Ultrospec 7500 Spectrophotometer

USER MANUAL

Intentionally blank

TABLE OF CONTENTS

Intentionally blank

ESSENTIAL SAFETY NOTES

Hazards and Warnings

This section describes potential hazards which may exist in the operation of these units. Several warning labels and symbols are affixed to your instrument. These symbols are used to inform you of potential dangers which may exist or where caution is required. Before installing your new unit, please take time to familiarise yourself with these warnings and symbols.

N.B. THE PROTECTION GIVEN BY THE EQUIPMENT MAY BE IMPAIRED IF USED IN A MANNER NOT SPECIFIED BY THE MANUFACTURER.

This instrument is subject to the following identified hazards:

This unit uses a Xenon lamp. The lamp energy is mainly confined within the unit but traverses the cell holder when a measurement is being taken. Although the energy present is low and intermittent you are advised not to stare into the beam or attempt to deflect the beam as prolonged exposure could result in permanent eye damage.

High voltages exist within the power supply unit and the Xenon lamp housing. Repair and maintenance should only be carried out by individuals trained to work on these instruments.

There are no biohazardous materials within the unit, however, this unit may be exposed to biohazardous samples during normal laboratory use. To protect users against these hazards we recommend the following decontamination procedures:

- Wipe the exterior casework with disinfectant cleaning wipes.
- Remove cuvettes and cuvette holders.
    - Wash with disinfectant appropriate for the biohazard in question.
    - Rinse with distilled water.
    - Allow to dry thoroughly before reuse.

To further reduce the possibility of biohazards:

- Include an appropriate decontamination certificate for equipment returned for repair.
- Ensure that the operator of the equipment is provided with a safe working environment.
- Use, store and dispose of any chemicals in accordance with manufacturer's guidelines and local safety regulations.
- Provide suitable ventilation when working with volatile solvents or toxic substances.
- Dispose of solvents and chemicals that may be classed as hazardous waste in accordance with local regulatory practice.
- Determine if personal protective equipment (PPE) is required for handling laboratory samples.

All models can be connected to and operated from a PC. To preserve the integrity of the measuring equipment it is essential that the attached PC itself conforms to basic safety and EMC standards and is set up in accordance with the manufacturers' instructions. If in doubt, consult the information that came with your PC.

The following safety precautions should be observed when operating a PC:

- To reduce the chance of eye strain, set up the PC display with the correct viewing position, free from glare and with appropriate brightness and contrast settings.
- To reduce the chance of cross contamination from biological samples, use appropriate personnel protection measures and disinfectant wipes on keyboard and mouse.

In the event of contamination, malfunction or hazard occurring, the operator should disconnect the unit, by removing the power cord, and isolate for decontamination and/or repair.

INTRODUCTION

The Biochrom Ultrospec Spectrophotometers

Spectrophotometers are ubiquitous among modern laboratories. Ultraviolet (UV) and Visible (VIS) spectrophotometry has become the method of choice in most laboratories concerned with the identification and quantification of organic and inorganic compounds across a wide range of products and processes. Applied across research, quality, and manufacturing, with continuing focus on life science and pharmaceutical environments, they are equally as relevant in agriculture, animal husbandry and fishery, geological exploration, food safety, environmental monitoring, and many manufacturing industries to name a few.

The Ultrospec spectrophotometers are quick, accurate, and reliable. They require only small demands on the time and skills of the operator. This operating manual details the processes in taking basic measurements using the Ultrospec 7500 spectrophotometer.

The Ultrospec 7500 instrument is UV-VIS split-beam spectrophotometers with a 2 nm spectral bandwidth and comes as standard, with a 10 mm pathlength 8 position cell changer, however a range of alternative accessories are available.

INSTALLATION

Unpacking and Contents List

The following items and quantities are supplied as standard with the Ultrospec 7500 (p/n 80-2140-60). Please check this contents list against the actual content in the box. If any discrepancies are found, please contact Biochrom or your local dealer.

Item Description	Qty
Ultrospec 7500	1
USB memory stick (includes this User Manual and PVC software latest version)	1
Calibration Certificate	1
Dust Cover	1
European Power Line Cord (220V)	1
US Power Line Cord (110V)	1
UK Power Line Cord (220V)	1
Power Supply Unit (19V)	1
USB Cable	1

- The unit weighs ~13 kg. No special handling is required.
- Please keep the original packaging for transport for service or repair as it has been specifically designed to protect the unit from damage during transit.
- Inspect the instrument and its power supply for any signs of damage caused during transit. If any damage is discovered, do not use the instrument, and report the problem to Biochrom or your local dealer.

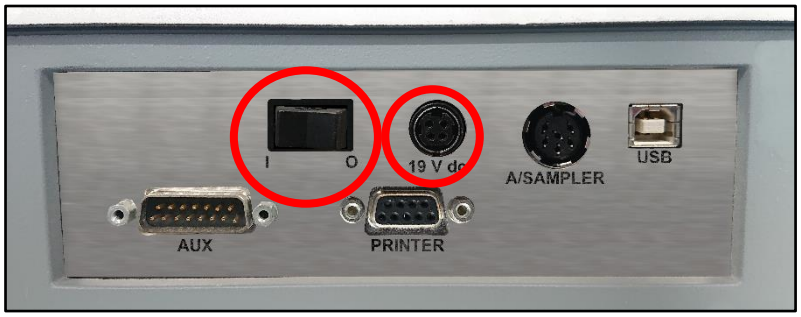
Positioning

- Ensure your proposed installation site conforms to the environmental conditions for safe operation:
    - Indoor use
    - 5 to 40°C
    - Maximum relative humidity 90% up to 31°C decreasing linearly to 50% at 40°C.
- Extremes of temperature may require recalibration of the unit for optimal performance.
- The instrument must be placed on a stable, level bench or table capable of supporting its weight allowing sufficient space around the instrument for air to circulate freely.

- The instrument should be positioned so that the power supply cable may be readily removed in the event of a hazard or malfunction.
- Locate the instrument in an atmosphere free from dust and corrosive fumes. Use the dust cover to further protect the instrument when not in use.

Installing

- If the instrument has been stored in a cold environment, then it should be allowed to come to room temperature before turning it on to avoid compromising the internal calibration procedure.
- The equipment is operated using a 19 VDC power supply adapter unit. Always use the power supply adapter and mains cords supplied with the instrument.
- Mains power requirements are as follows:
  - 100 to 240 VAC~
  - 50 or 60 Hz
- The UK style mains cord plug has a user replaceable 3A fuse. Replace only with the same rating and type 3A BS1362.
- The unit maximum power rating is 90 VA.
- Connect the instrument to the mains power using the main power cord and the 19 VDC power supply adapter unit, then turn the instrument's main switch to the on (I) position, this will Power on the instrument followed by a series of self-diagnostic checks.



Main switch and 19 VDC power supply socket

WARRANTY AND REPAIR

Warranty Policy

Biochrom warrants these instruments for a period of 24 months (2 years), and an additional 12 months (3 years in total) for the xenon lamp, from the date of purchase. Where appropriate, Biochrom will repair or replace the unit for defects of workmanship or materials. This warranty does not extend to damage resulting from misuse, neglect, or abuse, normal wear and tear, or accidental damage. This warranty extends only to the original purchaser.

Products failing within the first 30 days of end user operation are considered dead on arrival (DOA) and where appropriate a replacement will be given if a repair is not possible. In the instance of a DOA Biochrom will incur the return shipping charges.

IN NO EVENT SHALL BIOCHROM BE LIABLE FOR INCIDENTAL OR CONSEQUENTIAL DAMAGES. Some states do not allow the exclusion or limitation of incidental or consequential damages so the above limitation to exclusion may not apply to you. THERE ARE NO IMPLIED WARRANTIES OF MERCHANTABILITY, FITNESS FOR A PARTICULAR USE, OR OF ANY OTHER NATURE. Some states do not allow this limitation on an implied warranty, so the above limitation may not apply to you.

