3: Wavescan

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

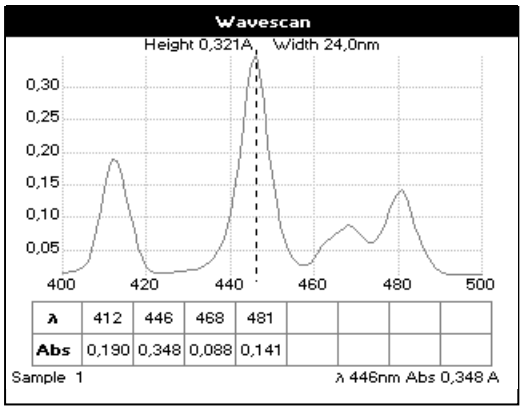


Step 1
Set start wavelength by using keypad numbers or left and right arrows.
Press the down arrow key.
Step 2
Set end wavelength by using keypad numbers or left and right arrows.
Press the down arrow key.
Step 3
Select the mode, *Absorbance* or *%T*, using the left and right arrows.
Step 4
To enter the measurements screen with the selected parameters press OK
OR
Cancel the selections and return to the Applications Folder by pressing Cancel .



Step 5
Insert the reference. Press Blank key. This will be used for all subsequent samples until changed.
Step 6
Insert sample and press .

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.

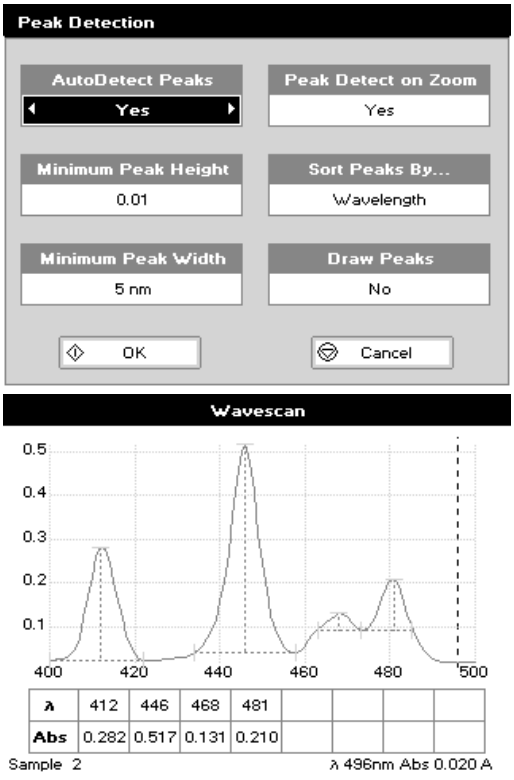
Press to return to the Applications Folder.

Press Options 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.
5. 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...
6. 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 , or wait.



Peak Detection (Shortcut button 4)

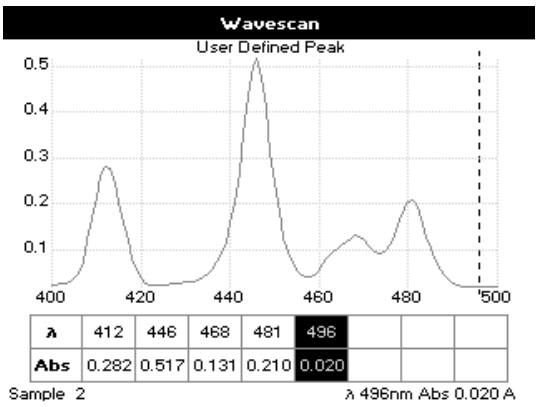
AutoDetect Peaks: Turns on and off the automatic peak detection. The following options determine how peaks are detected:
Minimum peak height: Minimum height the peak has to be above the higher of the two adjacent minima for the peak to be detected
Minimum peak width: Minimum width of the peak as determined 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).

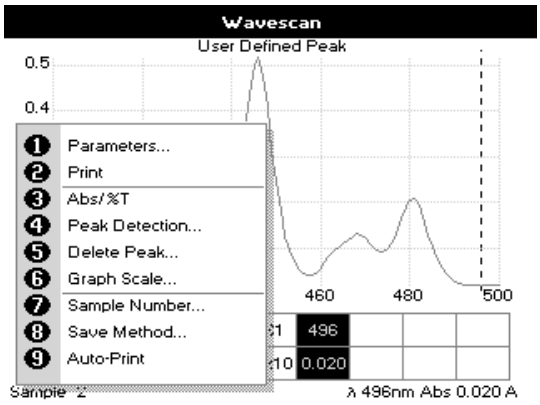
Peak Detect on Zoom: Determines whether peaks are re-assessed and tabulated when the user zooms into a region of the wavescan. If off leaves the peak detection as determined on the un-zoomed display

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 ignores the selection, pressing accepts them.

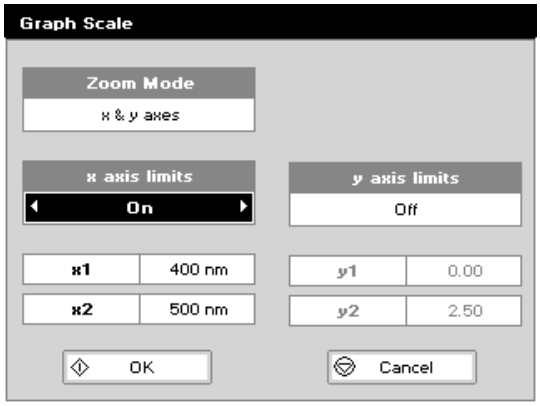




Add Peak... (Shortcut button 5)

Adds a used defined peak at the current cursor position. The entry is then displayed 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 ignores the selection; pressing accepts them and displays the required graph.

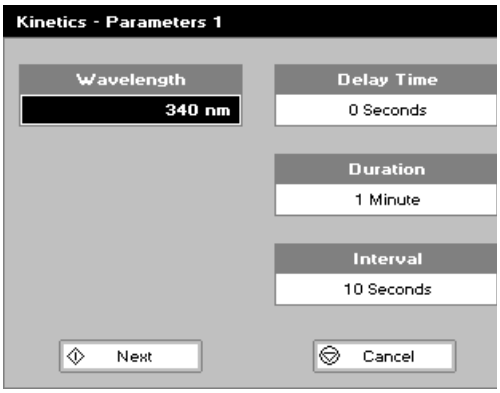
4: Simple Kinetics

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*.

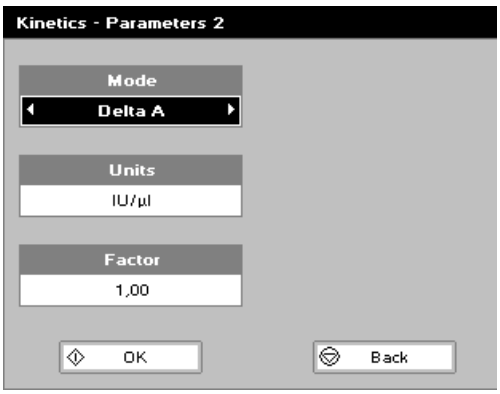
For this reason, the change in absorbance per minute **(ΔA/min**), concentration (**ΔA/min x factor**) and **correlation coefficient** (calculated from a best fit of the data points) are displayed. They may not be relevant for simple kinetics experiments.

The procedure to define a new method is as follows:



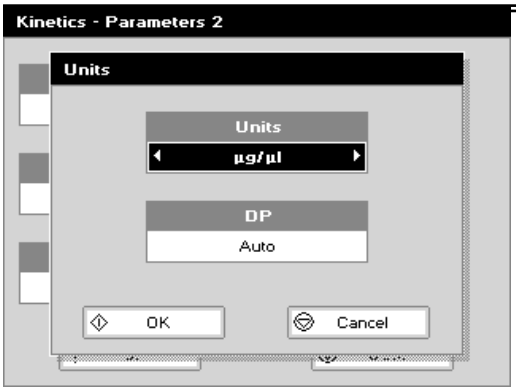
Kinetics Parameter 1 Screen
Step 1 (Wavelength)
Enter all numerical values using the keypad numbers or the left and right arrows. Use the up and down arrow keys to move between boxes.
Step 2 (Delay time)
Enter the delay time in seconds before measurements are taken. This can be a maximum of 600 seconds (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 to go to the next parameters screen
OR



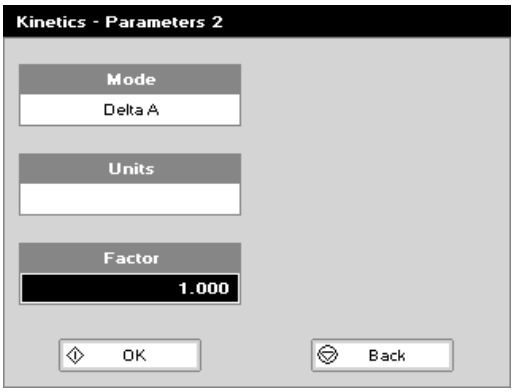
Press Cancel to return to the Applications Folder.

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: The user can enter a text string up to 8 characters long. To access a list of pre-defined units press the Options key Options 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 .



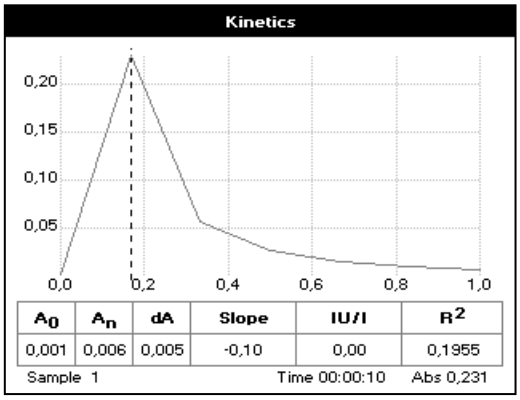
Step 8

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 to enter the Results screen
OR

Press Cancel to return to the Parameters 1 screen.



Results

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

Insert the sample and press to start the run.

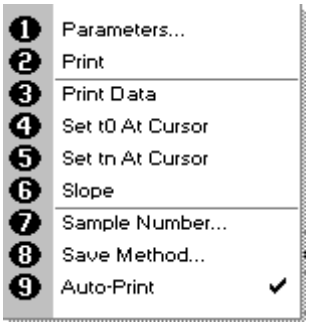
Time (min) is displayed at the bottom of the screen, and absorbance data are plotted on the graph as testing proceeds. The table below the graph gives: absorbance values at T_0 (start of calculation), T_n (finish of calculation, change in absorbance, slope, regression parameter (R^2) of the calculated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

Press Options to display available Options which are described below.



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_n position (finishing point for the slope and dA calculation) at the current cursor position. Value is retained for subsequent samples.
6. Toggle the calculated slope line on and off.
   Note: if any data points enclosed by t_0 and t_n are beyond the range of the instrument (>2.5A or <-0.3A) then this option is greyed out.
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.

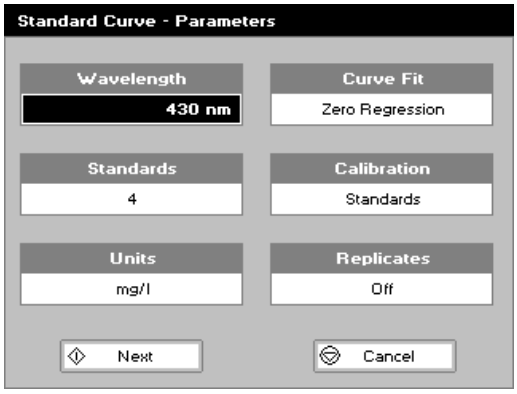
Exit options by pressing  , or wait.

5: Standard Curve

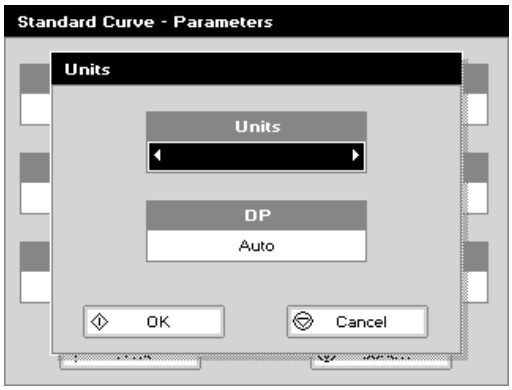
The construction of a multi-point calibration curve from standards of known concentration to quantify unknown samples is a fundamental use of a spectrophotometer; this instrument has the advantage of being able to store this curve as a method, using up to 9 standards.

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.

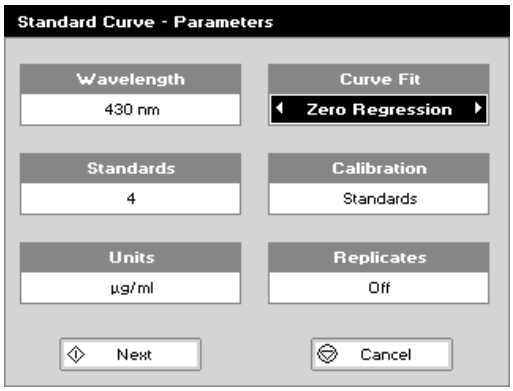
The procedure is as follows:



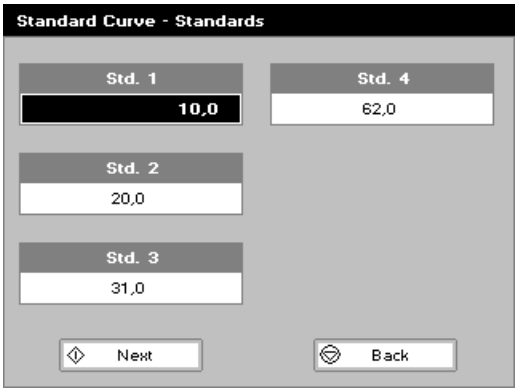
Step 1
Select the wavelength using the keypad numbers or left and right arrows.
Press the down arrow.
Step 2
Enter the number of standard concentration points to be used in the curve (1-9).
Press the down arrow.



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 Options 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 .



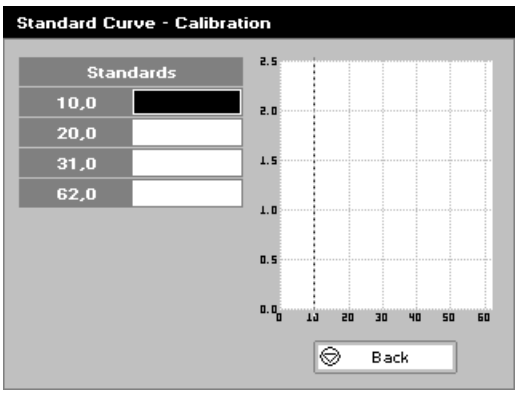
Step 4
Select the type of curve fit using the left and right arrows.
Options: straight line regression, a zero regression (this forces the straight line through the origin), interpolated or cubic spline.
Step 5
Select the calibration mode: either Standards (measure prepared standards) or Manual (keypad 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 to 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 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.

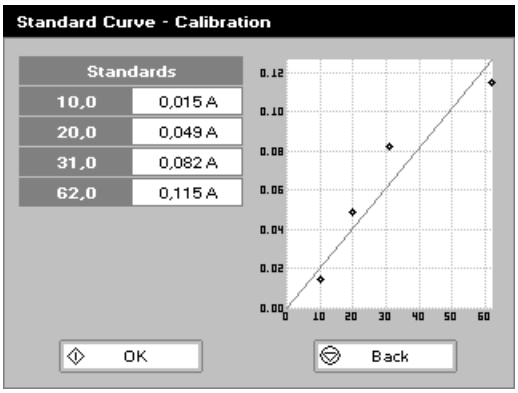


Calibration Screen (replicates off)
This shows the calibration values and allows standards to be measured.
Step 10

Insert the reference. Press Blank 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 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 to accept the calibration and go to the Results screen (see below)
OR

Press Back to return to the Standards screen.

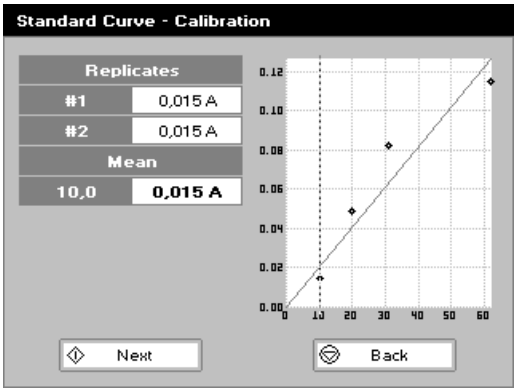
Calibration Screen (replicates on)
This shows the calibration values and allows standards to be measured.
Step 10
Insert the reference. Press Blank key.
This will be used for all subsequent samples until changed.
Step 11
Press  to display the replicate entry boxes. Use C to clear previously stored results 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  to accept the calibration and go to the Results screen (see below)
OR
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  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 Blank key. This will be used for all subsequent samples until changed.
Step 14
Insert the sample and press  .
The concentration of the sample is taken and displayed.
Repeat step 14 for all samples.
Press  to return to the Applications Folder.

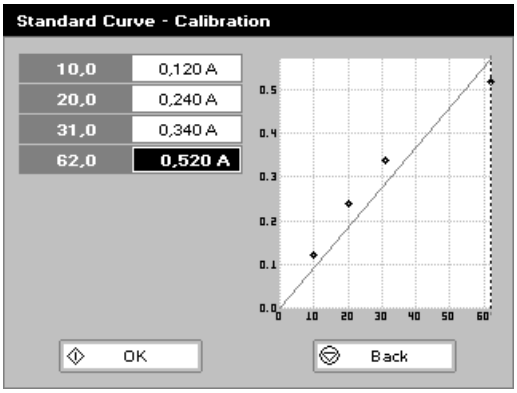
Press Options to display available Options which are described below.