Returns

If any defect arises within or outside the warranty period, please contact:

US Office Technical Support	
Email	support@hbiosci.com
Online Returns form	https://support.biochrom.co.uk/hc/en-us/requests/new?ticket_form_id=1500000731442
Telephone (Toll Free)	+1 800 272 2775
Telephone (Outside the US)	+1 508 893 8999
Address	84 October Hill Road Holliston MA 01746 USA
UK Office Technical Support	
Email	support@hbiosci.com
Online Returns form	https://support.biochrom.co.uk/hc/en-us/requests/new?ticket_form_id=1500000731442
Telephone	+44 (0) 1223 423 723
Address	Unit 7, Enterprise Zone 3970 Cambridge Research Park Beach Drive, Waterbeach Cambridge, United Kingdom CB25 9PE

Goods will not be accepted for return unless an RMA (Return Materials Authorization) number has been issued. The unit must be returned only once the online RMA form has been completed and submitted, and an RMA number has been issued. The customer is responsible for shipping charges unless the failure is within 30 days of receiving the goods. Please allow a reasonable amount of time for completion of repairs or replacement.

INSTRUMENT OVERVIEW

Scope

This user manual covers the following range of Biochrom UV/Visible spectrophotometers:

Part Number	Description
80-2140-60	Ultrospec 7500

Spectrophotometer Principle and Intended Use

UV/Visible spectrophotometers measure the transmission of light through a sample. Samples absorb light based on their unique molecular composition. The amount of absorbance is directly proportional to the sample concentration and the pathlength, which is the distance that the light travels through the sample.

UV/Visible spectrophotometers are used in a number of different laboratory environments including life science, clinical, healthcare and industrial laboratories. In a life science laboratory, UV/Visible spectrophotometers are commonly used to measure the concentration of nucleic acids and proteins.

Hardware

Your spectrophotometer is a simple-to-use UV/Visible instrument with two silicon photodiodes. A 1200 lines/mm aberration corrected concave grating mounted on a calibrated motor, which is the basis of the quick and accurate scanning operating system.

Technical Specifications

Wavelength range	190 to 1100 nm
Monochromator	1200 lines/mm Aberration corrected concave grating
Wavelength calibration	Automatic upon switch on
Beam Height	15 mm
Spectral bandwidth	<2 nm
Wavelength accuracy	±1 nm
Wavelength reproducibility	±0.5 nm
Light sources	Xenon flash lamp
Detector	two silicon photodiodes
Photometric range	-3.000 to 3.000 A, 0.1 to 100 %T
Photometric accuracy	±0.5 % or ±0.003 A whichever is greater at 546 nm
Photometric reproducibility	±0.5 % to 3.000 A at 546 nm
Stray Light	<0.05 %T at 220 nm using NaI or at 340 nm using $NaNO_2$ <0.10 %T at 380 using $NaNO_2$
Stability	±0.001 A/h at 340 nm for 0 A
Noise	±0.002 peak to peak ± 0.0005 RMS at 340 nm for 0 A
Digital output	USB Flash Drive, PC via PVC software
Data Export	USB Flash Drive: .tsv, native PVC format PC via PVC: .csv, .emf, .xlsx, .xls, .rtf, .tsv, native PVC format
Method Storage	156 with PIN number protection
Graphical Display	Yes, zoom and track function
Sample ID	Yes
Languages	English, German, French, Spanish, Italian, Japanese, Chinese
Dimensions	510 × 350 × 160 mm
Weight	13.00 kg
Power input	19 VDC at max 90 VA from a supplied 100 to 240 V~, 50/60 Hz Mains Power Adapter

Touchscreen Display

The instrument has an 800 × 480-pixel resolution backlit LCD colour display with touch panel for navigating the instrument's built-in firmware. The instrument is very energy efficient.

Instrument Connections

USB connector for PC connection

USB connector for USB memory stick

PVC PC Software

The instrument is supplied with the PVC software program (supplied with its own devoted operating manual) on the accompanying USB flash drive. The instrument can be connected to a PC onto which the PVC software has been installed, via a USB A to USB B cable. This enables the operator to “print through” the PC directly to the printer that is connected to it. The data may also be stored as a comma-separated value (.csv), enhanced meta file (.emf), Excel spreadsheet (.xlsx, .xls), rich text format (.rtf), tab-separated value (.tsv) or in a native PVC format file.

Biochrom Resolution PC Software

When connected to a PC the spectrophotometer can be controlled using the Biochrom Resolution PC software packages (sold separately). Operation using Biochrom Resolution PC software is described in the Resolution user manual or Resolution help file.

Instrument Data Output

A printer accessory is available for the instrument. This is an optional accessory for end-user installation.

Measurement data can also be exported to a USB flash drive via the USB A socket on the side of the instrument, as either a tab-separated value (.tsv) or native PVC format file.

Performing a Measurement

The optical height (z value) of the instrument is 15 mm. The light path is directed from LEFT to RIGHT through the cell chamber.

The 8-position cell changer supplied as standard with the instrument accepts 10 mm pathlength quartz, glass, or plastic cuvettes. When using a cuvette with a pathlength less than 10 mm, ensure the cell is inserted to the far right of the cell holder and secured using an appropriate packing piece.

Please consult the User Interface section of this user manual for more detail on taking a measurement using the spectrophotometer. In summary, how to perform a measurement is outline below.

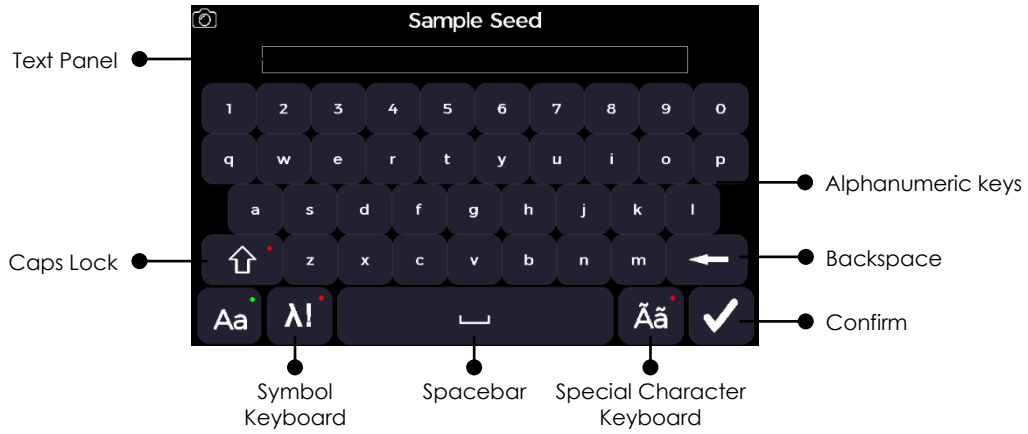
1. Open the desired application on the spectrophotometer.
2. Insert a cuvette containing the reference sample into cell 1 of the 8-position cell changer and insert a cuvette containing the sample into cell 2. If measuring more than one sample, insert cuvettes containing samples into cells 2 – 8.
3. Set the appropriate parameters, moving through the parameter screens.
4. When you get to the measurements screen, take measurements by pressing the batch measurement icon  ; you will be prompted to load the 8-cell changer and confirm  or cancel  . The acquired reference baseline is applied to any subsequent sample measurements until a new reference baseline is taken, or the application is closed.

Colour Touchscreen

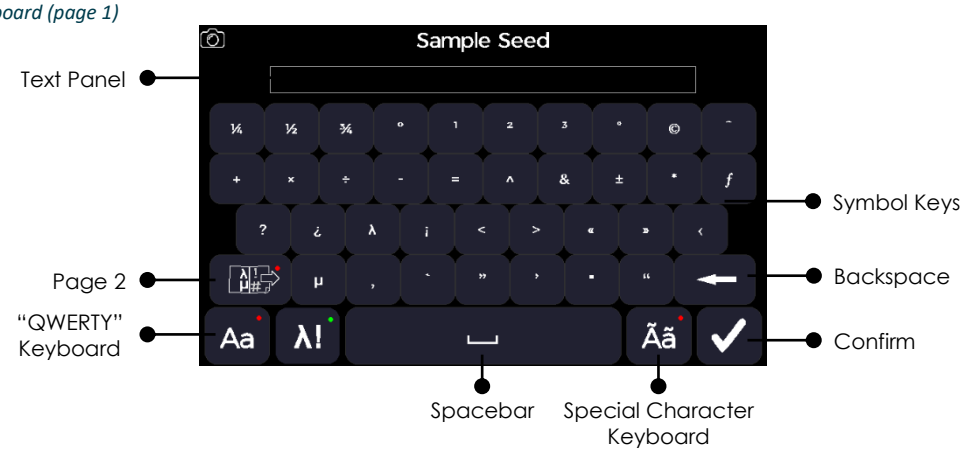
The instrument is controlled using the colour display and touchscreen. The onscreen keyboards and number pad, and frequently used icons are detailed in this section of this operating manual.