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.

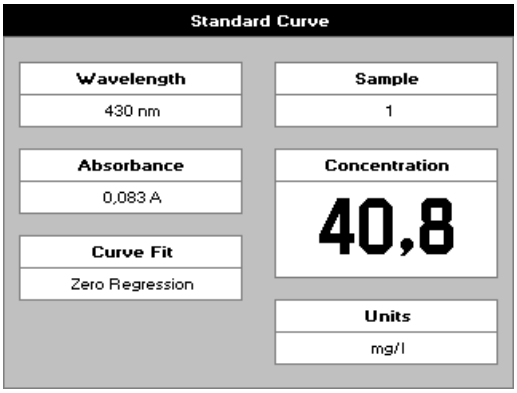
Exit options by pressing , or wait.



Options (select using key pad numbers)

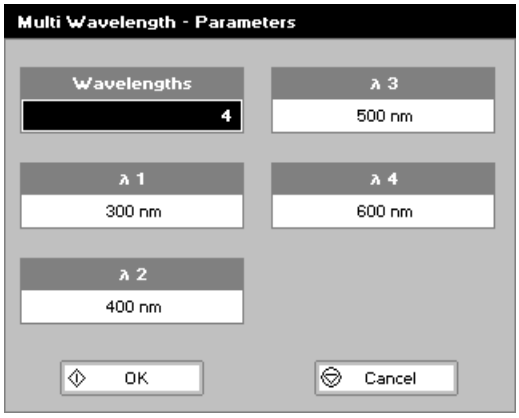
1. Return to parameters screen (step 1 above).
2. Print result via selected method.
4. Print graph using selected method. Grayed out if no data are available.
7. Sample number – add a prefix to the sample number and reset the incrementing number to the desired value.
8. Save method – use the left and 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 , or wait.



6: Multiple Wavelength

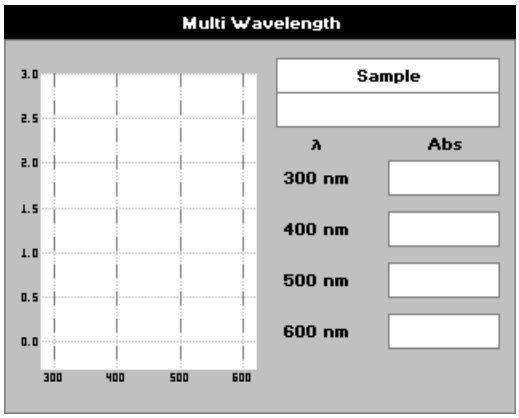
This makes up to 5 absorbance measurements on the same sample. The procedure is as follows:



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 to enter the results screen
OR

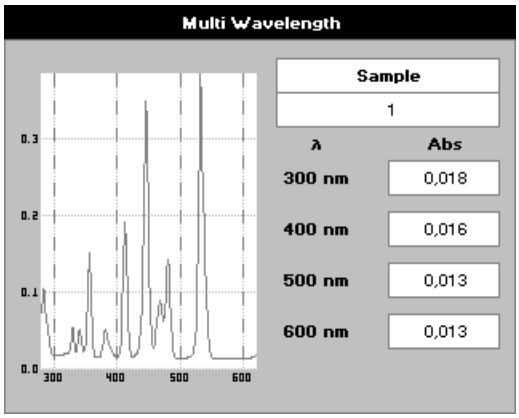
Press Cancel to return to the Applications Folder.



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 .

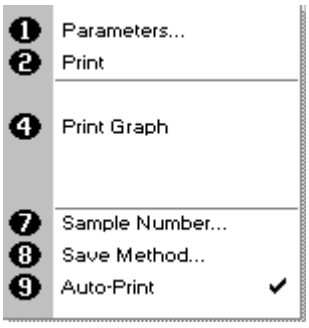
Repeat step 5 for all samples.



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

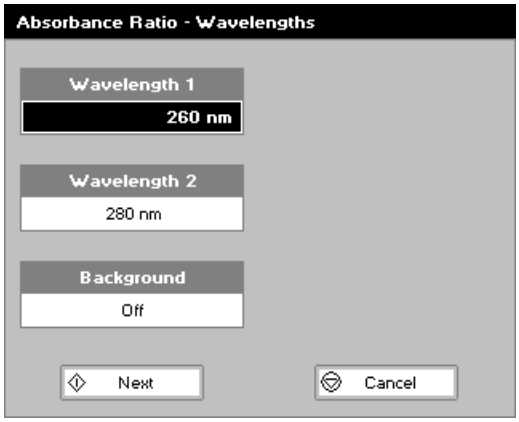
Press to return to the Applications Folder.

Press Options to display available Options which are described below.



7: Absorbance Ratio

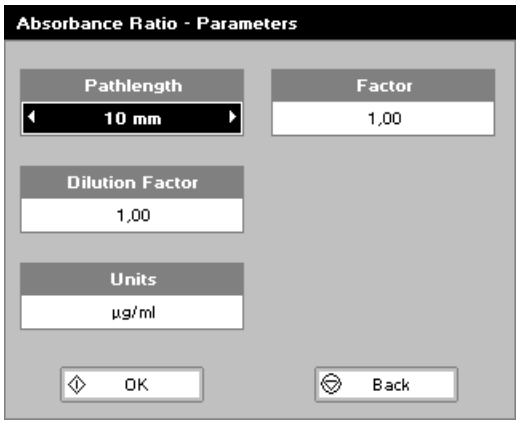
This makes simple absorbance ratio measurements on samples, measuring the amount of light that has passed through a sample relative to a blank (this can be air) at two wavelengths. The procedure is as follows:



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

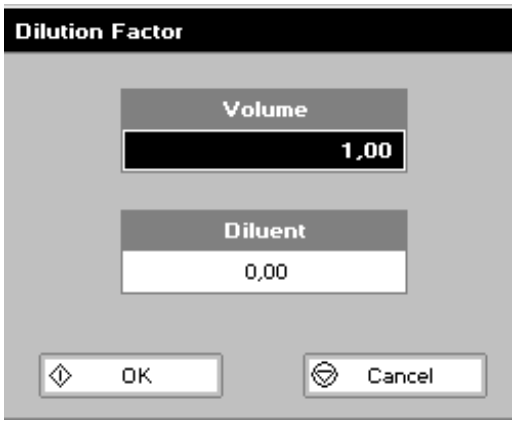
Press Cancel to return to the Applications Folder.

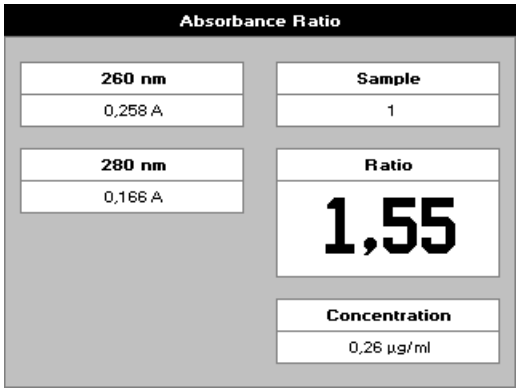


Absorbance Ratio – Parameters Screen
Step 6
Select the pathlength (5 or 10 mm) using the left and right arrows.
Press the down arrow.
Step 7 (Dilution Factor known)
Enter a dilution factor by using the keypad numbers within the range 1.00 – 9999.
OR
Step 7 (Calculate Dilution Factor)
Press the options key: Options.
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 to calculate the dilution factor and return to the Parameters screen (or press Back to cancel selections).
Step 8
Select units of measurement, using left and right arrows. Options are: µg/ml, ng/µl, µg/µl.
Press the down arrow.
Step 9
Enter the factor using the keypad numbers (Range 0.001 to 9999).

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





Results Screen
Step 10

Insert the reference. Press Blank key. This will be used for all subsequent samples until changed.

Step 11

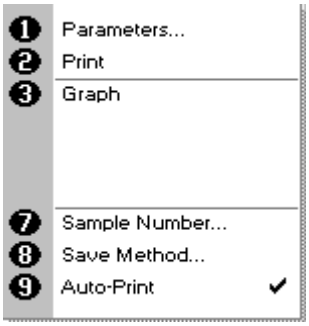
Insert sample and press .

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 to return to the Applications Folder.

Press Options 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 , or wait.

THE LIFE SCIENCE FOLDER

Life Science Folder

This contains three sub folders; Nucleic Acids, Protein and Cell Count. Contents of these sub folders are detailed below:

1. Nucleic Acids	1	DNA	Concentration and purity check for DNA samples
	2	RNA	Concentration and purity check for RNA samples
	3	Oligo	Concentration and purity check for oligo samples
2. Protein	1	UV protein (Christian Warburg)	Protein determination at 280nm
	2	BCA	Protein determination at 562nm
	3	Bradford	Protein determination at 595nm
	4	Lowry	Protein determination at 750nm
	5	Biuret	Protein determination at 546nm
3. Cell Count	1	OD600	Cell culture OD600 with correction factor

DNA, RNA and oligonucleotide characterization

Nucleic Acid Quantification (NAQ)

- Nucleic acids can be quantified at 260 nm because it is well established that a solution of DNA in a 10 mm pathlength cell with an optical density of 1.0 has a concentration of 50 µg/ml, or 40 µg/ml in the case of RNA. Oligonucleotides have a corresponding factor of 33 µg/ml, although this does vary with base composition; this can be calculated if the base sequence is known.
  Concentration = Abs260 * Factor
- The instrument uses factors 50, 40 and 33 as defaults for DNA, RNA and oligonucleotides, respectively, and compensates for dilution and use of cells which do not have 10 mm pathlength; dilution factor and cell/tip pathlength can be entered.

Nucleic Acid Purity Checks

- Nucleic acids extracted from cells are accompanied by protein, and extensive purification is required to separate the protein impurity. The 260/280 ratio gives an indication of purity; it is only an indication, however, and not a definitive assessment. Pure DNA and RNA preparations have expected ratios of ≥ 1.8 and ≥ 2.0, respectively; deviations from this indicate the presence of impurity in the sample, but care must be taken in interpretation of results.
- The 260 nm reading is taken near the top of a broad peak in the absorbance spectrum for nucleic acids, whereas the 280 nm reading is taken on a steep slope (i.e. small changes in wavelength cause large changes in absorbance). Consequently, small variations in wavelength at 280 nm will have a greater effect on the 260/280 ratio than variations will at 260 nm. Thus different instruments of the same and different types may give slightly different ratios due to variations in wavelength accuracy. But each instrument will give consistent results within itself.
- Concentration also affects 260/280 readings. If a solution is too dilute, the readings will be at the instrument's detection limit, and results may vary as there is less distinction of the 260 peak and 280 slope from the background absorbance. This is one reason why the Abs260 value should be greater than 0.1 for accurate measurements.
- An elevated absorbance at 230 nm can indicate the presence of impurities as well; 230 nm is near the absorbance maximum of peptide bonds and also indicates buffer contamination since This, EDTA and other buffer salts absorb at this wavelength. When measuring RNA samples, the 260/230 ratio should be > 2.0; a ratio lower than this is generally indicative of contamination with guanidinium thiocyanate, a reagent commonly used in RNA purification and which absorbs over the 230 - 260 nm range. A wavelength scan of the nucleic acid is particularly useful for RNA samples.
- The instrument can display 260/280 and 260/230 ratios, and compensates for dilution and use of cells that do not have 10 mm pathlength; dilution factor and cell pathlength can be entered.

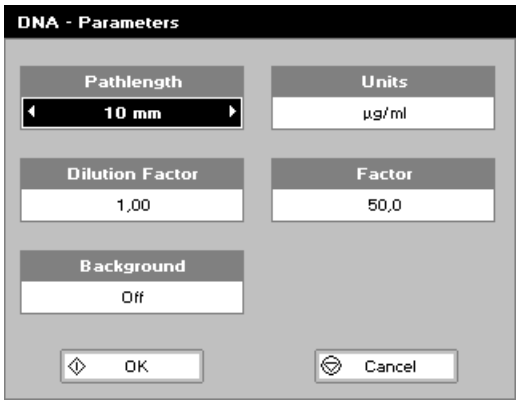
Use of Background Correction

- Background correction at a wavelength totally separate from the nucleic acid and protein peaks at 260 and 280 nm, respectively, is sometimes used to compensate for the effects of background absorbance. The wavelength used is 320 nm and it can allow for the effects of turbidity, high absorbance buffer solution and the use of reduced aperture cells. The instrument can use background correction.
- If it is used, there will be different results from those when unused, because Abs320 is subtracted from Abs260 and Abs280 prior to use in equations:
  Concentration = (Abs 260 - Abs 320) * Factor
  Abs ratio = (Abs 260 - Abs 320) / (Abs 280 - Abs 320)
  Abs ratio = (Abs 260 - Abs 320) / (Abs 230 - Abs 320)
- If your laboratory has not used background correction before, set this option to NO.
- The use of background correction can remove variability due to handling effects of low volume disposable cells.

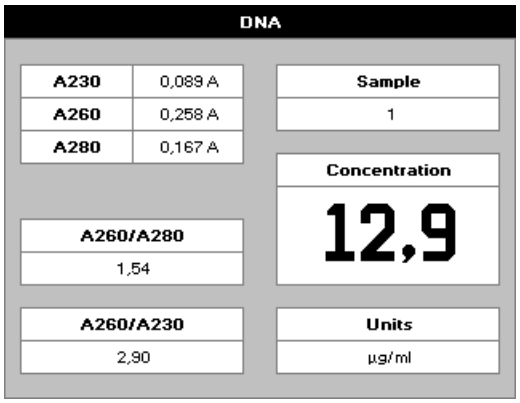
Note:

- absorbance maximum near 260 nm and absorbance minimum near 230 nm
- flat peak near 260 nm and steep slope at 280 nm
- very little absorbance at 320 nm

1: DNA
The procedure is as follows:



Step 1
Press 1 to select DNA mode.
Step 2
Select path length using the left and right arrows. Options are 1mm (tip) or 10 mm (cuvette).
Press the down arrow.
Step 3 (dilution factor known)
Enter the dilution factor using the keypad numbers. Range 1.00 to 9999. Use the C button to backspace and clear the last digit entered.
OR
Step 3 (calculate dilution factor)
Press Options to enter the dilution factor screen.
Enter the volume of the sample using the keypad numbers. Range 0.01 to 9999.
Press the down arrow.
Enter the volume of the diluent using the keypad numbers. Range 0.01 to 9999.
Press to calculate the dilution factor and return to the Parameters screen.
OR Press to cancel the selections and return to the Parameters screen.
Step 4
Select whether the background correction at 320 nm is used or not with the left and right arrows.
Press the down arrow.
Step 5
Select the units of measurement using the left and right arrows. Options: µg/ml, ng/µl, µg/µl.
Press the down arrow.
Step 6
Enter the factor using the keypad numbers. Default value is 50, range is 0.01 to 9999.
Step 7
Press OK to enter the Results screen
OR
Cancel to return to the Nucleic Acids folder

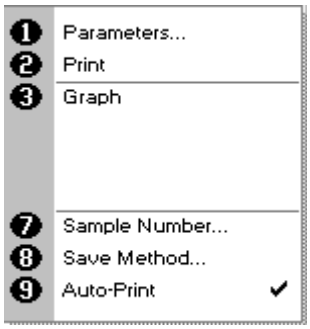


Results Screen
Step 8
Insert the reference. Press Blank Key. This will be used for all subsequent samples until changed.
Step 9
Insert sample and press . This measures at the selected wavelengths and displays the results. The ratio of wavelengths 1 and 2 absorbencies are calculated (both corrected by the background wavelength value if selected). Gives concentration based on absorbance at wavelength 1.

Repeat step 9 for all samples.

Press to return to the Nucleic acid folder.

Press Options to display available Options which are described below.

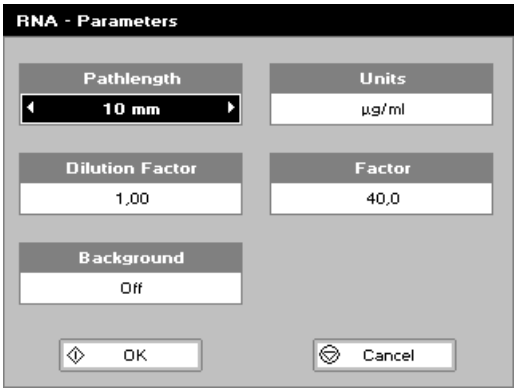


Options (select using key pad numbers)

1. Return to parameters screen (step 1 above).
2. Print result via selected method.
3. Toggle graph on/off. The graph shows a wavescan plot across the range 220 nm to 320 nm with cursors denoting 230, 260, 280 and (if background correction selected) 320 nm.
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  , or wait.