Onscreen Keyboards and Number Pad

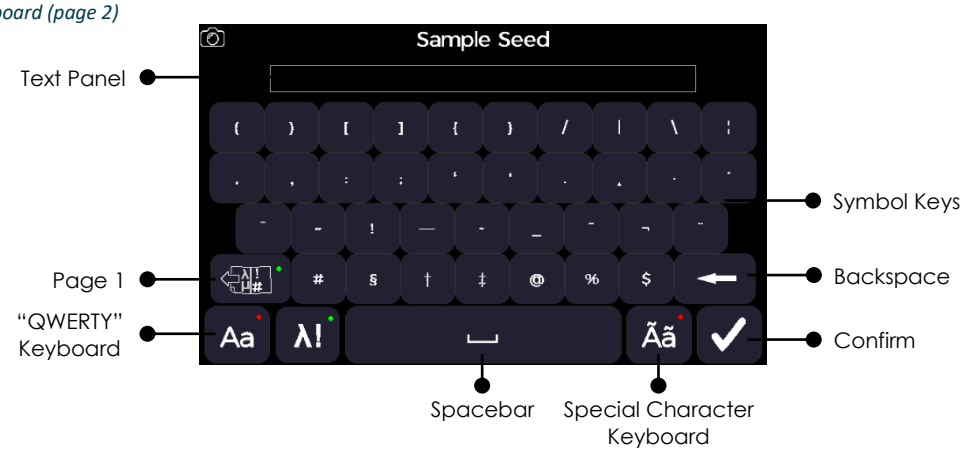
"QWERTY" Alphanumeric Keyboard



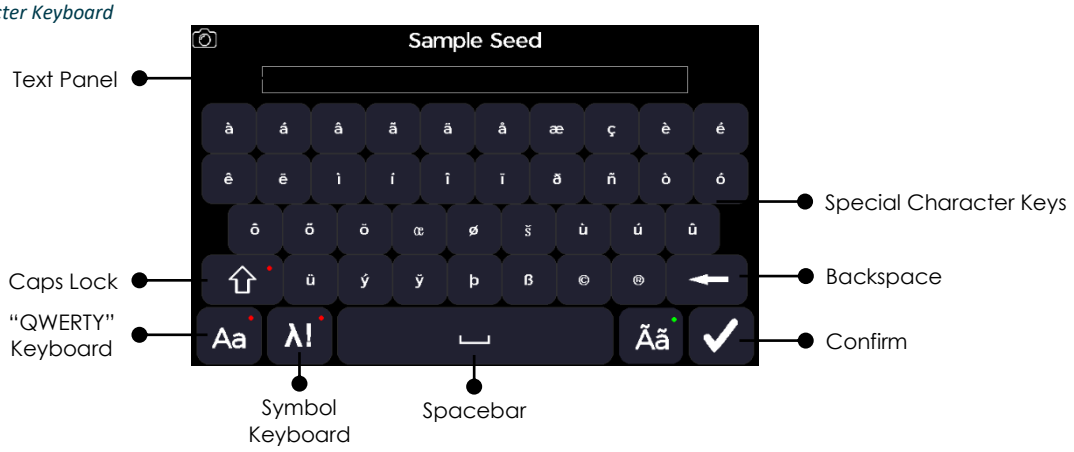
Symbols Keyboard (page 1)



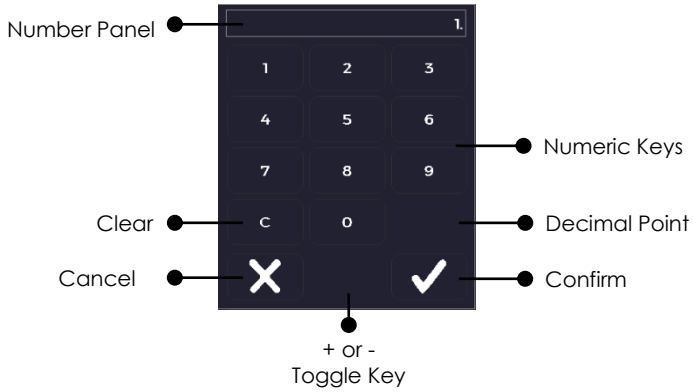
Symbols Keyboard (page 2)



Special Character Keyboard



Number Pad



Frequently Used Icons

The frequently used icons detailed in this section are to support the quick-start operation of the instrument. Method specific icons are detailed in the relevant method section.

Navigating Icons

	**Right/forward arrow**	Progress to the next screen
	**Left/backward arrow**	Return to the previous screen
	**Confirm**	Confirm selection
	**Cancel**	Cancel selection

Common Icons on the Sample Measurement Screen

	**Reference measurement**	Take a reference measurement
	**Sample measurement**	Take a sample measurement
	**Batch measurement**	Take a batch measurement (with a cell changer)
	**Options**	Open the options menu
	**Parameters**	Return the method parameters

Common Icons on the Options Menu

	**Exit**	Exit the application and return to the application menu
	**Save data**	Save the sample data
	**Save method**	Save the method with the current parameter's settings
	**Print**	Print the sample data from the specified printer
	**Auto-print**	Toggle auto print on (green) or off (red)
	**Go to std Curve**	Takes you to standard curve screen
	**Load sample**	Open saved sample data

Home Screen Toolbar Icons

	**Settings**	Accesses the instrument settings
	**GLP Status**	Open the GLP Application
	**Switch user**	Open the user login window (only seen if 'show log in' is activated in user settings)
	**Information**	Accesses the instrument information

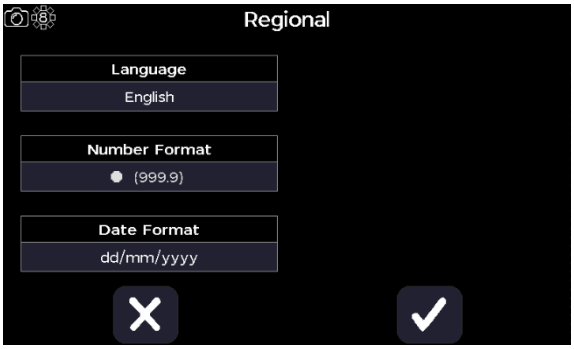
Instrument Firmware

The instrument firmware uses an intuitive menu arrangement that is navigated using the colour display icons and touchscreen.

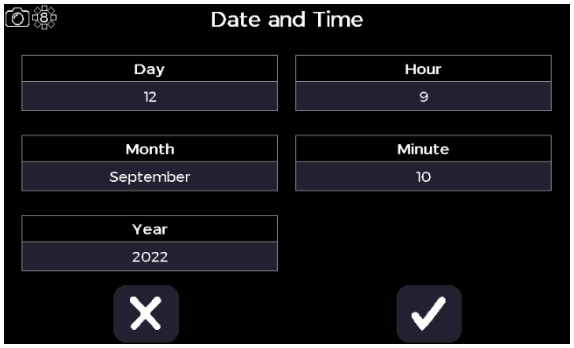
First-time Start-Up

Upon first powering up of the instrument, the following screen sequence is displayed.