2: RNA
The procedure is as follows:



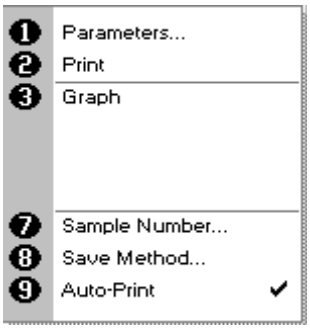
Step 1
Press 2 to select RNA mode.
Step 2
Select path length using the left and right arrows. Options are 1mm (tip) or 10 mm (cuvette).
Press the down arrow.
Step 3 (dilution factor known)
Enter the dilution factor using the keypad numbers. Range 1.00 to 9999. Use the C button to backspace and clear the last digit entered.
OR
Step 3 (calculate dilution factor)
Press Options to enter the dilution factor screen.
Enter the volume of the sample using the keypad numbers. Range 0.01 to 9999.
Press the down arrow.
Enter the volume of the diluent using the keypad numbers. Range 0.01 to 9999.
Press to calculate the dilution factor and return to the Parameters screen.
OR Press to cancel the selections and return to the Parameters screen.
Step 4
Select whether the background correction at 320 nm is used or not with the left and right arrows.
Press the down arrow.
Step 5
Select the units of measurement using the left and right arrows. Options: µg/ml, ng/µl, µg/µl.
Press the down arrow.
Step 6
Enter the factor using the keypad numbers. Default value is 40, range is 0.01 to 9999.
Step 7
Press OK to enter the Results screen
OR
Cancel to return to the Nucleic Acids folder

Results Screen
Step 8
Insert the reference. Press Blank Key. This will be used for all subsequent samples until changed.
Step 9
Insert sample and press . This measures at the selected wavelengths and displays the results. The ratio of wavelengths 1 and 2 absorbencies are calculated (both corrected by the background wavelength value if selected). Gives concentration based on absorbance at wavelength 1.

Repeat step 9 for all samples.

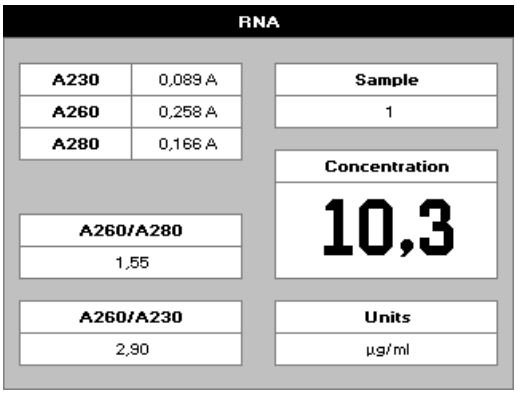
Press to return to the Nucleic acid folder.

Press Options to display available Options which are described below.



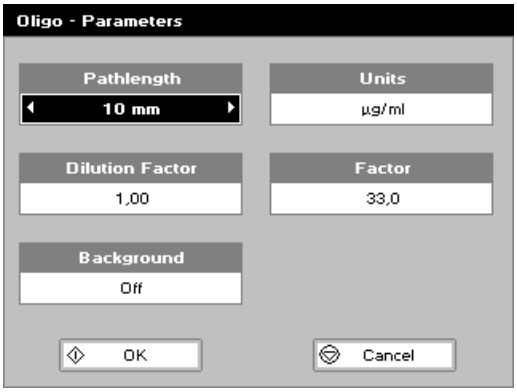
Options (select using key pad numbers)

1. Return to parameters screen (step 1 above).
2. Print result via selected method.
3. Toggle graph on/off. The graph shows a wavescan plot across the range 220 nm to 320 nm with cursors denoting 230, 260, 280 and (if background correction selected) 320 nm.
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.

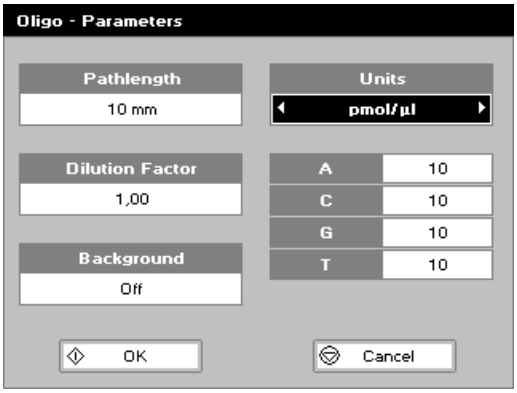


Exit options by pressing  , or wait.

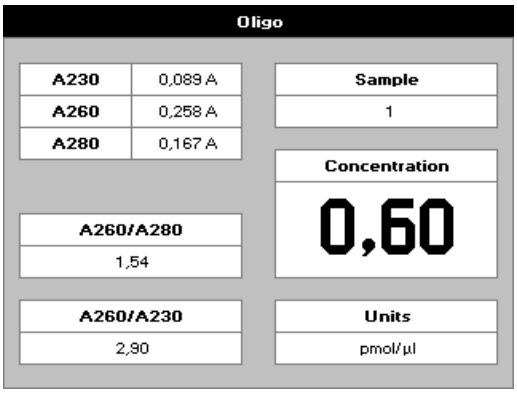
3: Oligo
The procedure is as follows:



Step 1
Press 3 to select Oligo mode.
Step 2
Select path length using the left and right arrows. Options are 1mm (tip) or 10 mm (cuvette).
Press the down arrow.
Step 3 (dilution factor known)
Enter the dilution factor using the keypad numbers. Range 1.00 to 9999. Use the C button to backspace and clear the last digit entered.
OR
Step 3 (calculate dilution factor)
Press Options to enter the dilution factor screen.
Enter the volume of the sample using the keypad numbers. Range 0.01 to 9999.
Press the down arrow.
Enter the volume of the diluent using the keypad numbers. Range 0.01 to 9999.
Press to calculate the dilution factor and return to the Parameters screen.
OR Press to cancel the selections and return to the Parameters screen.
Step 4
Select whether the background correction at 320 nm is used or not with the left and right arrows.
Press the down arrow.
Step 5
Select the units of measurement using the left and right arrows. Options: µg/ml, ng/µl, µg/µl and pmol/µl. If pmol/µl is selected the factor changes to a selection table denoting the ratios of the 4 bases in the structure.
Press the down arrow.



Step 6 (units not pmol/µl)
Enter the factor using the keypad numbers. Default value is 33, range is 0.01 to 9999.
OR
Step 6 (units pmol/µl)
Enter the proportions of bases present using the keypad numbers and up and down arrows to move between boxes. Default is 10 for each, range is 0 to 9999.
Step 7
Press OK to enter the Results screen
OR
Cancel to return to the Nucleic Acids folder



Results Screen
Step 8

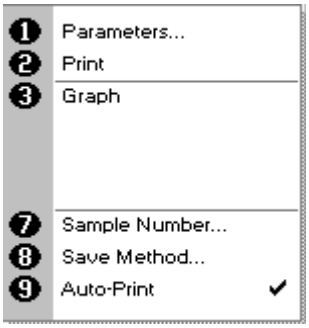
Insert the reference. Press Blank Key. This will be used for all subsequent samples until changed.

Step 9

Insert sample and press . This measures at the selected wavelengths and displays the results. The ratio of wavelengths 1 and 2 absorbencies are calculated (both corrected by the background wavelength value if selected). Gives concentration based on absorbance at wavelength 1.
Repeat step 9 for all samples.

Press to return to the Nucleic acid folder.

Press Options 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. The graph shows a wavescan plot across the range 220 nm to 320 nm with cursors denoting 230, 260, 280 and (if background correction selected) 320 nm.
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 , or wait.

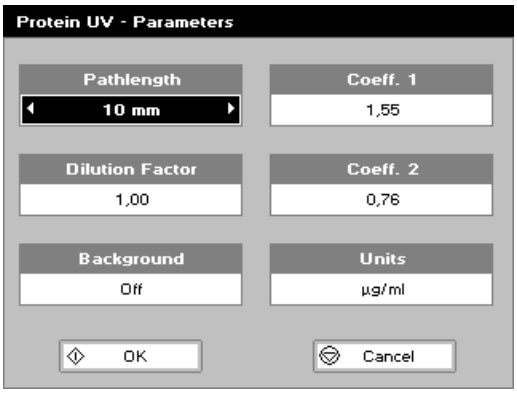
Protein Determination

Protein Determination at 280 nm

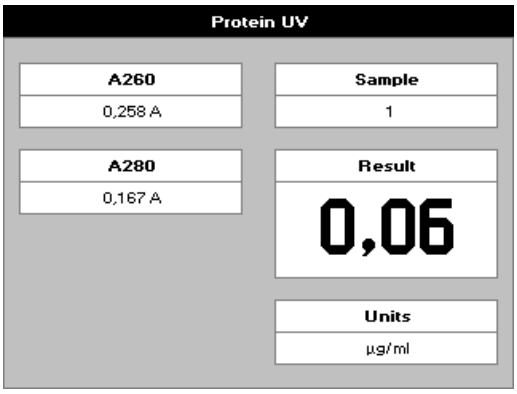
- Protein can be determined in the near UV at 280 nm due to absorption by tyrosine, tryptophan and phenylalanine amino acids; Abs 280 varies greatly for different proteins due to their amino acid content, and consequently the specific absorption value for a particular protein must be determined.
- The presence of nucleic acid in the protein solution can have a significant effect due to strong nucleotide absorbance at 280 nm. This can be compensated by measuring Abs 260, and applying the equation of Christian and Warburg for the protein crystalline yeast enolase (Biochemische Zeitung 310, 384 (1941)):
  Protein (mg/ml) = 1.55 * Abs 280 - 0.76 * Abs 260
  or, Protein conc. = (Factor 1 * Abs 280) - (Factor 2 * Abs 260)
- This equation can be applied to other proteins if the corresponding factors are known. The instrument can determine protein concentration at 280 nm and uses the above equation as default; the factors can be changed, and the use of background correction at 320 nm is optional.
- To customise the equation for a particular protein, the absorbance values at 260 and 280 nm should be determined at known protein concentrations to generate simple simultaneous equations; solving these provides the two coefficients. In cases where Factor 2 is found to be negative, it should be set to zero since it means there is no contribution to the protein concentration due to absorbance at 260 nm.
- Set Factor 2 = 0.00 for direct λ280 UV protein measurement; Factor 1 is based on the extinction coefficient of the protein. If BSA (bovine serum albumin) is an acceptable standard, setting Factor 1 = 1.115 will give linear results from 0 to 0.8 mg/ml protein.
  Protein (mg/ml) = 1.115 * Abs 280
- Rapid measurements such as this at Abs 280 are particularly useful after isolation of proteins and peptides from mixtures using spin and HiTrap columns by centrifuge and gravity, respectively.
  Protein Determination at 595, 546, 562 and 750 nm
- The Bradford method depends on quantitating the binding of a dye, Coomassie Brilliant Blue, to an unknown protein and comparing this binding to that of different, known concentrations of a standard protein at 595 nm; this is usually BSA, bovine serum albumin.
- The Biuret method depends on reaction between Cupric ions and peptide bonds in an alkali solution, resulting in the formation of a complex absorbing at 546 nm.
- The BCA method also depends on reaction between cupric ions and peptide bonds, but in addition combines this reaction with the detection of cuprous ions using bicinchoninic acid (BCA), giving an absorbance maximum at 562 nm. The BCA process is less sensitive to the presence of detergents used to break down cell walls.
- The Lowry method depends on quantifying the colour obtained from the reaction of Folin-Ciocalteu phenol reagent with the tylsryl residues of an unknown protein and comparing with those derived from a standard curve of a standard protein at 750 nm; this is usually BSA, bovine serum albumin
- Detailed protocols are supplied with these assay kits, and must be closely followed to ensure accurate results are obtained.
- The use of plastic disposable cells is recommended. To use a zero concentration standard include it in the number of standards to be entered and enter 0.00 for concentration; use this when required to enter standard 1.
- A linear regression analysis of the calibration standard data points is calculated; the result, together with the correlation coefficient, can be printed out. A correlation coefficient of between 0.95 and 1.00 indicates a good straight line.

1: Protein UV

This is the Christian and Warburg assay discussed previously. The procedure is as follows:



Step 1
Press 1 to select Protein UV mode.
Step 2
Select path length using the left and right arrows. Options are 1mm (tip) or 10 mm (cuvette).
Press the down arrow.
Step 3 (dilution factor known)
Enter the dilution factor using the keypad numbers. Range 1.00 to 9999. Use the C button to backspace and clear the last digit entered.
OR
Step 3 (calculate dilution factor)
Press Options to enter the dilution factor screen.
Enter the volume of the sample using the keypad numbers. Range 0.01 to 9999.
Press the down arrow.
Enter the volume of the diluent using the keypad numbers. Range 0.01 to 9999.
Press to calculate the dilution factor and return to the Parameters screen.
OR Press to cancel the selections and return to the Parameters screen.
Step 4
Select whether the background correction at 320 nm is used or not with the left and right arrows.
Press the down arrow.
Step 5
Enter co-efficient 1 (280 nm) using the keypad numbers. Default value is 1.55, range is 1.00 to 9999.
Press the down arrow.
Step 6
Enter co-efficient 2 (260 nm) using the keypad numbers. Default value is 0.76. Range is 1.00 to 9999.
Press the down arrow.
Step 7
Select the units of measurement using the left and right arrows. Options: µg/ml, ng/µl and µg/µl.
Step 8
Press OK to enter the Results screen
OR
Cancel to return to the Protein folder



Results Screen
Step 9

Insert the reference. Press Blank Key. This will be used for all subsequent samples until changed.

Step 10

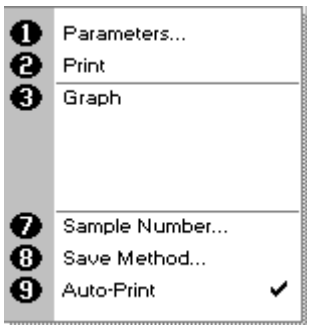
Insert sample and press . This measures at both 260 and 280 nm wavelengths and displays the result. Protein concentration is calculated (corrected by background wavelength value if selected)

.

Repeat step 10 for all samples.

Press to return to the Protein folder.

Press Options to display available Options which are described below.



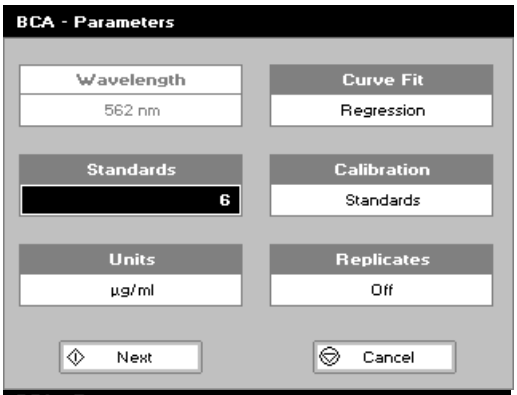
Options (select using key pad numbers)

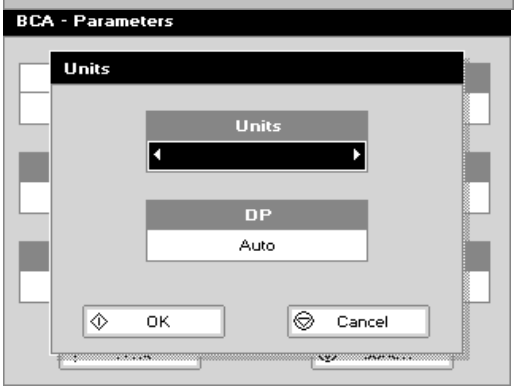
1. Return to parameters screen (step 1 above).
2. Print result via selected method.
3. Toggle graph on/off. The graph shows a wavescan plot across the range 250 nm to 330 nm with cursors denoting 230, 260, 280 and (if background correction selected) 320 nm.
7. Sample number – add a prefix to the sample 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.

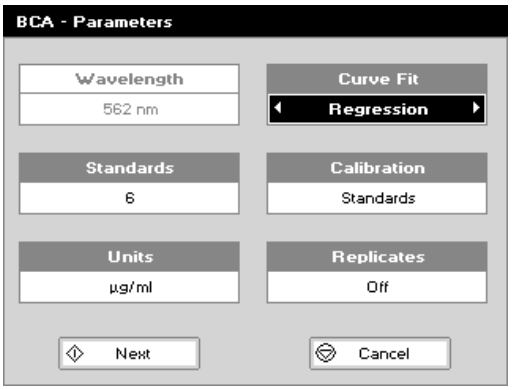
Exit options by pressing , or wait.

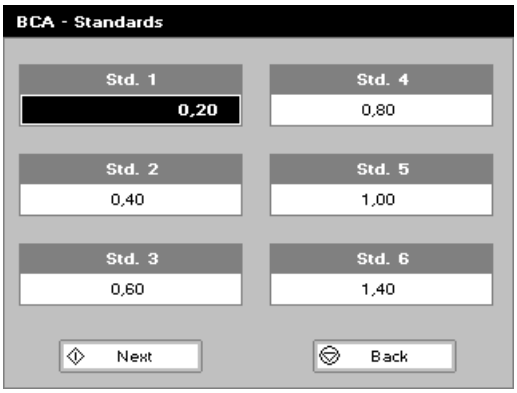
2: BCA

The procedure is as follows:









Step 1
Press 2 to select BCA mode.
Step 2
Wavelength for this stored method is pre-set to 562nm
Step 3
Enter the number of standard concentration points (1-9) to be used in the curve using the keypad numbers or left and right arrows.
Press the down arrow.