1: Self-calibration routine screen



2: Regional settings page. Select the appropriate settings according to your location



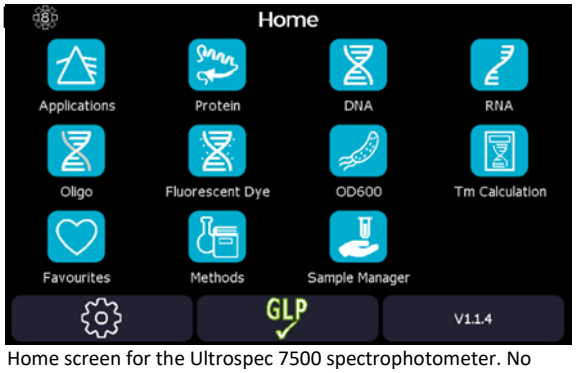
3: Date and time setup page. Set the date and time according to your location

4: Home page

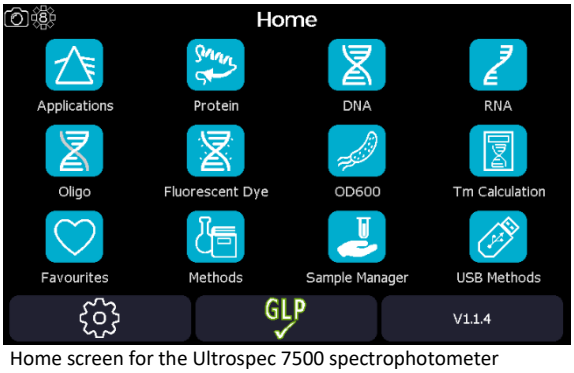
After the first-time start-up, any future instrument starts-up will only display the self-calibration routine screen followed by the Home page.

Home Screen

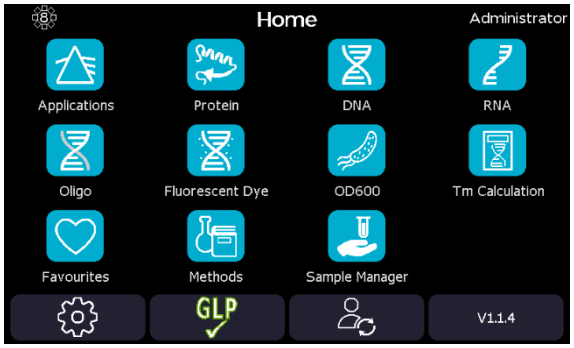
All applications can be accessed from the instrument Home screen using the icon-based menu. The Settings icon, the Switch User icon (if enabled in the User Access Control Page) and the information icons are in the toolbar across the bottom. The pictures below represent the different Home screen configurations possible.



Home screen for the Ultrospec 7500 spectrophotometer. No user login has been set.



Home screen for the Ultrospec 7500 spectrophotometer displaying the USB memory stick application and screenshot camera icon, made available when a USB flash drive is inserted



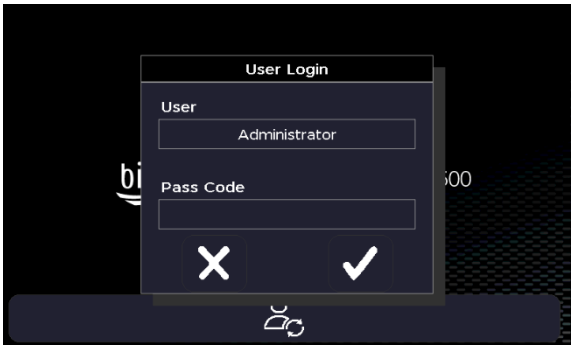
Home screen for the Ultrospec 7500 spectrophotometer when User Login is available

Home screen for the Ultrospec 7500 spectrophotometer displaying the USB memory stick application and screenshot camera icon, made available when a USB flash drive is inserted and User Login available

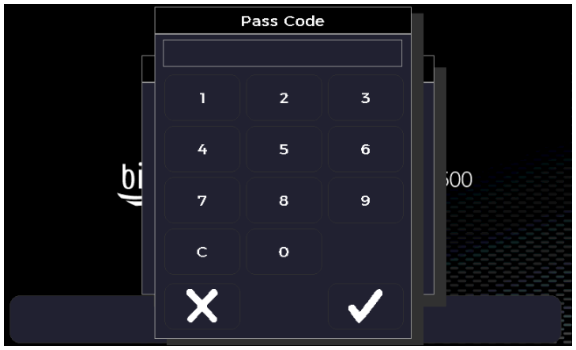
Login Screen

The instrument Ultrospec 7500 login screen is the first screen displayed after self-initialisation of the instrument if the 'Show Login' setting has been enabled on the 'Edit User Access – Parameters' screen for the default Administrator user (see **User Access** section below for more details). Once enabled, the unit can be unlocked using the switch user icon  . Then with the user set to the default "Administrator", enter "1000" as the passcode and select the confirm  icon. Confirm the user login details using the confirm icon to progress to the instrument home screen.

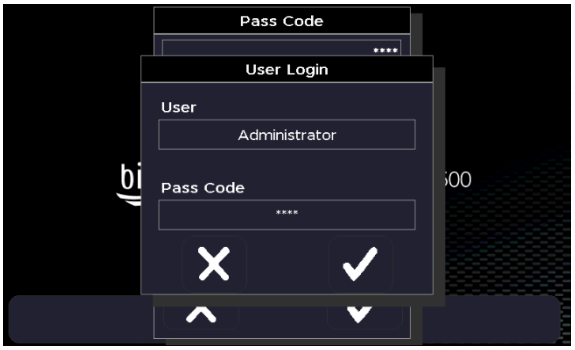
Login screen for the Ultrospec 7500 spectrophotometer



User login window displayed after selecting the switch user icon



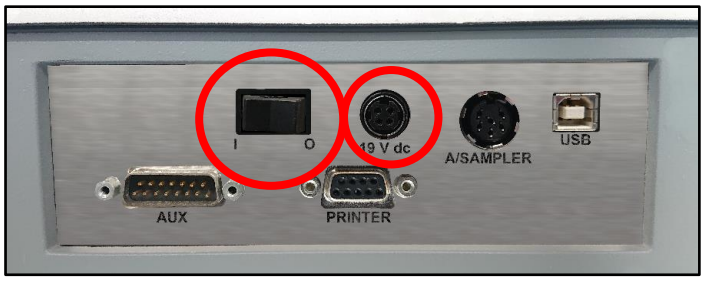
The pass code number pad displayed after selecting the pass code entry box



The user login showing the default user login details

Power Off

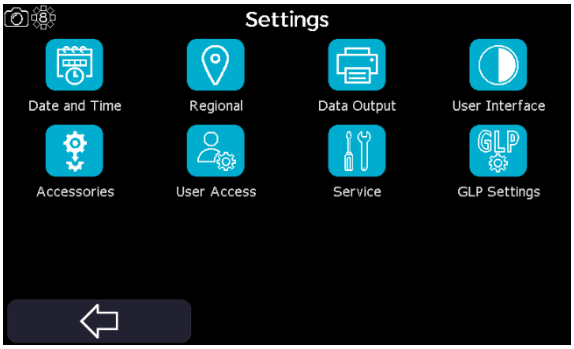
To Power off the instrument switch off the main switch to the off (0) position.



Main switch and 19 VDC power supply socket

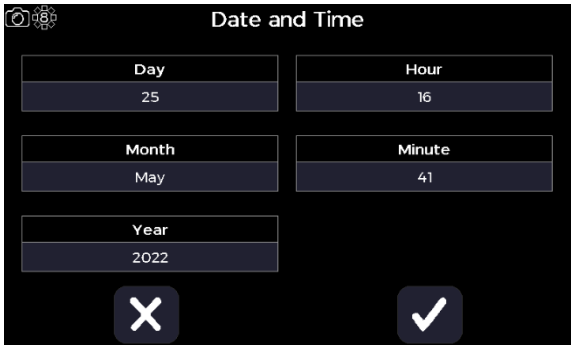
Settings

The Settings screen is accessed from the home screen settings icon  . The settings screen can be used to adjust the instrument settings: date and time, regional, data output, user interface, accessories, user access, service and GLP settings. Note that the service application is used by engineers and a passcode is required.



Date and Time

The Date and Time application is accessed from the Settings screen. It can be used to adjust the date and time stamp applied to measurement data outputs.