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 Options 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 5
Enter the type of curve fit. Options are straight line regression, zero regression (forces the straight line through the origin), interpolated or cubic spline.
Press the down arrow.
Step 6
Select the calibration mode, either standards (measure prepared standards) or manual (keypad data entry)
Step 7 (if standards selected)
Select the number of replicates using the left and right arrows. This determines the number of standards to be measured and averaged at each standard concentration point. Can be OFF (1), 2 or 3.
Step 8

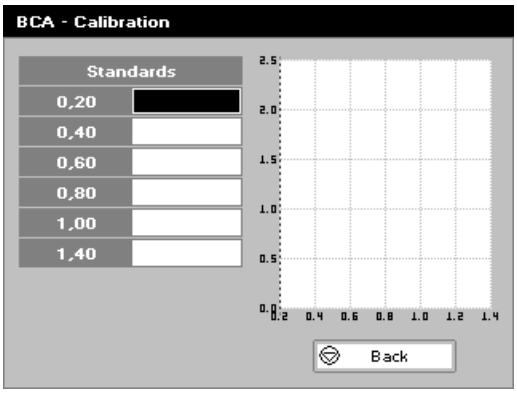
Press Next to enter the Standards screen
OR

Press Cancel to cancel selections and return to the Protein folder.

Standards Screen
Step 9
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. C button backspaces and clears the last digit entered.
Step 10

Press Next to enter the Calibration screen. If there are duplicate or non-monotonic (increasing) entries 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 11

Insert the reference sample. Press Blank key.
This will be used for all subsequent samples until changed.

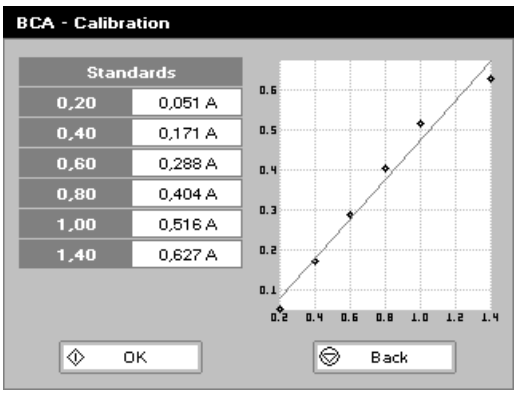
Step 12

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

Press to measure the standard and store the result.

Repeat step 12 for all standards. A graph will display the results and the fitted curve as the measurements are made.

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 13

When all standards are measured the OK box appears. Press to accept the calibration an go to the Results screen (see below)
OR

Press Back to cancel selections and return to the Standards screen.

Calibration Screen (replicates on)
This shows the calibration values and allows standards to be measured.

Step 11

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

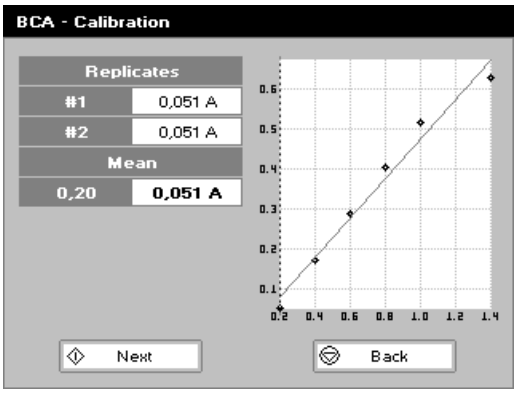
Step 12

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

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

Repeat for all replicates and standards.

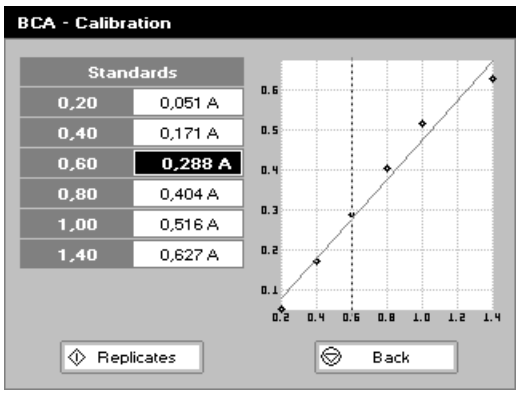
A graph will display the results and the fitted curve as the measurements are input.
Use the up and down arrows to select a standard to be repeated if a poor reading has been obtained. Use C to clear the previous reading.



Step 13

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

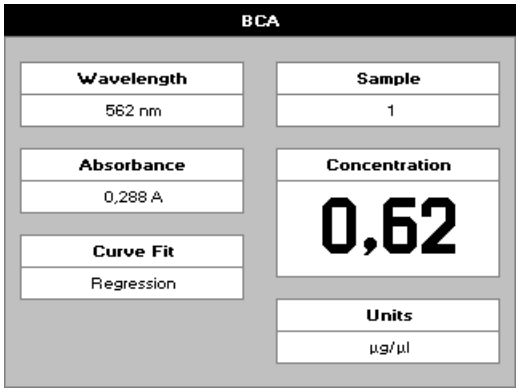
Press Back to return to the Standards screen.



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

Press Back to return to the Standards screen.



Results screen
Step 14

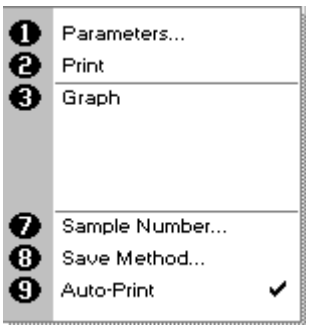
Insert the reference and press the Blank key. This will be used for all subsequent samples until changed.

Step 15

Insert the sample and press .
The concentration of the sample is taken and displayed.
Repeat step 15 for all samples.

Press to return to the Protein Folder.

Press Options to display available Options which are described below.



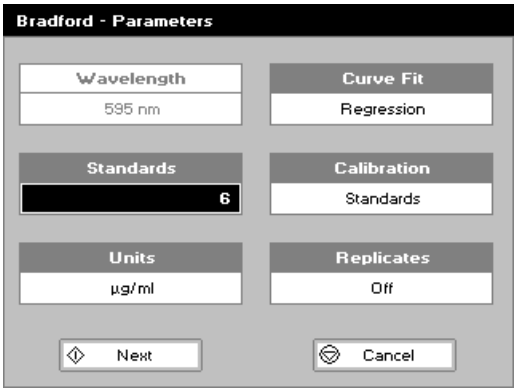
Options (select using key pad numbers)

1. Return to parameters screen (step 1 above).
2. Print result via selected method.
3. Toggle graph on/off. Displays the 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.

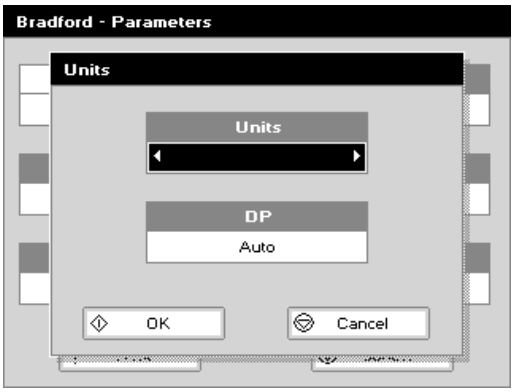
Exit options by pressing , or wait.

3: Bradford

The procedure is as follows:

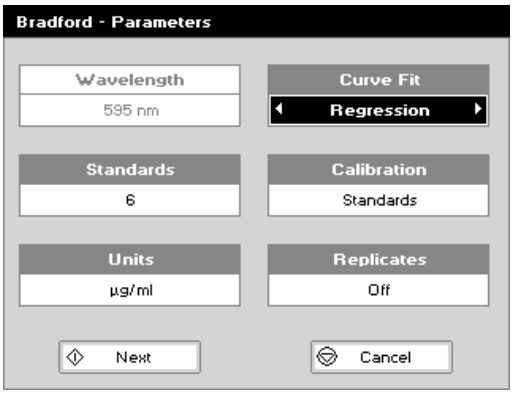


Step 1
Press 3 to select Bradford method.
Step 2
Wavelength for this stored method is pre-set to 595 nm.
Step 3
Enter the number of standard concentration points (1-9) to be used in the curve using the keypad numbers or left and right arrows.
Press the down arrow.



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 Options 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).

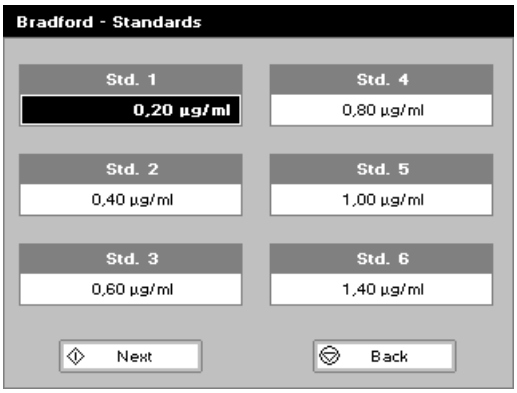
Press OK to store the chosen parameters or Cancel .



Step 5
Enter the type of curve fit. Options are: straight line regression, zero regression (forces the straight line through the origin), interpolated or cubic spline.
Press the down arrow.
Step 6
Select the calibration mode, either standards (measure prepared standards) or manual (keypad data entry)
Step 7 (if standards selected)
Select the number of replicates using the left and right arrows. This determines the number of standards to be measured and averaged at each standard concentration point. Can be OFF (1), 2 or 3.
Step 8

Press Next to enter the Standards screen
OR

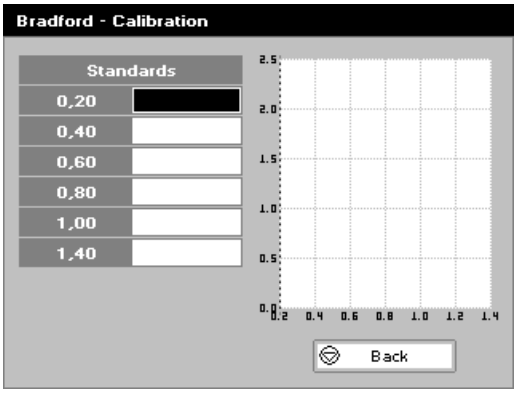
Press Cancel to cancel selections and return to the Protein folder.



Standards Screen
Step 9
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. C button backspaces and clears the last digit entered.
Step 10

Press Next to enter the Calibration screen. If there are duplicate or non-monotonic (increasing) entries 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 11
Insert the reference. Press 0A/100% key.
This will be used for all subsequent samples until changed.

Step 12
Insert the standard (use C to clear previously stored results before measuring)

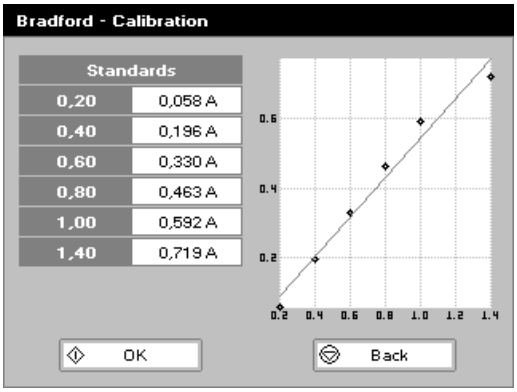
Press to measure the standard and store the result.

Repeat step 12 for all standards. A graph will display the results and the fitted curve as the measurements are made.

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 13
When all standards are measured the OK box appears. Press to accept the calibration an go to the Results screen (see below)
OR

Press Back to cancel selections and return to the Standards screen.



Calibration Screen (replicates on)
This shows the calibration values and allows standards to be measured.

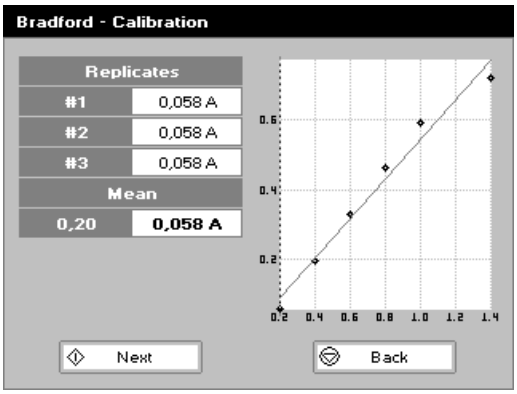
Step 11
Insert the reference. Press Blank key.
This will be used for all subsequent samples until changed.

Step 12
Press to display the replicate entry boxes. Use C to clear previously stored results before measuring.

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

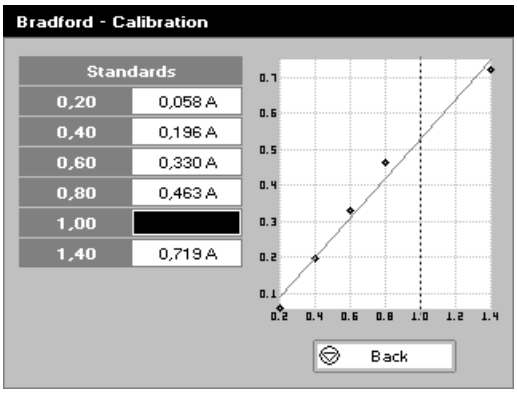
Repeat for all replicates and standards.

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 13
Press to accept the calibration and go to the Results screen (see below)
OR

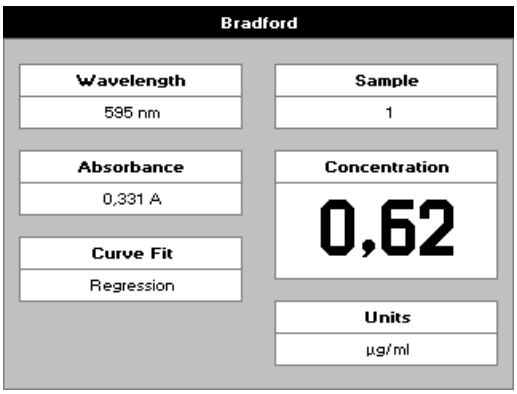
Press Back to return to the Standards screen.



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

Press Back to return to the Standards screen.



Results screen
Step 14

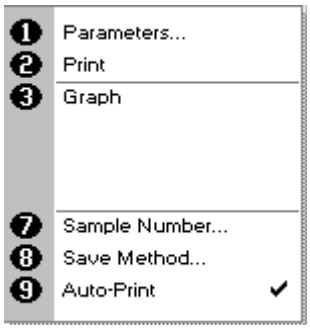
Insert the reference and press the Blank key. This will be used for all subsequent samples until changed.

Step 15

Insert the sample and press .
The concentration of the sample is taken and displayed.
Repeat step 15 for all samples.

Press to return to the Protein Folder.

Press Options to display available Options which are described below



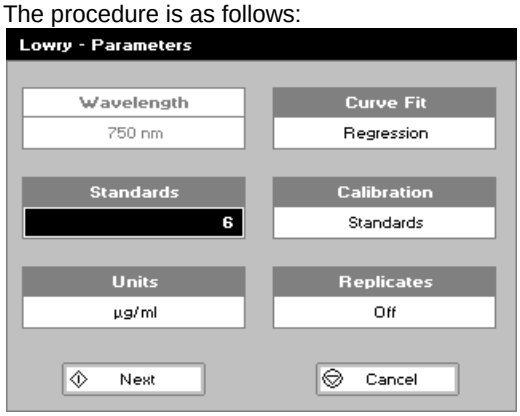
Options (select using key pad numbers)

1. Return to parameters screen (step 1 above).
2. Print result via selected method.
3. Toggle graph on/off. Displays the 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.

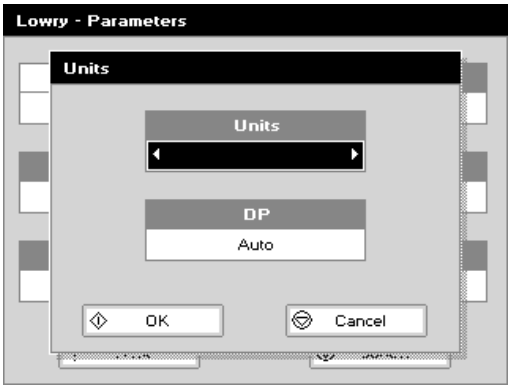
Exit options by pressing , or wait.

4: Lowry

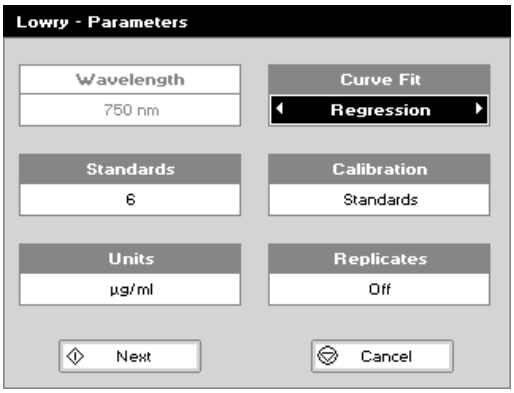
The procedure is as follows:



Step 1
Press 4 to select Lowry method.
Step 2
Wavelength for this stored method is pre-set to 750 nm.
Step 3
Enter the number of standard concentration points (1-9) to be used in the curve using the keypad numbers or left and right arrows.
Press the down arrow.



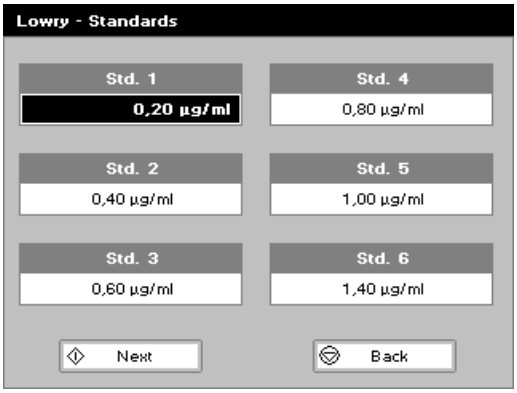
Step 4
Units: The user can enter a text string up to 8 characters long. To access a list of pre-defined units press the Options key Options and then use the left/right arrows (µg/ml, µg/µl, pmol/µl, mg/dl, mmol/l, µmol/l, g/l, mg/l, µg/l, U/l, %, ppm, ppb, conc or none). These units can also be edited 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 5
Enter the type of curve fit. Options are straight line regression, zero regression (forces the straight line through the origin), interpolated or cubic spline.
Press the down arrow.
Step 6
Select the calibration mode, either standards (measure prepared standards) or manual (keypad data entry)
Step 7 (if standards selected)
Select the number of replicates using the left and right arrows. This determines the number of standards to be measured and averaged at each standard concentration point. Can be OFF (1), 2 or 3.

Step 8 Press Next to enter the Standards screen
OR

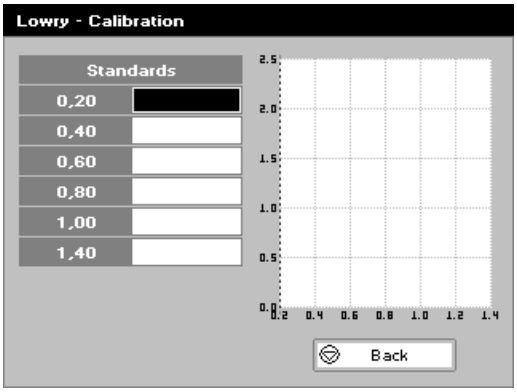
Press Cancel to cancel selections and return to the Protein folder.



Standards Screen
Step 9
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. C button backspaces and clears the last digit entered.
Step 10

Press Next to enter the Calibration screen. If there are duplicate or non-monotonic (increasing) entries 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 11

Insert the reference. Press Blank key.
This will be used for all subsequent samples until changed.

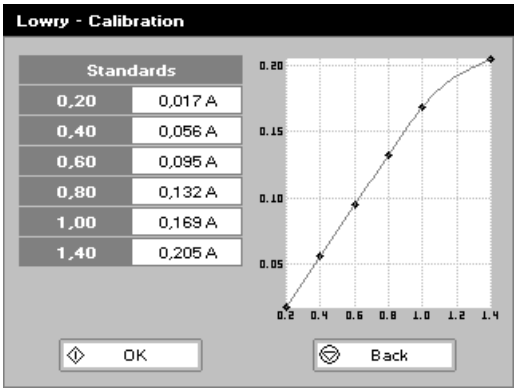
Step 12

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

Press to measure the standard and store the result.

Repeat step 12 for all standards. A graph will display the results and the fitted curve as the measurements are made.

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 13

When all standards are measured the OK box appears. Press to accept the calibration an go to the Results screen (see below)
OR

Press Back to cancel selections and return to the Standards screen.

Calibration Screen (replicates on)
This shows the calibration values and allows standards to be measured.

Step 11

Insert the reference sample. Press Blank key.
This will be used for all subsequent samples until changed.

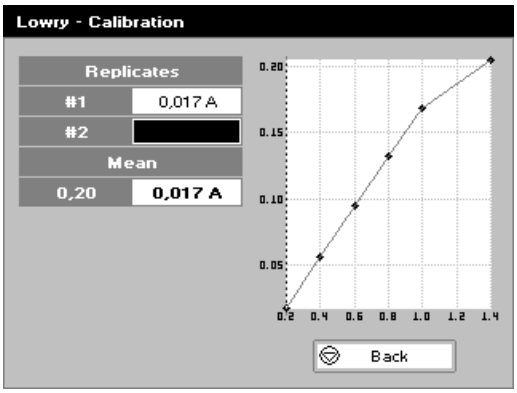
Step 12

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

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

Repeat for all replicates and standards.

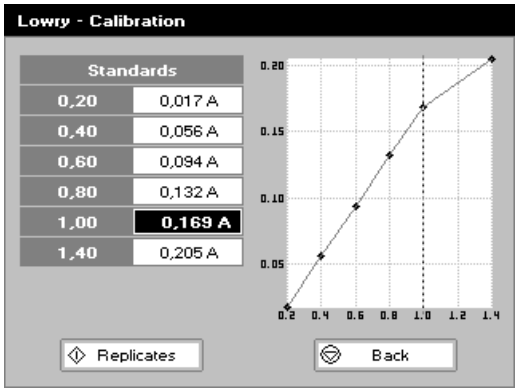
A graph will display the results and the fitted curve as the measurements are input.
Use the up and down arrows to select a standard to be repeated if a poor reading has been obtained. Use C to clear the previous reading.



Step 13

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

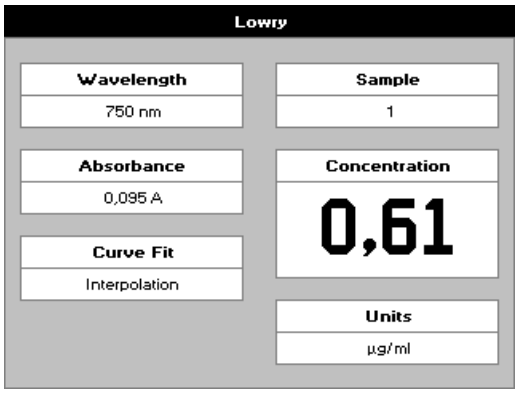
Press Back to return to the Standards screen.



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

Press Back to return to the Standards screen.



Results screen
Step 14

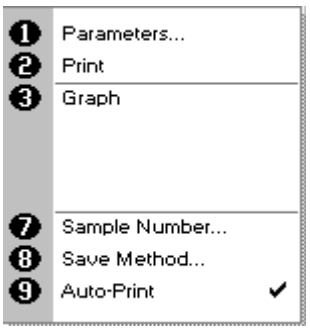
Insert the reference and press the Blank key. This will be used for all subsequent samples until changed.

Step 15

Insert the sample and press .
The concentration of the sample is taken and displayed.
Repeat step 15 for all samples.

Press to return to the Protein Folder.

Press Options to display available Options which are described below



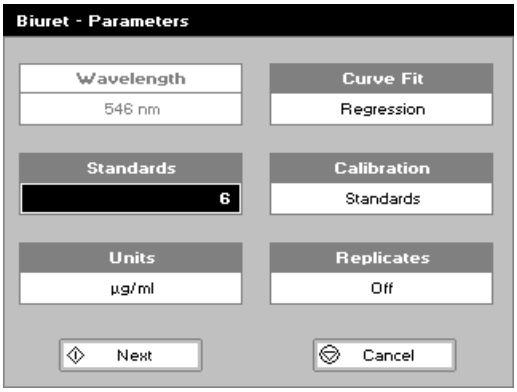
Options (select using key pad numbers)

1. Return to parameters screen (step 1 above).
2. Print result via selected method.
3. Toggle graph on/off. Displays the 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.

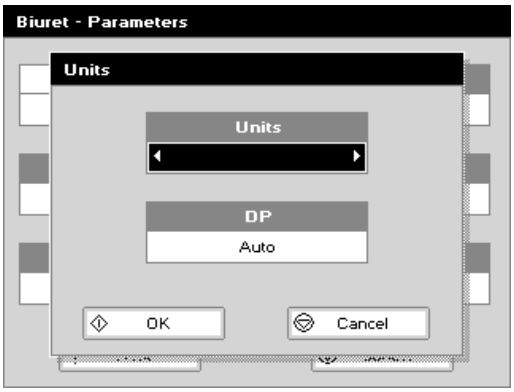
Exit options by pressing , or wait.

5: Biuret

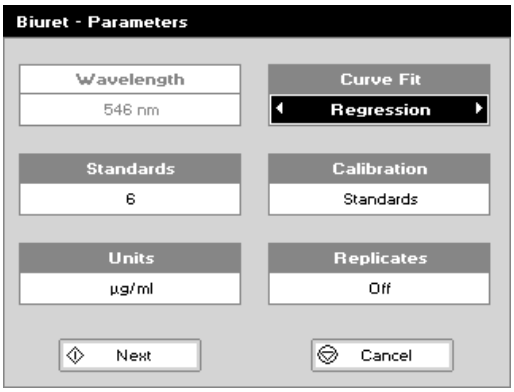
The procedure is as follows:



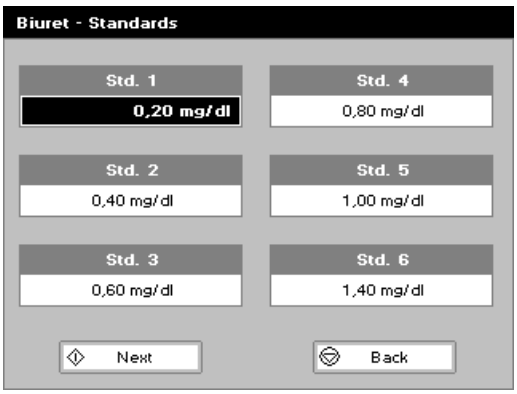
Step 1
Press 5 to select Biuret method.
Step 2
Wavelength for this stored method is pre-set to 546 nm.
Step 3
Enter the number of standard concentration points (1-9) to be used in the curve using the keypad numbers or left and right arrows.
Press the down arrow.



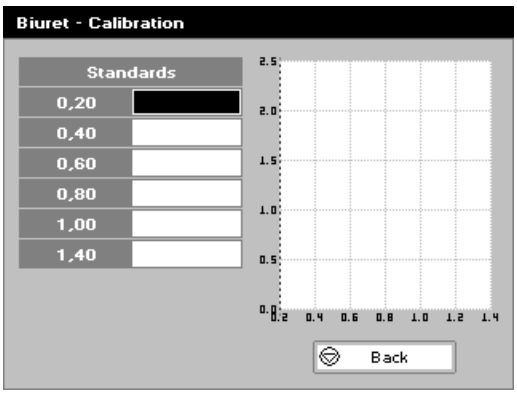
Step 4
Units: The user can enter a text string up to 8 characters long. To access a list of pre-defined units press the Options key Options and then use the left/right arrows (µg/ml, µg/µl, pmol/µl, mg/dl, mmol/l, µmol/l, g/l, mg/l, µg/l, U/l, %, ppm, ppb, conc or none). These units can also be edited 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 5
Enter the type of curve fit. Options are straight line regression, zero regression (forces the straight line through the origin), interpolated or cubic spline.
Press the down arrow.
Step 6
Select the calibration mode, either standards (measure prepared standards) or manual (keypad data entry)
Step 7 (if standards selected)
Select the number of replicates using the left and right arrows. This determines the number of standards to be measured and averaged at each standard concentration point. Can be OFF (1), 2 or 3.
Step 8
Press Next to enter the Standards screen
OR
Press Cancel to cancel selections and return to the Protein folder.



Standards Screen
Step 9
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. C button backspaces and clears the last digit entered.
Step 10
Press Next to enter the Calibration screen
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 11

Insert the reference. Press Blank key.
This will be used for all subsequent samples until changed.

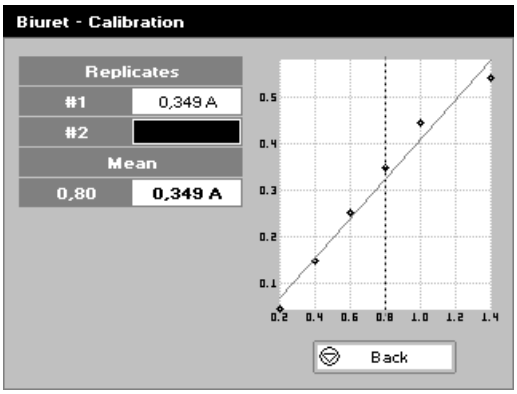
Step 12

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

Press to measure the standard and store the result.

Repeat step 12 for all standards. A graph will display the results and the fitted curve as the measurements are made.

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 13

When all standards are measured the OK box appears. Press to accept the calibration an go to the Results screen (see below)
OR

Press Back to cancel selections and return to the Standards screen.

Calibration Screen (replicates on)
This shows the calibration values and allows standards to be measured.

Step 11

Insert the reference. Press Blank key.
This will be used for all subsequent samples until changed.

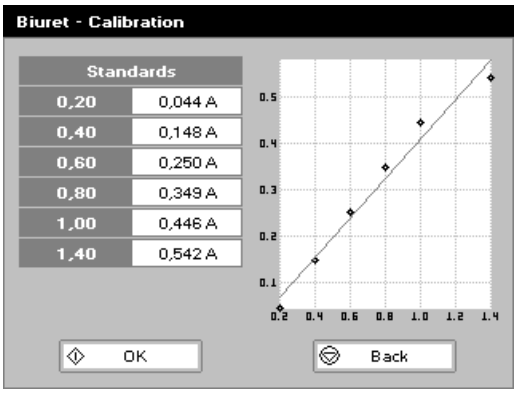
Step 12

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

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

Repeat for all replicates and standards.

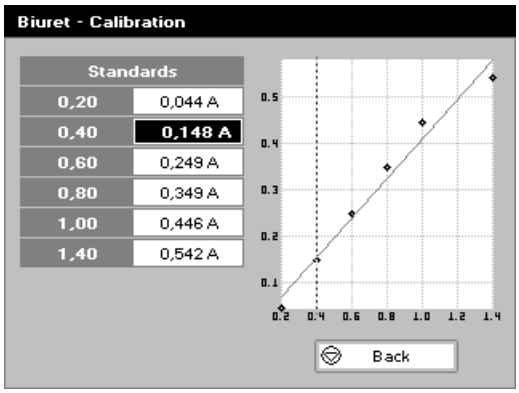
A graph will display the results and the fitted curve as the measurements are input.
Use the up and down arrows to select a standard to be repeated if a poor reading has been obtained. Use C to clear the previous reading.



Step 13

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

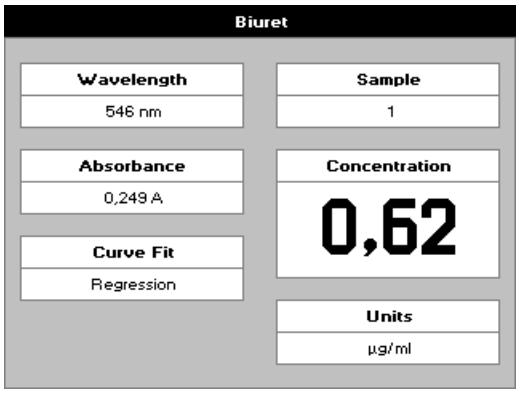
Press Back to return to the Standards screen.



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

Press Back to return to the Standards screen.



Results screen
Step 14

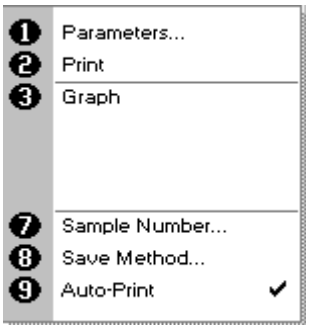
Insert the reference and press the Blank key. This will be used for all subsequent samples until changed.

Step 15

Insert the sample and press .
The concentration of the sample is taken and displayed.
Repeat step 15 for all samples.

Press to return to the Protein Folder.

Press Options to display available Options which are described below



Options (select using key pad numbers)

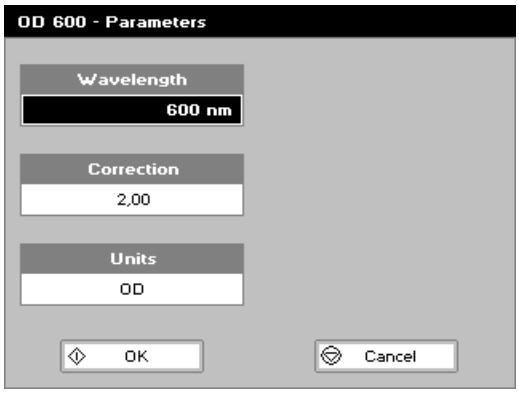
1. Return to parameters screen (step 1 above).
2. Print result via selected method.
3. Toggle graph on/off. Displays the calibration graph, cursors give values for last measured sample.
7. Sample number – add a prefix to the sample number and reset the incrementing number to the desired value.
8. Save 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 , or wait.

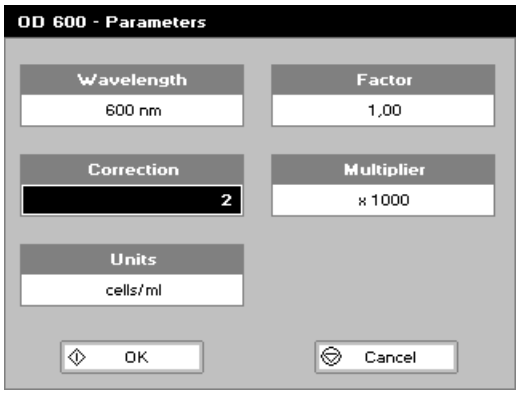
Bacterial Cell Culture Measurement (OD600)

- Bacterial cell cultures are routinely grown until the absorbance at 600 nm (known as OD600) reaches approximately 0.4 prior to induction or harvesting. A linear relationship exists between cell number (density) and OD 600 up to approx. 0.6.
- It is important to note that for turbid samples such as cell cultures, the absorbance measured is due to light scattering, and **not** the result of molecular absorption. The amount of scatter is affected by the optics of the system (distance between the cell holder and instrument exit slit, geometry of this slit and the monochromator optics). Different spectrophotometer types therefore give different responses for the same turbid sample; to compare results, they must be normalised using calibration curves.
- A calibration curve can be determined by comparing measured OD 600 to expected OD 600. Expected OD 600 is determined by counting cell number using an alternative technique (for example microscope slide method) and converting to OD 600 using the rule of thumb that 1 OD 600 = 8 x 10^8 cells/ml for E. Coli.
- Your Piccodrop instrument has much smaller optics than most conventional spectrophotometers, and more light is transmitted through to the detector resulting in lower than expected OD 600 values. Results obtained by comparing measured OD 600 with expected OD 600 (see above) indicate that a correction factor of 2.0 is required to make the data comparable to larger instruments; this factor is included as a default value in set up.
- The use of 10 mm pathlength disposable cells is recommended for optical density measurements of cell culture solutions; to prevent the suspension settling too quickly and giving an OD that changes with time, glycerol should be added to the sample.