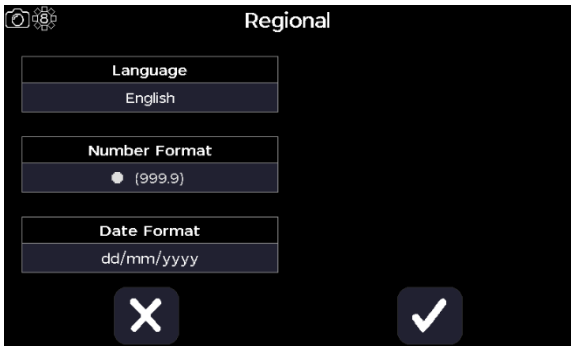
There are several setting options available.

1. Set the day using the number pad.
2. Select the month from the selection menu.
3. Set the year using the number pad.
4. Set the hour (24-hour format) using the number pad.
5. Set the minute using the number pad.

Confirm any changes using the confirm icon.

Regional

The Regional application is accessed from the Settings screen. It can be used to change the language and decimal separator number format.



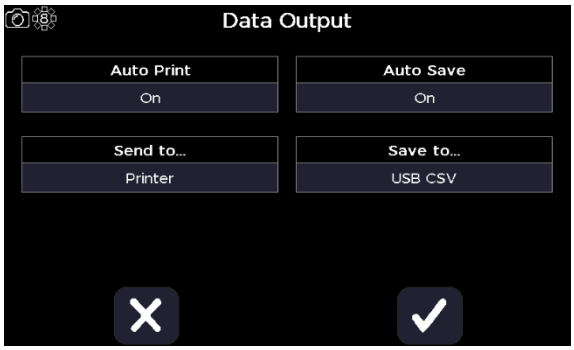
There are several setting options available.

1. Select the language from the selection menu.
2. Toggle between full stop and comma number format decimal separator.
3. Select the date format from the selection menu.

Confirm any changes using the confirm icon.

Data Output

The Data Output application is accessed from the Settings screen. It can be used to define the default printer and data output settings.



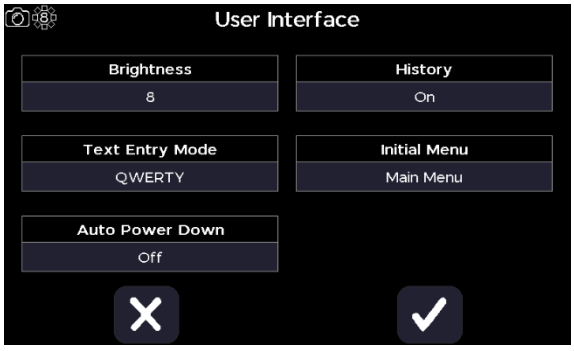
There are several setting options available.

1. Set auto print to “On” or “Off”.
2. If auto print is set to “On”, select the print to hardware from “Internal Printer”, “PC via USB”, or “USB Mass Storage” depending on what hardware is connected to the instrument.
3. Set auto save to “On” or “Off”.
4. If auto save is set to “On”, select the save to hardware from “USB CSV”, “USB”, or “Internal” depending on what hardware is connected to the instrument.

Confirm any changes using the confirm icon.

User Interface

The User Interface application is accessed from the Settings screen. It can be used to define the user interface preferences.



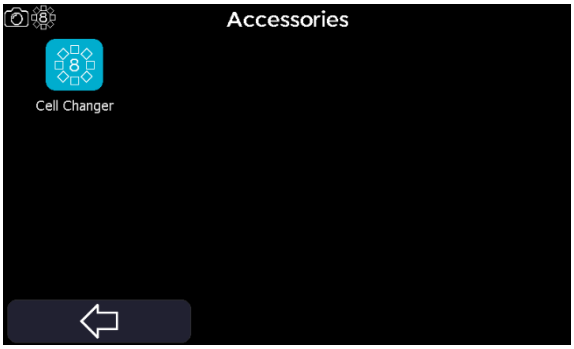
There are several setting options available.

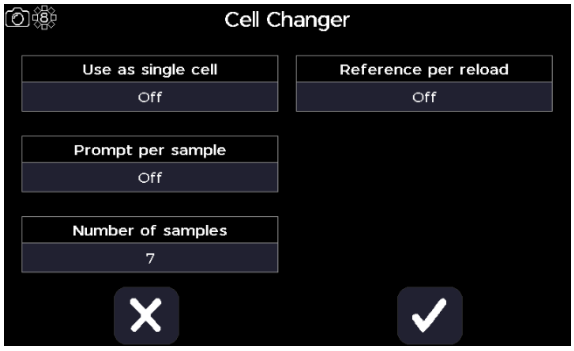
1. Set the brightness from the selection menu on a scale of 0 to 8.
2. Toggle between “QWERTY” or “A to Z” keyboards for text entry.
3. Set the screensaver activation time from the selection menu, from “Off”, “5 minutes”, “10 minutes”, “30 minutes”, or “1 hour”.
4. Toggle the parameter history to “On” or “Off”, to store application parameters for future use or not.
5. Select the initial menu from the selection menu.

Confirm any changes using the confirm icon.

Accessories

The Accessories application is accessed from the Settings screen. It can be used to identify which accessories are fitted to the instrument and to define their default settings. The example below shows the 10 mm pathlength, 8 position cell changer that comes as standard with the Ultrospec 7500.





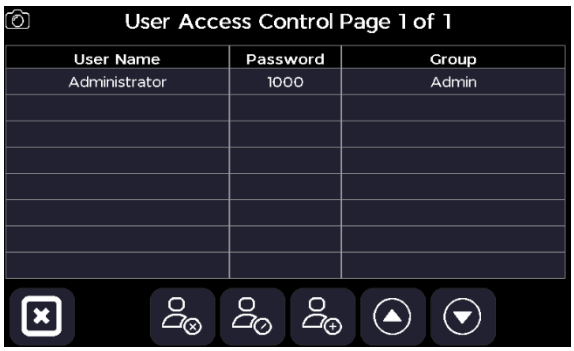
There are several settings options available depending on the fitted accessory (see Accessories section).

1. Toggle whether to use as single cell holder to “On” or “Off”.
2. Toggle whether to prompt per sample to “On” or “Off”, to require a ‘take measurement’ icon press per sample or just once per loaded changer.
3. Set the default number of sample to run to between 1 and 7.
4. Toggle whether to reference per reload to “On” or “Off”, to take a fresh reference measurement when reloading the cell change within the same batch of measurements or not.

Confirm any changes using the confirm icon.

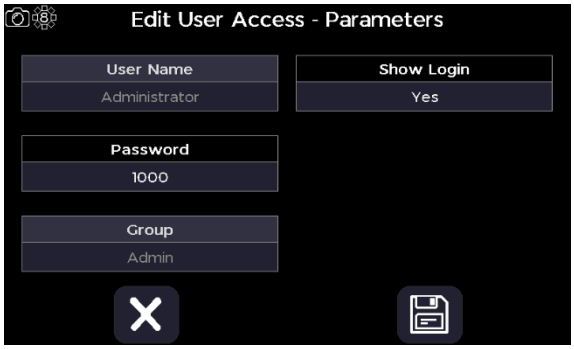
User Access

The User Access application is accessed from the Settings screen. It can be used to manage user access logins, passwords, and permission group.

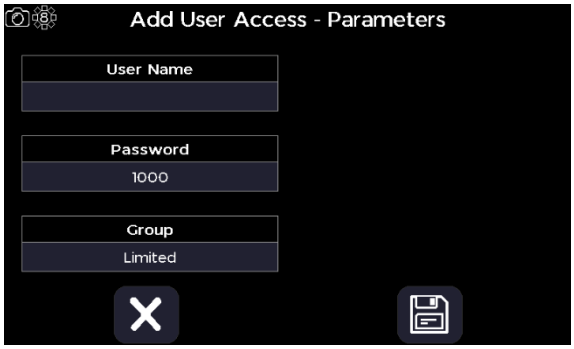


There are several setting options available.

1.  Delete selected user account.
2.  Edit selected user account.
3.  Add new user account.



Editing the "Administrator" user account allows to enable the login function upon start-up of the unit for all the users.
Select "Yes" in the ***Show Login*** box to enable the user login upon start up. The default Administrator password is 1000.