The procedure is as follows:



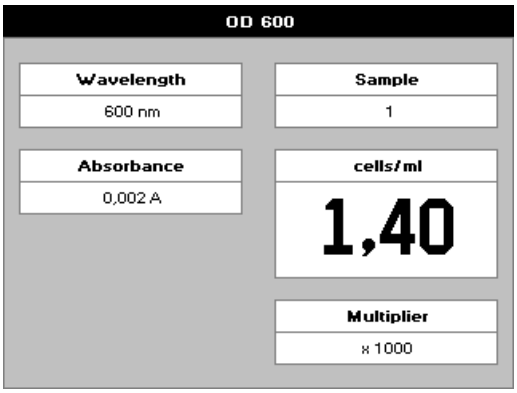
Step 1
Select the wavelength. Default value is 600 nm.
Press the down arrow.
Step 2
Enter the factor to compensate for different optical configurations between this and other instruments. Default value is 2.
Press the down arrow.
Step 3
Select the units. Options are OD or cells/ml. If cells/ml is selected two further parameters are displayed.



Step 4 (if cells/ml selected)
Enter the factor using the keypad numbers. Range 0.00 to 9999.
C button backspaces and clears the last digit entered.
Press the down arrow.
Step 5 (if cells/ml selected)
Select the multiplier using the left and right arrows. Options are 1000 or 1,000,000.
Step 6

Press OK to enter the Results screen
OR

Press Cancel to cancel selections and return to the Life Science folder.



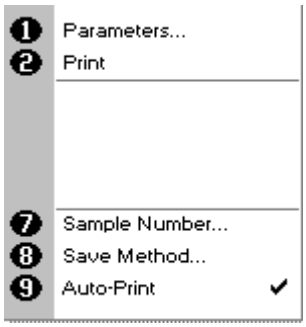
Results Screen
Step 8
Insert the reference and press the 0A/100%T key. This will be used for all subsequent samples until changed.
Step 9

Insert the sample and press  .
The wavelength, absorbance and OD600 value is displayed.

Repeat step 9 for all samples.

Press  to return to the Life Science Folder.

Press  to display available Options which are described below.



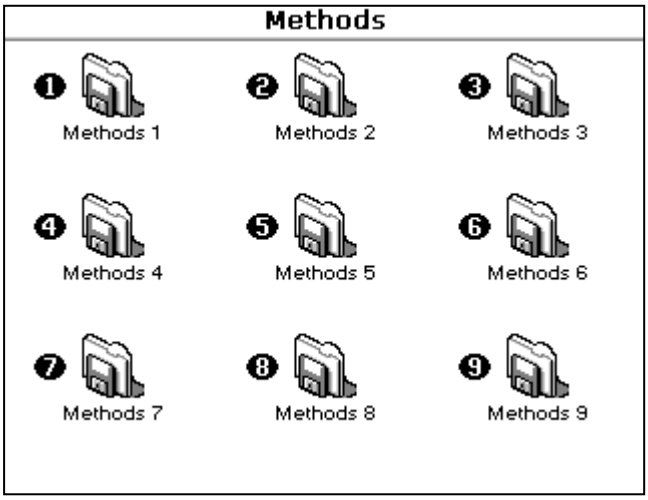
Options (select using key pad numbers)
1. Return to parameters screen (step 1 above).
2. Print result via selected method.
7. Sample number – add a prefix to the sample number and reset the incrementing number to the desired value.
8. Save method – use the left and right arrows to select a folder to 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  , or wait.

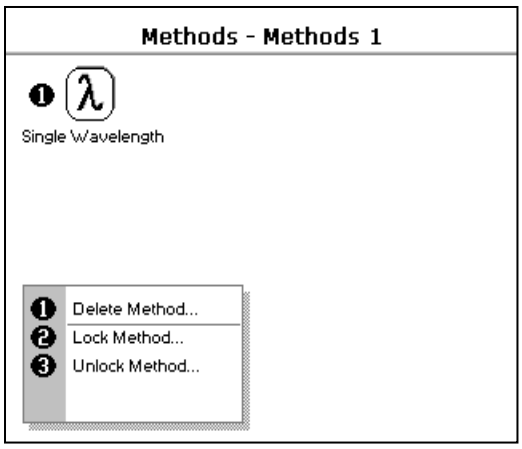
FAVOURITES AND METHODS FOLDERS

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 Options key.

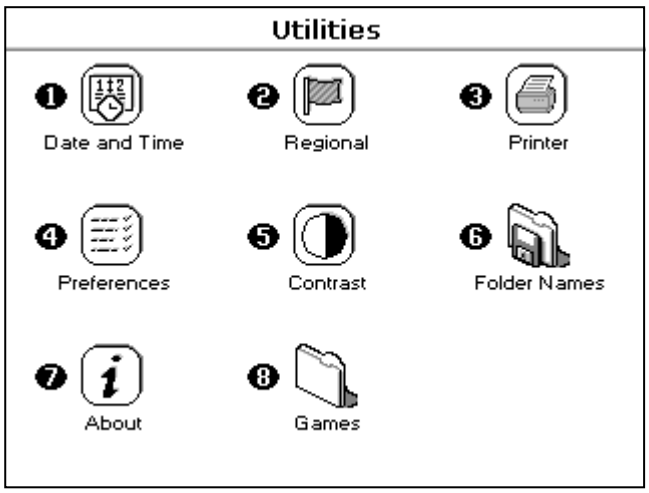


Delete Method
Press 1 to select delete method.
Select the method to be deleted using the left and right arrows.
Press to delete the method
OR cancel to return to Favourites/Methods folder.

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 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 to unlock the method
OR cancel to return to the Favourites/Methods folder.

UTILITIES FOLDER

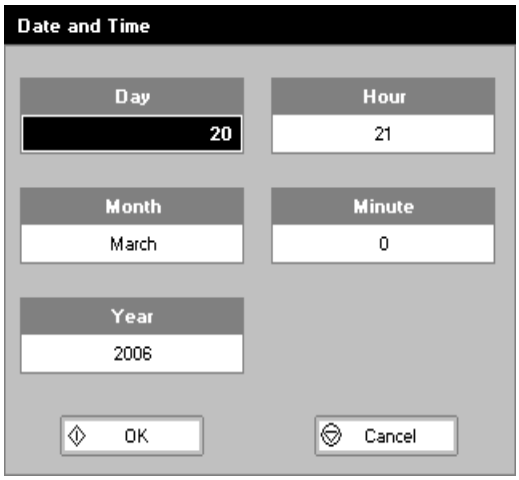


Summary Function	Keypad number	Description
1 Date and Time	1	Set correct time and date
2 Regional	2	Select preferred language and number format
3 Printer	3	Printer/output options
4 Preferences	4	Select screen layout (themes) and history
5 Contrast	5	Adjust screen contrast & brightness
6 Folder Names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

1: Date and Time

The procedure is as follows:

2: Regional

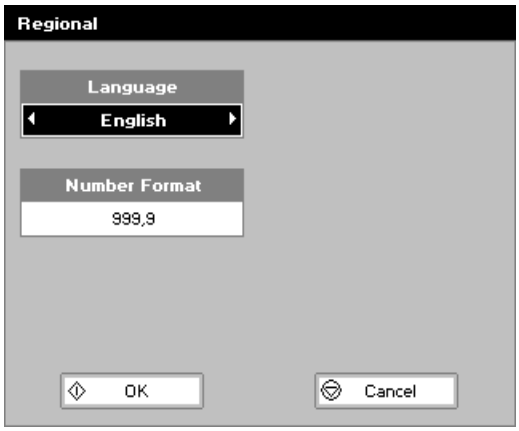


Enter the day using the keypad numbers or left and right arrows.
Press the down arrow.
Enter the month as above.
Press the down arrow.
Enter the year.
Press the down arrow.
Enter the hour.
Press the down arrow
Enter the minute. Seconds are zeroed when OK is pressed.

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

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

Sets Language and Number Format
The procedure is as follows:



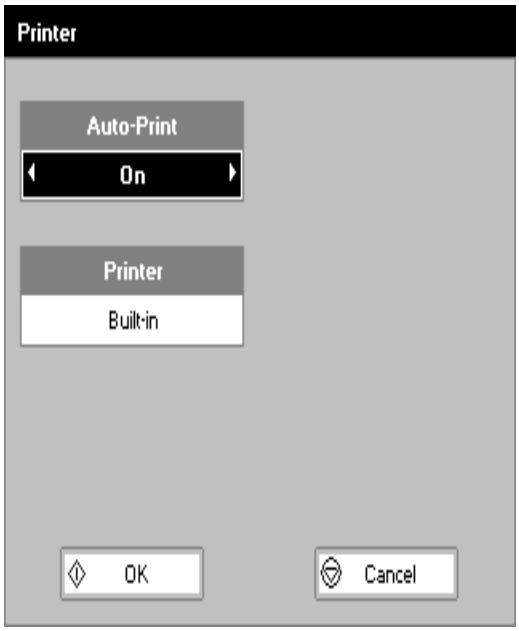
Select a language. Options are French, English, or Spanish. (German and Italian will be released in the near future).
Press the down arrow.
Set the decimal point style. Options are "," or ".".

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

Press Cancel 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 Options 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.

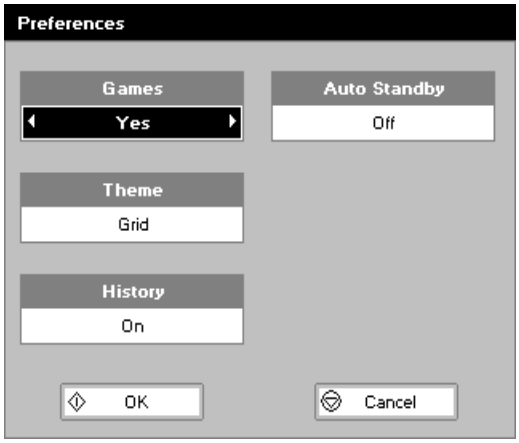
Press OK to store the settings and return to the Utilities folder
OR

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

4: Preferences

Sets user preferences
The procedure is as follows:

5: Contrast

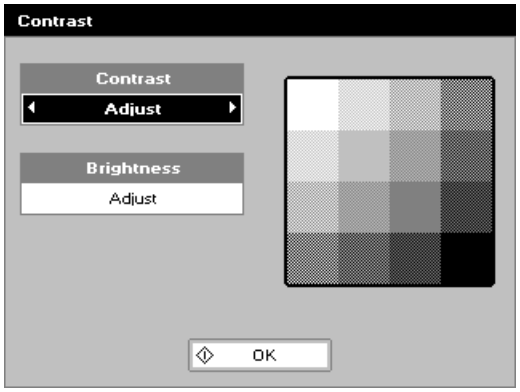


Select games function. This determines whether the games folder is displayed or not. Options are yes or no.
Press the down arrow.
Define the screen layout of folders. Options are either a grid format (default) or a list.
Press the down arrow.
Select whether to use previously entered parameters on switch 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 to store the settings and return to the Utilities folder
OR

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

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

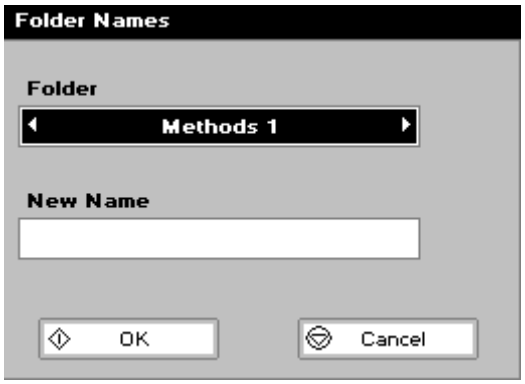


Adjust the contrast using the left and right arrows.
Press the down arrow.
Adjust the brightness using the left and right arrows.
Press the down arrow.

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

6: Folder Names

This folder allows you to rename the method or favourite folders

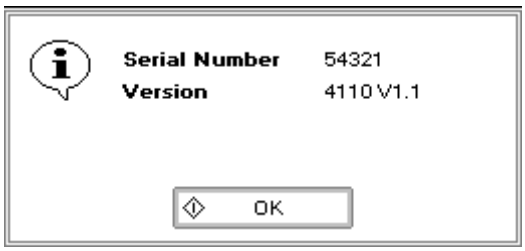


Select the folder you wish to rename using the left and right arrows.
Press the down arrow.
Input the new name for the folder.

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

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

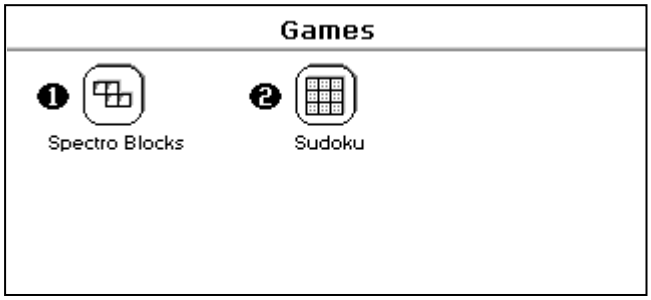
7: About



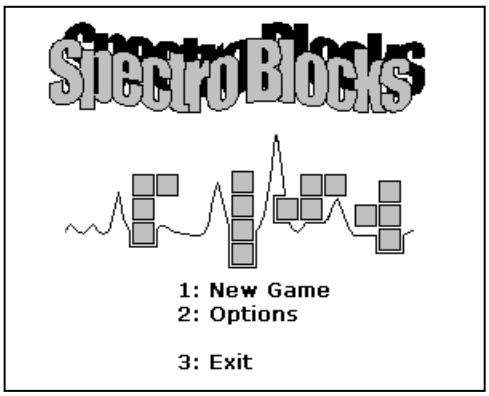
Displays the instrument serial number and software version.

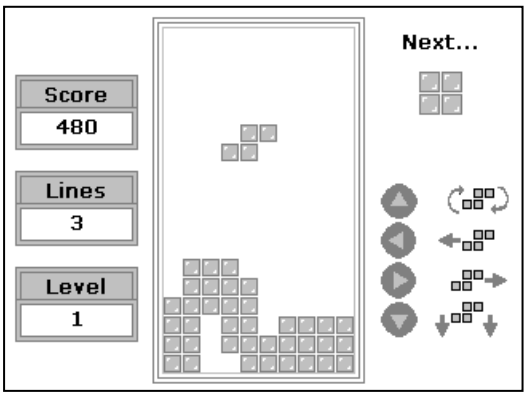
Press OK to close the window and return to the Utilities folder

8: Games



1: Spectroblocks





Classic block dropping game. Follow the instructions!

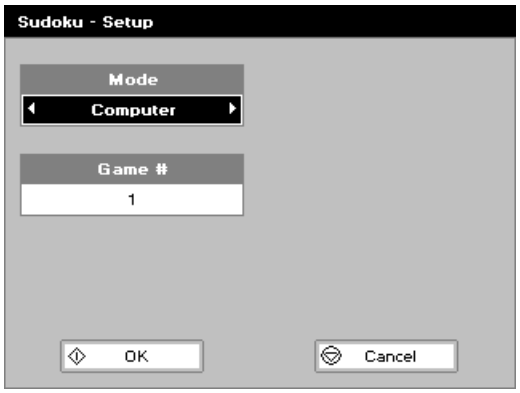
Press Cancel to return to the Utilities folder without storing the settings.

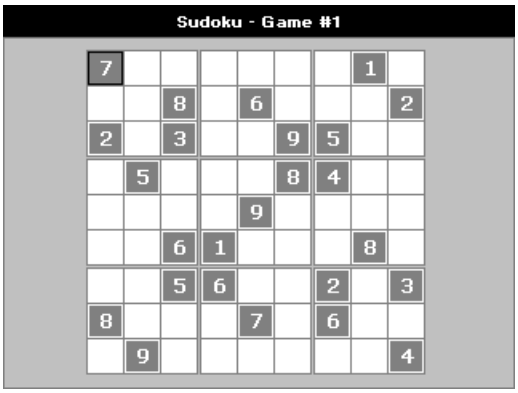
2: Su Doku

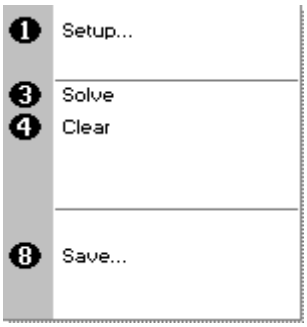
Can be set up as Computer mode (50 preset games) or User mode (enter your own pattern)

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

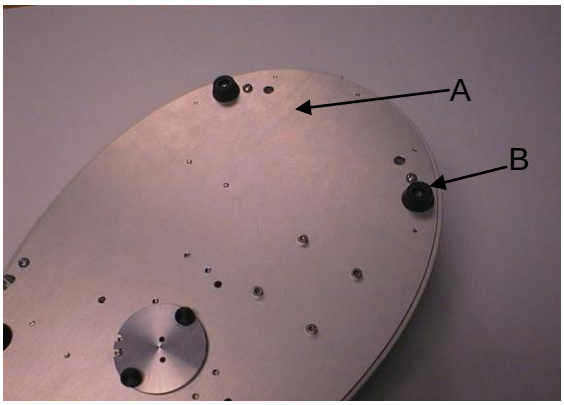
Press Options to display the options menu

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

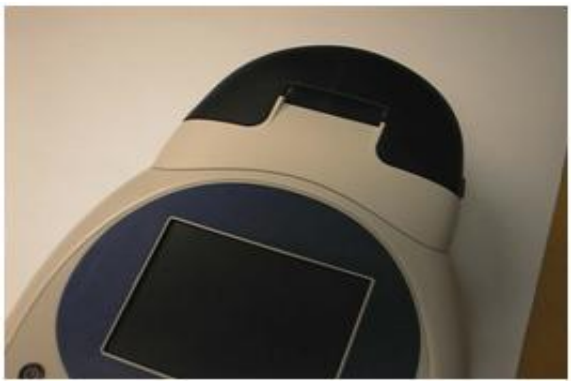


1. REMOVE THE POWER CABLE FROM THE INSTRUMENT. Turn the instrument over and remove cap head screws from positions A and B using the Allen key provided.

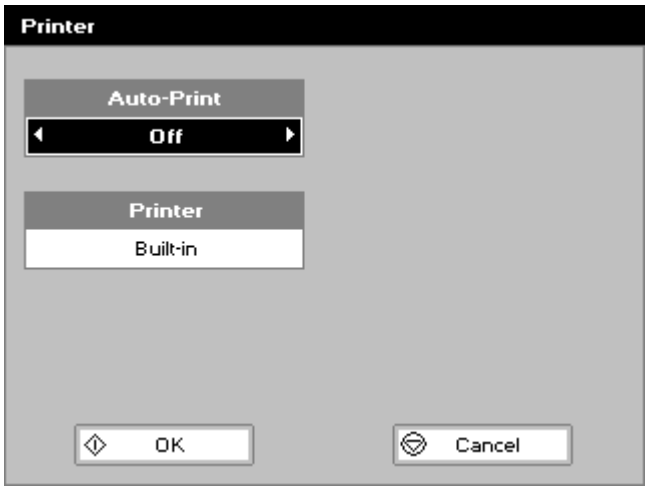
2. Turn the instrument back over and lift the accessory cover vertically upwards to remove. Remove the tie-wrap from the cable.

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

4. Plug the accessory cable into the printer.

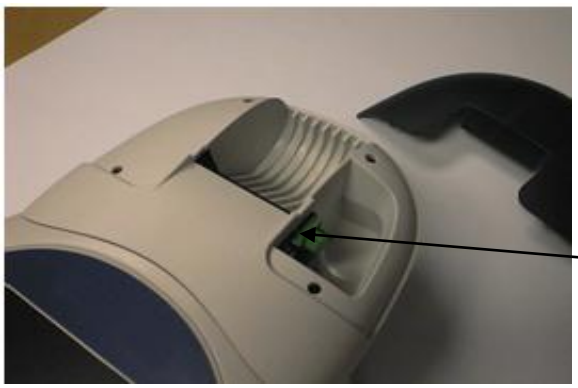


5. Lower the printer onto the locating bosses and push down firmly.



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

Loading / changing the printer paper

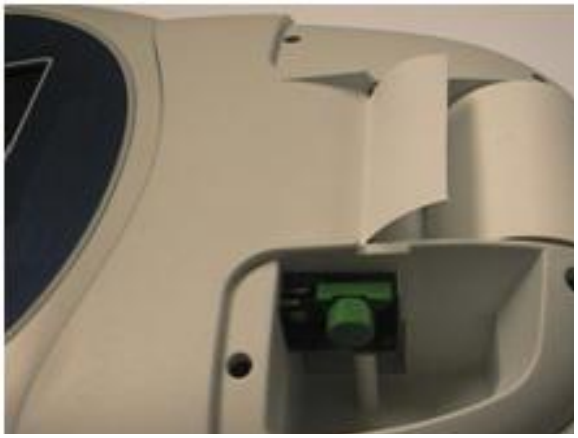


1. Lift off the paper cover.

Lock the platen and turn the knob to feed the paper

2. Feed in the paper.

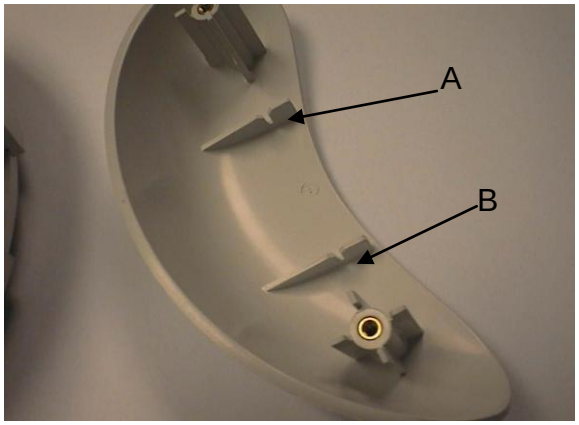
Sometimes it helps if the platen lock is released.



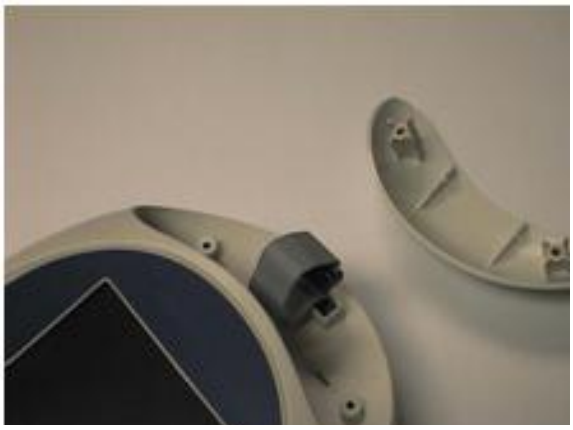
3. Paper gripped.

4. Replace Cover

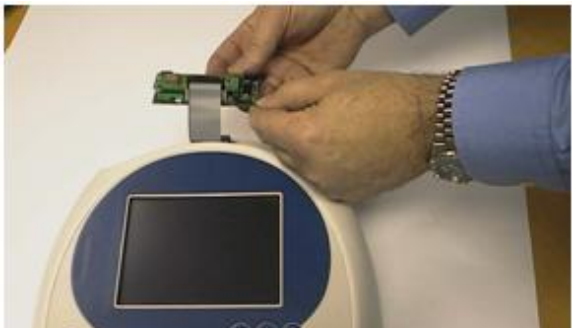
Changing to the Bluetooth Accessory



1. 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.

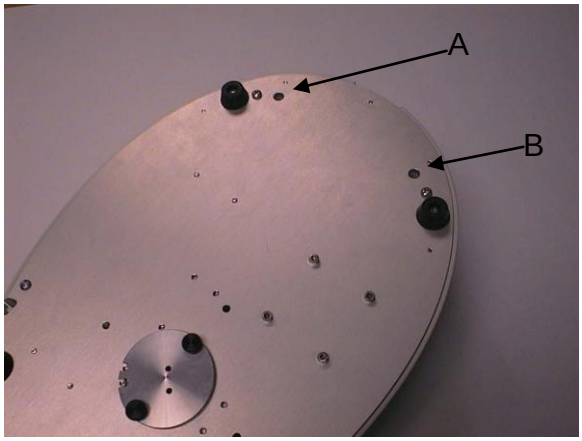


2. Turn the instrument back over and lift the accessory cover vertically upwards to remove. Remove the tie-wrap from the cable



3. Plug the accessory cable into the Bluetooth module.

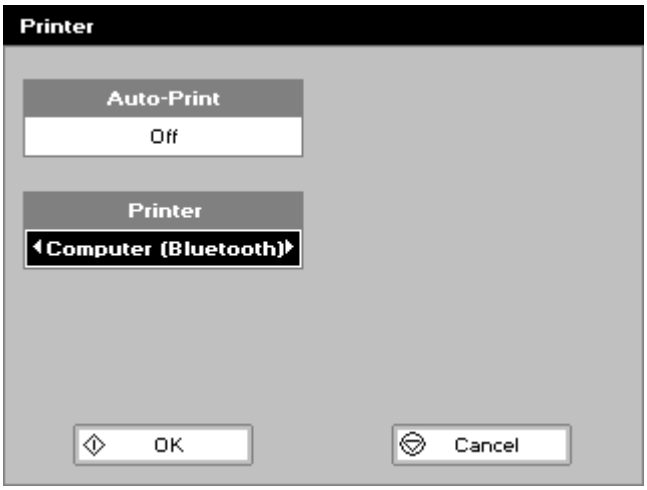
4. Note the slots in the base of the case. The two lugs on the Bluetooth module plug into these



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.



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

SD MEMORY CARD ACCESSORY

The SD memory card accessory can be fitted to a Pico200 spectrophotometer for the following operations:

- To export data from the instrument for loading into a PC
- To save methods
- To create a backup of all instrument methods for future restore
- To backup a specific instrument and restore these methods to other instruments in order to allow “cloning”

The SD memory card accessory is compatible with the following types of storage media

- SD memory cards
- SDHC memory cards

Data is stored on the SD memory card in a proprietary format with a *.pvc file extension and this data can only be accessed the Print Via Computer (PVC) software supplied with the accessory. This software should be installed onto a PC as detailed in the PVC user manual stored on the PVC CD.

Installation

Unpacking

- Remove the accessory from its packaging and inspect it for signs of damage.
- Within the SD memory card accessory you will have the following:
    - SD memory card PCB Module
    - New accessory cover
    - SD memory card
    - PVC (Print Via Computer) software CD
    - Allen key.
- If there are any signs of damage to the accessory of if any of the above components are missing, please contact your supplier immediately.

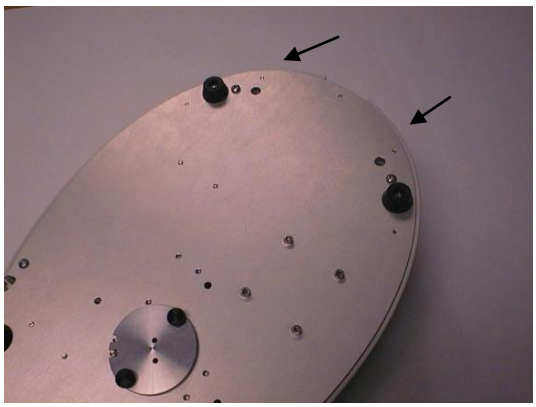
Safety

Read the safety instructions in the relevant instrument user manual. Disconnect all power from the instrument before fitting the SD memory card accessory.

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

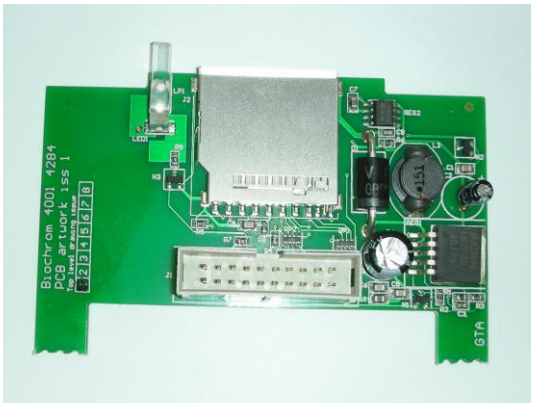
There are no user-serviceable parts inside this accessory.

Fitting SD memory card accessory

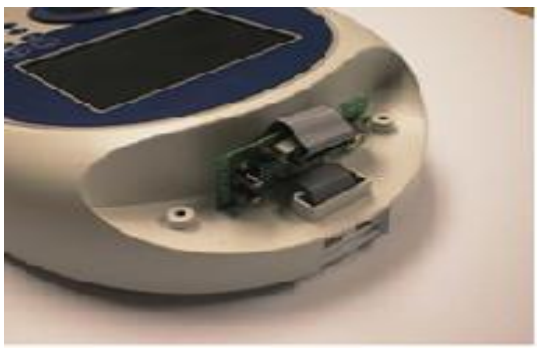


Step 1.
Remove the power cable from the instrument. Turn the instrument over and place onto a soft surface, for example a folded up towel, remove cap head screws from the positions indicated using the Allen key provided.

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



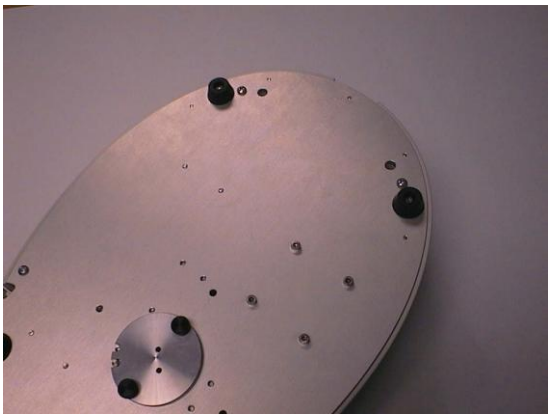
Step 3.
Plug the accessory cable into the SD memory card PCB module.



Step 4.
Note the slots in the base of the case. The two lugs on the SD memory card PCB module plug into these

Step 5.
Note the slots in the accessory cover supplied; these are, designed to engage with the SD memory card PCB module.

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

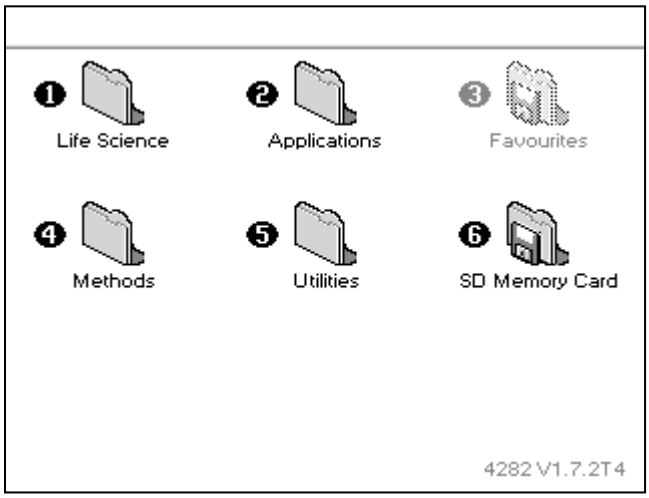


Step 7.
Invert the instrument and replace the cap head screws using the Allen key provided.

The accessory is now ready for use.

OPERATION

SD memory cards are inserted into the accessory with the contacts facing towards the user and the cut out corner on the right hand side (i.e. downwards). When a compatible SD card is inserted into the accessory, the red light flashes momentarily and the SD memory card icon appears on the instrument home page;



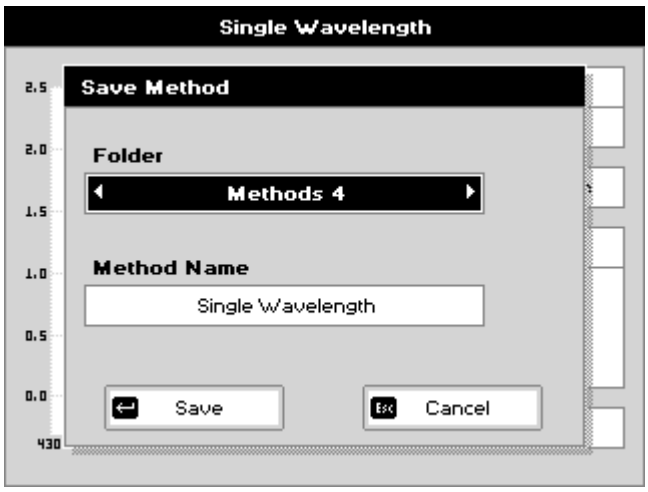
Saving methods to SD memory card

When an SD memory card is inserted into the accessory it is possible to save methods directly to the card.

Methods are stored on the card in a directory named ***Instrument Type*\Methods** (where instrument type will be dependant upon instrument being used)**;** this directory structure is evident when the SD card is connected to a PC.

To save a method to the SD memory card, the instructions for the relevant application from the instrument user manual must be followed. Typically:

- press the Options button (or relevant numerical short cut)
- press Save Method
- use the right and left arrows to select the folder on the SD memory card to which you wish to save the method
- change the filename if required
- press Save.



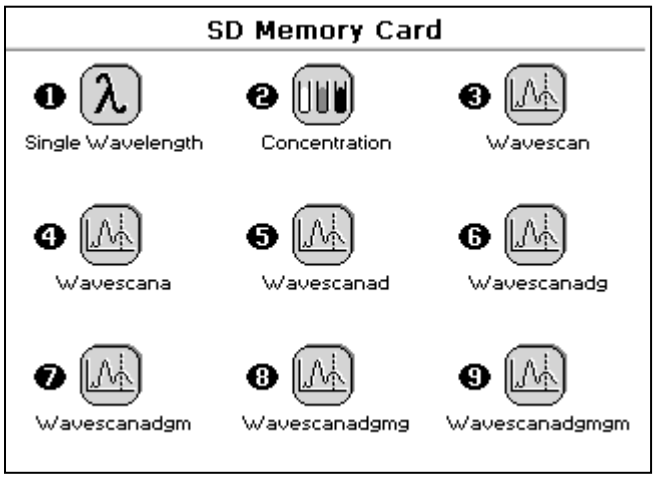
NOTE: a maximum of 9 methods can be stored in the SD memory card folder and in the *Instrument Type*\Methods directory.