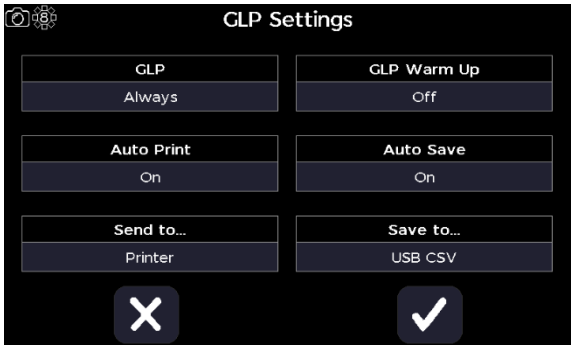


New accounts can be assigned to 3 permission groups.

Administrator – has access to all features and applications.
Supervisor – has access to all applications but not "User Access" settings or "Information" applications.
Limited – Only has access to applications but cannot save methods, does not have access to any "Settings" or "Information" applications.

GLP Settings

The GLP Settings application is accessed from the Settings screen. It can be used to define the GLP routine preferences.



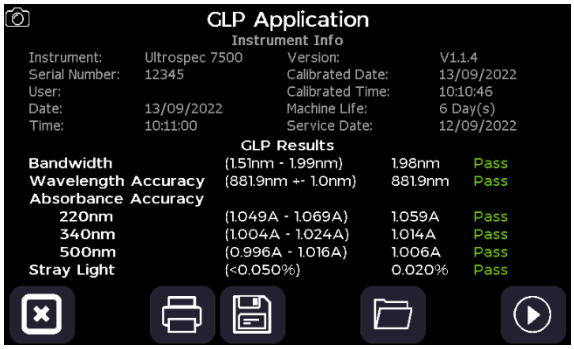
There are several GLP setting options available.

1. Set the GLP frequency from the selection menu, from “Always”, “Daily”, “Weekly”, “Monthly”, or “Quarterly”.
2. Set auto print to “On” or “Off”.
3. If auto print is set to “On”, select the print to hardware from “Printer”, or “USB Mass Storage” depending on what hardware is connected to the instrument.
4. Toggle GLP warm up to “On” or “Off”, to wait for 10 minutes before the GLP process begins.
5. Set auto save to “On” or “Off”.
6. If auto save is set to “On”, select the save to hardware from “USB CSV”, “USB”, or “Internal” depending on what hardware is connected to the instrument.

Confirm any changes using the confirm icon.

GLP application

The GLP application icon is visible on the home screen toolbar. It is used to view the latest GLP report, as well as print, save, view previous GLP reports, and rerun the GLP report.



There are several actions available.

1.  Open the GLP application.
2.  Print the GLP report.
3.  Save the GLP report.
4.  Open a previous GLP report.
5.  Rerun the GLP report.

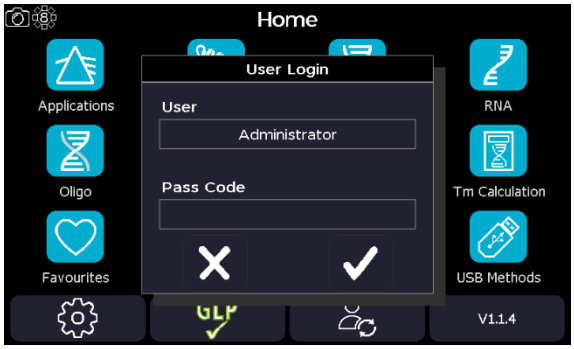
GLP error

In case one of the test parameters is out of its limits, the GLP will fail and will display a red GLP Icon like bellow.

In such an unlikely case please contact Harvard Bioscience Technical Support and share a screenshot from the GLP results screen.

Switch User

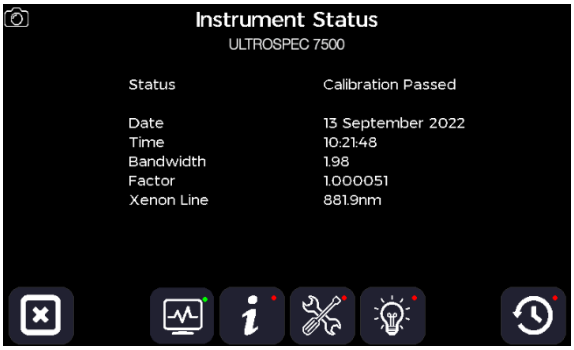
The switch user icon is visible on the home screen toolbar, providing the 'Show Login' setting is enabled on the 'Edit User Access – Parameters' screen for the default Administrator user. It is used to change the active user on the instrument without having to restart the instrument first.



Select the switch user icon, then cycle through the usernames and enter the appropriate pass code. Confirm or cancel the action using the confirm or cancel icons.

Instrument Status

The Instrument Status screen is accessed from the home screen toolbar icon. It can be used to view the Instrument Status and access the Instrument Information, the Instrument Settings, Lamp Settings, and Instrument Reset screens.



Instrument Information

The Instrument Information screen is accessed from the Instrument Status screen using the instrument information icon.

It can be used to view the basic instrument information.

Instrument Settings

The Instrument Settings screen is accessed from the Instrument Status screen. It can be used to create and store a new instrument baseline.

From the Instrument Status screen, select the instrument settings icon.

There are several setting options available.

1. Create a new temporary baseline, using the new baseline icon.

2. Save the temporary baseline as the permanent baseline, using the save baseline icon.

3. Restore the temporary baseline from the stored permanent baseline, using the reset baseline icon.

Lamp Settings

The Lamp Settings screen is accessed from the Instrument Status screen using the lamp setting icon.

It can be used to view the age of the lamp.

Instrument Reset

The Instrument Reset screen is accessed from the Instrument Status screen. It can be used to delete all the user data from the instrument.

From the Instrument Status screen, select the instrument reset icon.

There are several setting options available.

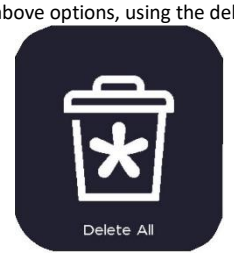
1. Delete all user samples, using the delete samples icon.

2. Delete all user logins, using the delete users icon.

3. Delete all user methods, using the delete methods icon.

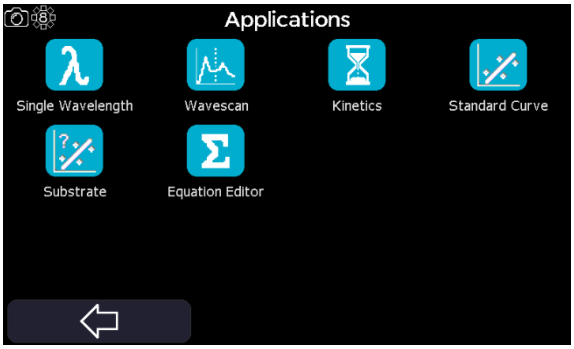
4. Reset all method folder names, using the delete method folder names icon.

5. Perform all of the above options, using the delete all icon.



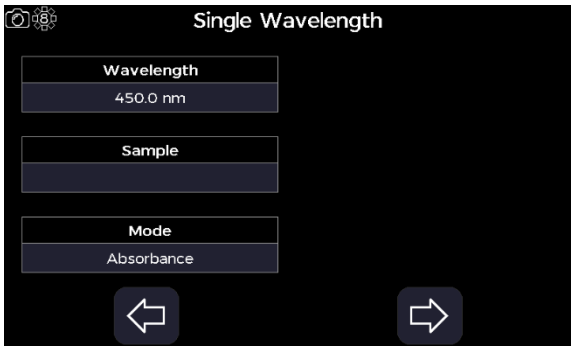
Applications

The Applications screen is accessed from the home screen. It contains basic applications with definable parameters to meet the needs of typical laboratory protocols.

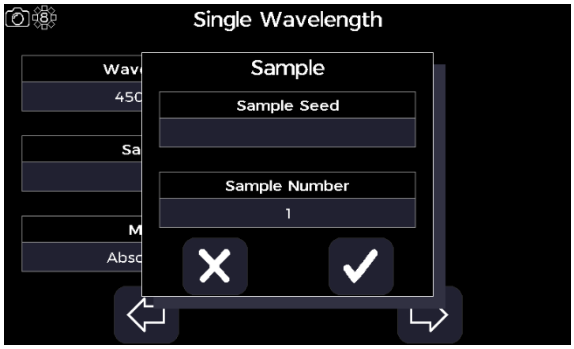


Single Wavelength

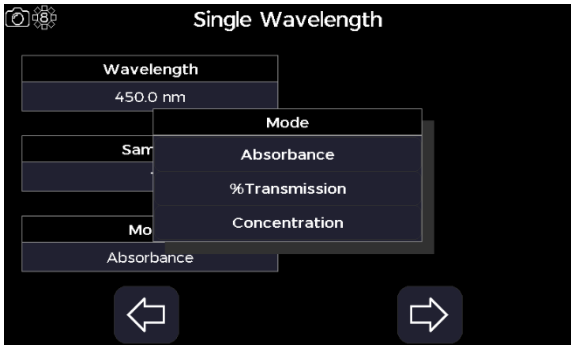
The Single Wavelength application is selected from the Applications screen. It can be used to perform simple absorbance (A) and % transmission (%T) measurements. It can also be used to determine the concentration of the sample by applying a known factor, or a factor determined using a standard of known concentration, to the single wavelength absorbance (A) measurement.



Step 1
Set the wavelength to between 190 and 1100 nm.

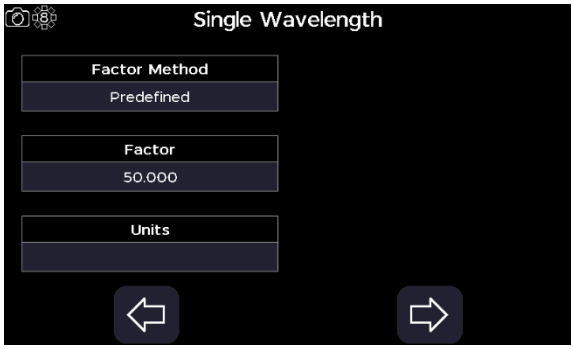


Step 2
Select the sample name to bring up the sample window, enter a sample seed prefix and the incremental sample number starting value. Confirm the settings using the confirm icon.



Step 3
Select the mode from "Absorbance", "%Transmission", or "Concentration".

If "Absorbance" or "%Transmission" mode is selected, skip steps 4 through 7 and go straight to step 8.

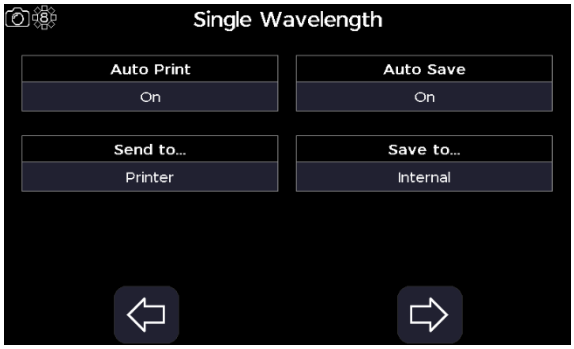


Step 4
If "Concentration" mode is selected. Proceed to the next parameter screen using the right/forward arrow.

Step 5
Select the factor method to be applied to the absorbance value from "Predefined" or "Standard".

Step 6
Set the factor or concentration value to between -9999 and 9999, according to the factor method selection of "predefined" or "Standard" respectively.

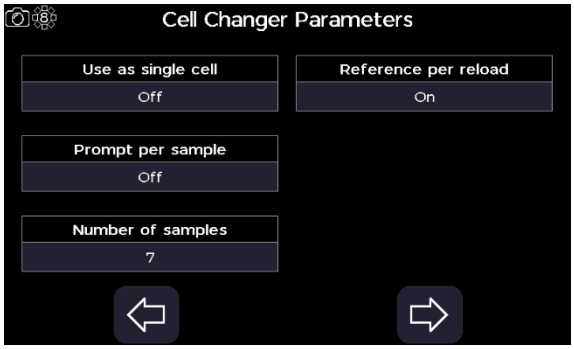
Step 7
Define the units that the concentration value will be reported in.



Step 8
Proceed to the next parameter screen using the right/forward arrow.

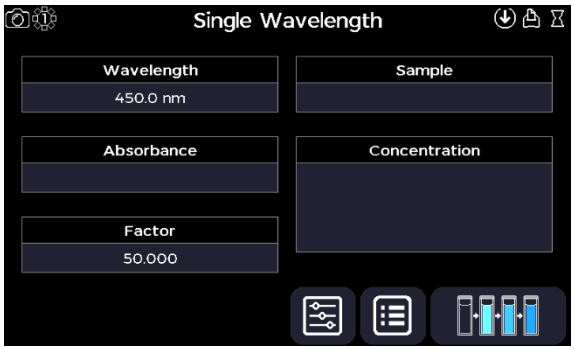
Step 9
Set auto print to "On" or "Off". If auto print is set to "On", select the print to hardware from "Internal Printer", "PC via USB", or "USB Mass Storage" depending on what hardware is connected to the instrument.

Step 10
Set auto save to "On" or "Off". If auto save is set to "On", select the save to hardware from "USB CSV", "USB", or "Internal" depending on what hardware is connected to the instrument.



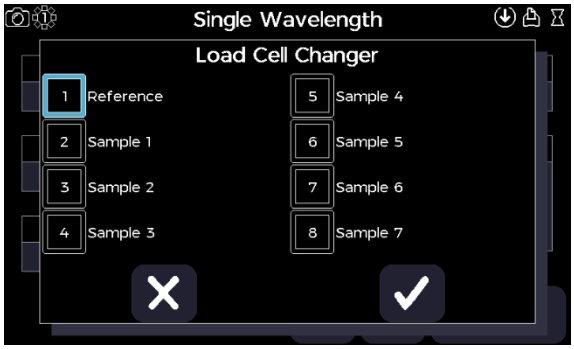
Step 11
Proceed to the next parameter screen using the right/forward arrow.

Step 12
Set whether to use as single cell to "On" or "Off". If set to "Off", set the position prompt per sample to "On" or "Off", set the number of samples to between 2 and 100, and set whether to retake the reference between reload to "On" of "Off".

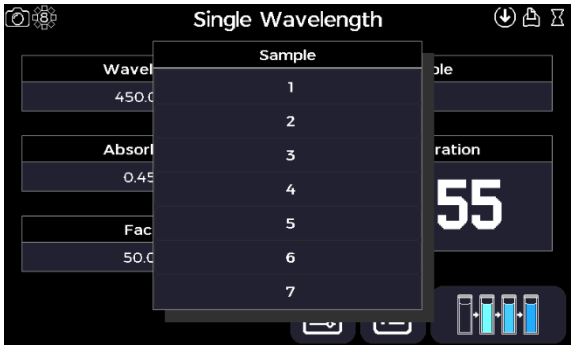


Step 13
Proceed to the measurement screen using the right/forward arrow.

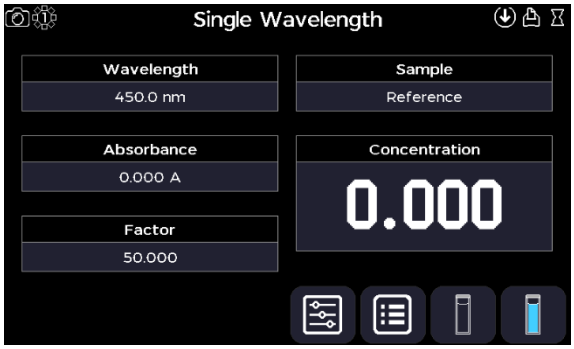
If using a single cell holder, or a cell changer set to use as a single cell holder, skip steps 14 through 15 and go straight to step 16.



Step 14
Press the batch measurement icon, then load the cell changer according to the cell changer prompt. Confirm when ready to take measurements.



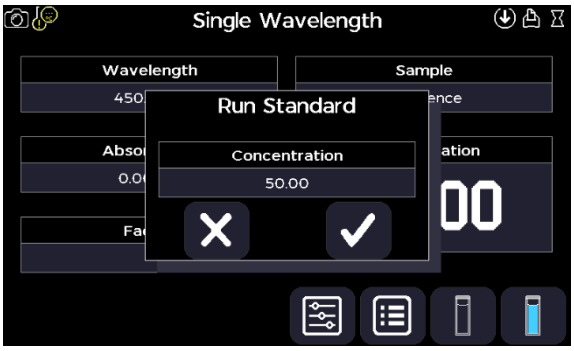
Step 15
The acquired reference sample baseline will be applied to all subsequent sample measurements. The sample measurements can be viewed by pressing the sample name test box and selecting the appropriate sample from the list.