These stored methods can also be opened on different instruments and then stored into other method folders if required.

When a method is being stored the LED next to the card will light up, the card **MUST** not be removed whilst the light is on otherwise the stored method will be corrupted.

Loading methods from SD memory card

Selecting the SD memory card by pressing the relevant number on the home page shows the methods stored on the card

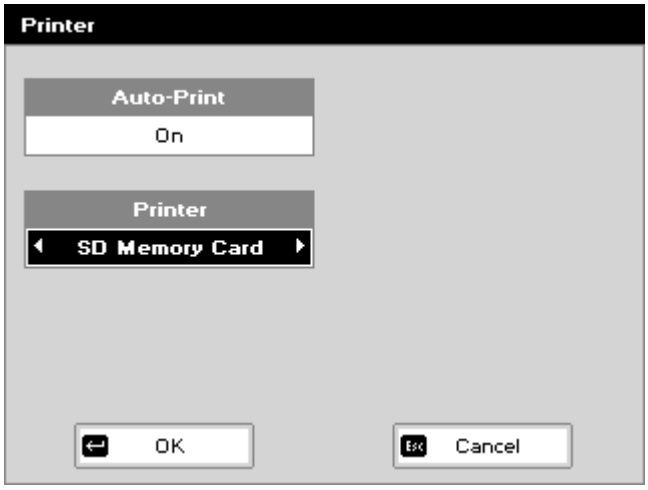


The required method can be loaded by pressing the relevant number on the keyboard and run in the same way as methods stored in any of the method folders on the instrument.

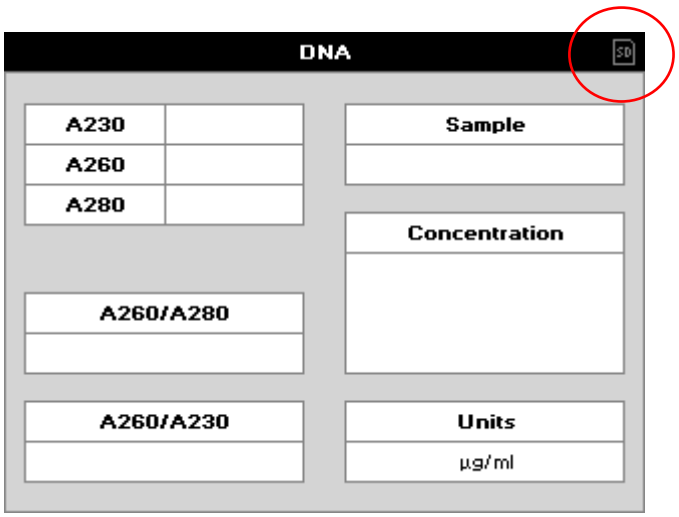
Saving data to SD memory card

Data from all applications on your instrument can be stored onto the SD memory card.

To enable data to be stored on the card, the SD memory card must be selected as the output device; to do this, **select utilities\printer** and under printer select SD memory card and ensure Auto-Print is selected.



Start an application or load a method in the usual way. Note that when SD memory card and Auto-Print are both selected in utilities, an SD icon appears in the top right hand corner of the display.



For applications that print continuously, **such as Single Wavelength or Nucleic Acids**, the LED next to the SD memory card will stay on continuously until the complete set of results is finished. To close the results file the application should be exited using the ESC key in the usual way. Removing the card whilst the LED is on will corrupt the collected data set. All results are stored as an individual file in a directory called ***Serial no*\PVC** on the SD memory card; this directory structure is evident when the SD card is connected to a PC.

Results are identified by the format of the filename; application type followed by an incrementing file ID. For example

DNA-A001.PVC for a DNA file
BCA001.PVC for a BCA Protein file

For applications that print whole documents in one go, such as **Wavescan or Kinetics**, the LED next to the SD memory card will go off after saving each measurement. In this case the card can be removed, when the light is off, without leaving the application. Each result is stored as an individual file in a directory called ***Serial no*\PVC** on the SD memory Card; this directory structure is evident when the SD card is connected to a PC.

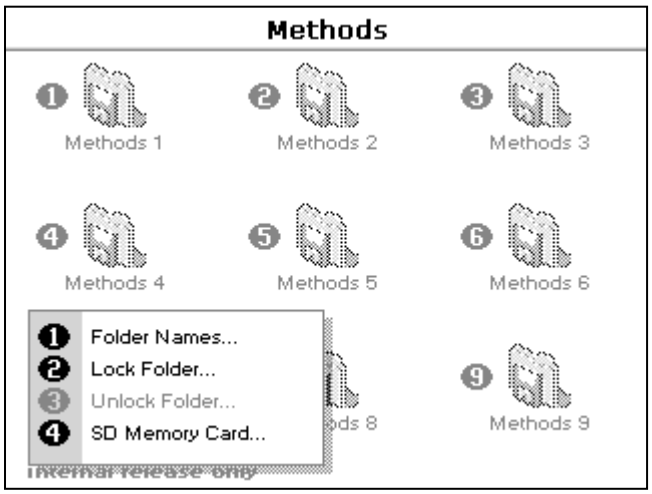
Results are identified by the format of the filename; application type followed by an incrementing file ID. For example

WAVE-001.PVC for a Wavelength scan
KINET001.PVC for a Kinetics file

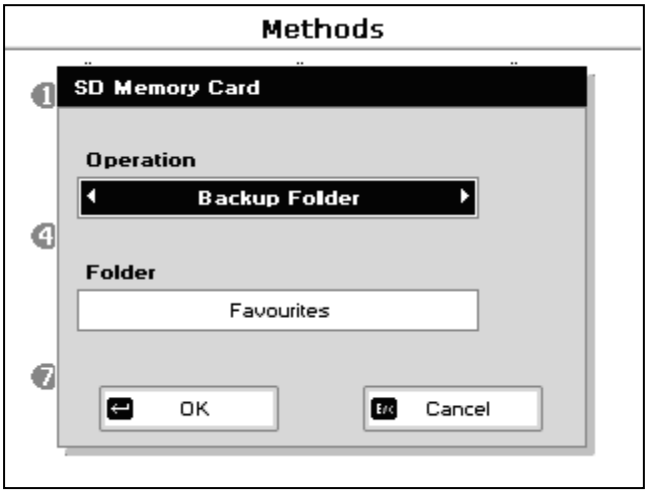
Backup of method folders

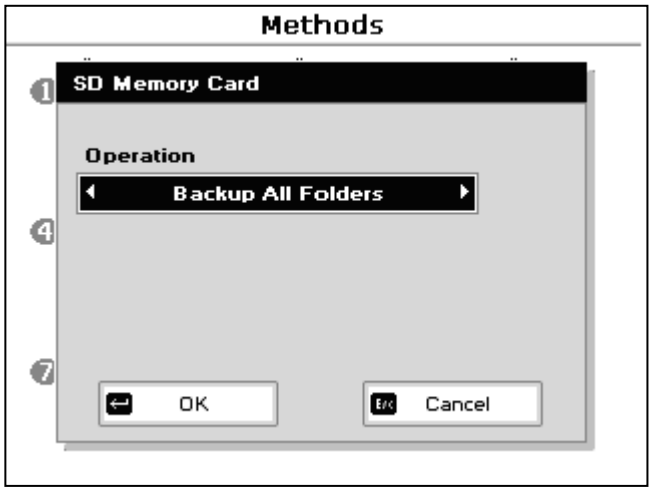
This function allows the user to make a copy of installed methods on an instrument; these methods can be restored back into the same instrument at a later date. There is the option to Backup/Load either a single folder of methods or to Backup/Load all folders on the instrument

To backup the METHOD Folder, go to methods and press OPTIONS followed by SD memory Card.



Then select Backup Folder together with the folder you wish to backup or select Backup All Folders

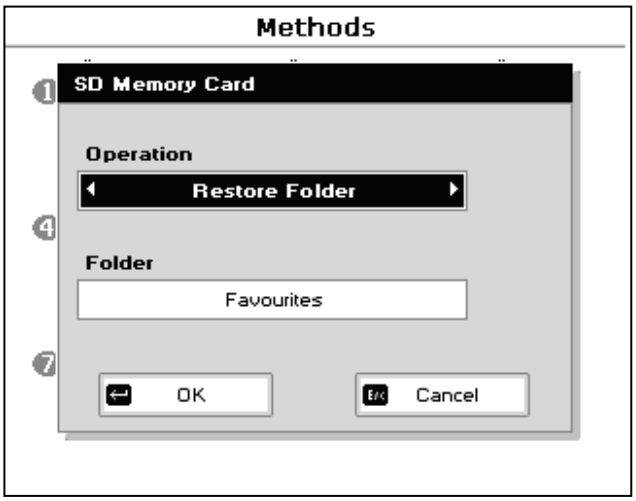


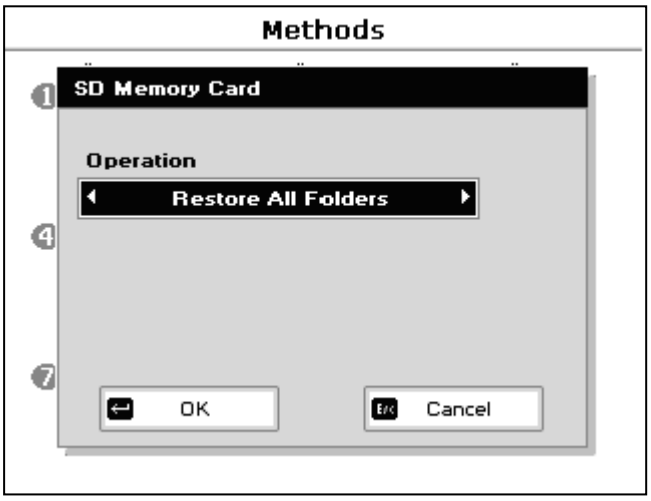


Alternatively, inserting an SD memory card whilst the method screen is displayed will also bring up the same screen.

Folder or folders will then be written to the SD memory Card, the card must not be removed when the LED is lit otherwise the Methods on the card will be corrupted. Methods are stored on the SD memory card in the ***\Serial no*\BACKUP** Directory. All folder names are also stored on the card. Different instruments can be backed up to the same card as methods will be stored under different sub directories due to the different instrument serial numbers.

Restoring method folders
This allows a previous backup of a method folder or all method folders to be restored to the original instrument.





NOTE: any methods currently on the instrument will be overwritten by the restore process and folder names will be changed to match those being restored.

Transfer of data to PC and file management

Methods, data and method folders stored on the SD card can be archived to PC; the data is accessed on the PC using the Print Via Computer (PVC) software supplied with the instrument and it can then be printed or saved in a variety of formats (graphics, text or Excel).

Refer to the user manual included on the PVC CD for installation and operating instructions of PVC.

To transfer data to a PC, the SD memory card should be inserted into the relevant SD memory card slot, either on the PC or on an SD card reader connected to the PC.

PVC Viewer is started and data opened using the Load File menu, navigating to the ***Serial no*\PVC** directory on the SD memory card and selecting the relevant file. Alternatively, you can double click on the file.

Individual method files can be manipulated on the PC using Windows Explorer. Files can be deleted, renamed (maximum 24 characters) or moved between folders (maximum 9 methods per folder).

NOTE: Folder names and method names edited on the PC are not restored to the instrument. Folders on the instrument should be renamed using options\Folder Names.

Restoring method folders to multiple instruments (cloning)

Using this function allows all instruments to be set with the same folder and method structure in a multi instrument lab environment.

- Back up all methods as detailed in section 3.4
- Take the SD Card from the instrument and load into a PC with SD memory card Reader
- Use Windows Explorer to locate the ***Serial no*\BACKUP** file for the instrument you have just backed up
- Rename ***Serial no*\BACKUP** to **\instrument type\BACKUP** where instrument type depends upon the instrument you are using
- This card can then be restored into any instrument of the same type as per section 3.5.

PRINT VIA COMPUTER

- PVC (Print Via Computer) is a small application running under Windows 2000™, Windows XP™, Vista or Windows 7 to enable a Pico200 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
SD Card Accessory	80-3005-00
Spare printer paper	80-3004-07
Excel Electronic Pipette 0.5-10ul with adapter for use with Picodrop	P3600L-10
Manual Gilson Pipetman Pipette 0.5-10ul with adapter for use with Picodrop	F144802
Interchangeable 10mm Cuvette block with lenses/fibres	P200Cuvette
PicoCal UV/Vis 260nm Wavelength Standard fluid, 750ul tube	PCAL01-GLP

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.

'Quick Clean' procedure for sample holder

In the event that sample leaks from a pipette tip or dust reduces the light transfer through the pipette holder simply unscrew the silver cable fibres from each side of the silver pipette holder (no tools required – silver screws should be only hand tight). Unscrew the circular base from the tube section. Unscrew the single screw on the tube to release the main tube from the bottom tip holder. Either soak the holder in hot water with detergent for 30mins and air or drip dry or alternatively simply wash with an ethanol or similar solvent. Reassemble and re-test instrument.

If this quick-clean procedure does not improve the results please follow the 'Service Clean' procedure described in the following section.

Important: Please note that as the lenses get dirty with use then the stability will decrease and this will accentuate any background noise and small variations between tips.

Service Clean Procedure

Detach the Silver fibres by unscrewing from each side of the silver pipette holder.

Use the '1.5mm Allen' key (as supplied) to loosen the 2 fibres by turning the two sunken screws in the front of the bottom of part of the pipette holder and then unscrew the two fibres away from the holder – and attach the LENS EXTRACTION TOOL (pictured below – as supplied) to each lens in place of the fibre and pull firmly on the lens extraction tool to pull out lenses.

Important: if the fibres do not come away from the holder easily please seek assistance from Picodrop service team (info@picodrop.com). DO NOT PULL ON THE FIBRE OPTIC CABLES

Check whether either lens, at the end of each cable, is wet. This happens when excess sample is picked up on the outside of the tips.

- Wash each lens with pure water and dry with tissue.
- Use a cotton tip soaked in acetone to clean and dry the lenses.
- Remove the round base from the holder by unscrewing.
- Thoroughly rinse the metal base unit in pure water and then allow to air dry.
- Re-assemble unit and repeat tests as detailed above.

Lens Extraction tool

SPECIFICATION AND WARRANTY

Wavelength range	220 - 950 nm
Sample Containment	Pipette Tip or 10mm Cuvette
Pathlength Recalibration	Not required as path length fixed by tip – No moving parts
Speed of sample measurement	4 seconds
Nucleic Acid Detection Range(cuvette)	0.4 - 120 ng/ul
Nucleic Acid Detection Range (tip)	3 – 1200 ng/ul
Protein Detection Range (cuvette)	0.02 – 2.5 mg/ml
Protein Detection Range (tip)	0.1 – 25 mg/ml
Monochromator	Flat grating
Wavelength calibration	Automatic upon switch on
Spectral bandwidth	5 nm
Wavelength accuracy	±2 nm
Wavelength reproducibility	±1 nm
Light sources	Pulsed xenon lamp
Detector	1024 element CCD array
Photometric 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 $NaNO_2$
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.

Fibre Optic Cable Handing and Limits of Warranty

Scope: The intent of this note is to define the recommended handling practices for fibre optic cables supplied by Picodrop Ltd. and to comment on warranty issues.

Section 1: Handling Guidelines

A) Strength and Proof Testing:

Do not proof test!

Strength or 'proof' testing is done by the fibre manufacturers. It is a carefully controlled process because it has been found that the more one stresses fibre near its strength limit, reduces it overall life time expectation to continue to resist failures. Therefore, it is specifically recommended that no fibre be proof tested or strength tested except for sample testing in order to gather nominal strength statistics. Any overall proof or strength testing is one at the risk of damaging the fibre. Specifically, proof testing a fibre at or below its bend radius minimum or in any way uncontrolled or unapproved by Picodrop Ltd. will void the warranty.

B) Bend Radius:

Do not bend the fibre past the minimum bend radius!

The rule of thumb regarding bend radius is that the **Minimum** bending radius for a particular fibre is 300x the cladding diameter (assuming the cladding is also glass).

C) Twisting:

Do not coil or twist the cable when spooling, un-spooling, coiling or uncoiling. Cables must be handled in a 'hand over hand' fashion at all times.

Fibre cables should not be handled like rope and coiled or uncoiled by twisting or untwisting one loop at a time. Fibre should always be handled in a 'hand over hand' fashion making coils or unrolling coils by moving the fibre in a circle, one hand over the other. To do otherwise is to induce a twisting stress in the cable and hence in the fibre within the cable. Very high stresses can be achieved if a cable is uncoiled incorrectly and damage or weakening can be the result and this type of handling will void the warranty.

D) Pulling:

Do not pull fibre cables by their end fittings! And, be careful of pulling in general, unless your specific cable has been rated to withstand this stress.

Fibres are usually very strong in direct tension, relative to the cross section, but when fibres are small it is very easy to break them.

E) Other Handling Comments:

Optical fibre is not wire or rope and can't be handled as such.

Any fibre optic cable, constructed with quality techniques and materials, should survive as long as care is taken when handling it. The most precarious moments in a fibre cables life occur during inspection, testing and installation. It is very easy to exceed the bend radius guideline, especially when working with large core cables and performing these tasks.