If using a cell changer, skip steps 16 through 19 and go straight to step 20.

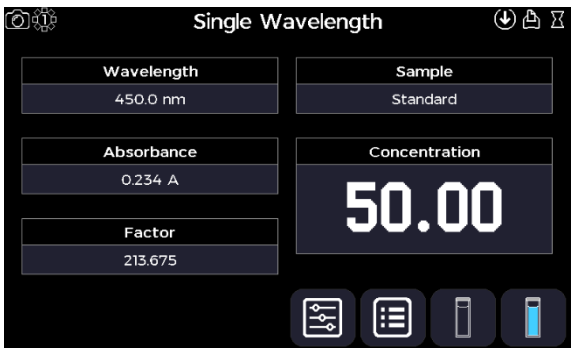
Step 16
Insert the reference sample then take a reference measurement using the reference measurement icon.

The acquired reference sample baseline will be applied to all subsequent sample measurements.

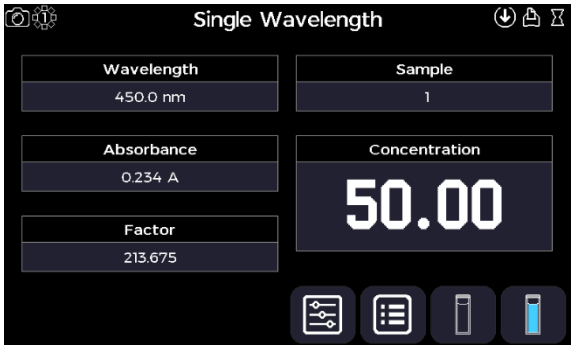


If “Absorbance”, “%Transmission” or “Concentration” mode with the “Predefined” factor method is selected, skip steps 17 through 18 and go straight to step 19.

Step 17
If “Concentration” mode with the “Standards” factor method is selected, replace the previous sample with a standard sample of known concentration then take a sample measurement using the sample measurement icon.



Step 18
Enter the concentration value of the standard sample. Confirm the settings using the confirm icon.



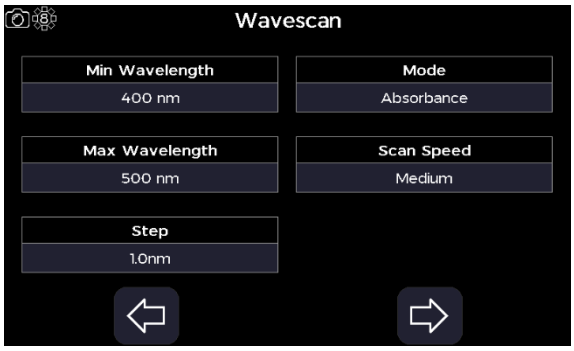
Step 19
Replace the previous sample with a test sample then take a sample measurement using the sample measurement icon.

Repeat for all samples.

Step 20
Return to the Applications screen using the exit icon in the options menu **OR** use the additional options to save, print, and, or load previous measurements (see the Additional Options section).

Wavescan

The Wavescan application is selected from the Applications screen. It can be used to perform absorbance (A) or % transmission (%T) measurements across a range of wavelengths creating an absorbance, or transmission, spectrum.



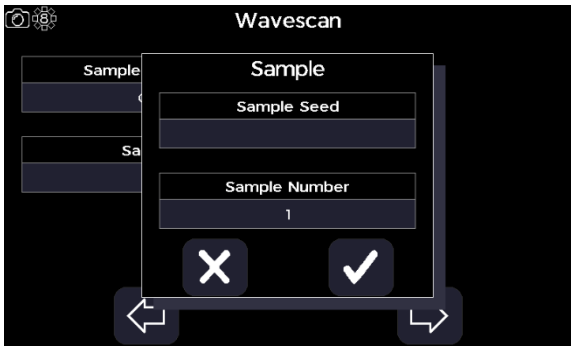
Step 1
Set the minimum wavelength to between 190 to 1090 nm.

Step 2
Set the maximum wavelength to between 200 to 1100 nm.

Step 3
Select the step size from “1.0nm”, “0.5nm”, “0.2nm”, or “0.1nm”.

Step 4
Select the mode from “Absorbance” or “%Transmission”.

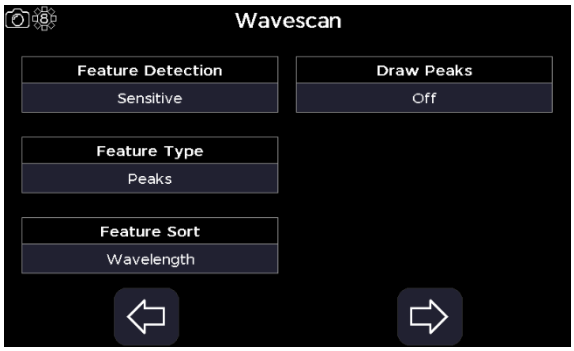
Step 5
Select the scan speed from “Slow”, “Medium”, “Fast”, or “Survey”.



Step 6
Proceed to the next parameter screen using the right/forward arrow.

Step 7
Set the number of sample overlays limit to “Off”, “2”, “3”, “4”, “5”, “6”, “7”, or “8”.

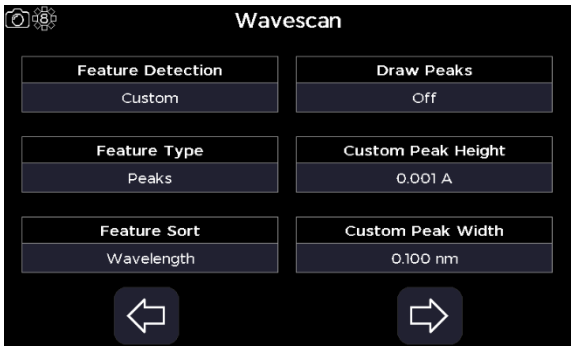
Step 8
Select the sample name to bring up the sample window, enter a sample seed prefix and the incremental sample number starting value. Confirm the settings using the confirm icon.



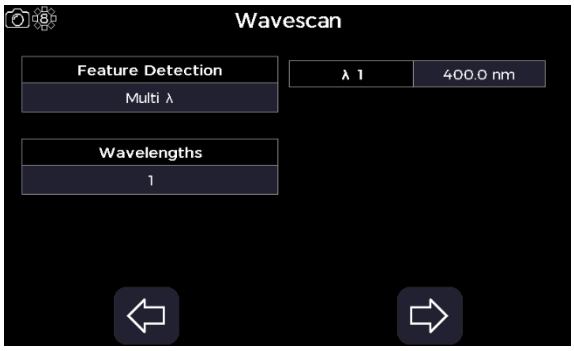
Step 9
Proceed to the next parameter screen using the right/forward arrow.

Step 10
Set feature detection to “Off”, “Coarse”, “Sensitive”, “Custom”, or “Multi λ”.

Step 11
If feature detection is set to “Coarse”, “Sensitive”, or “Custom” select the trigger feature type from “Peaks” or “Valleys”, the feature sort from “Wavelength” or “Magnitude”, and the draw peaks from “On” or “Off”.

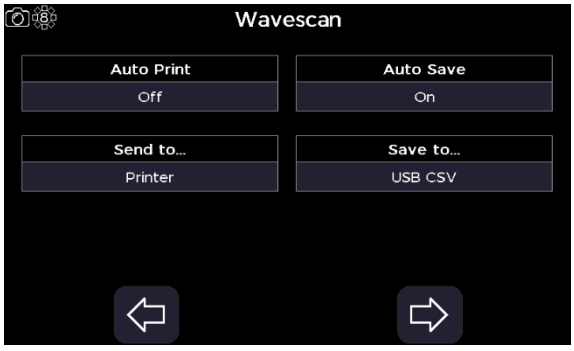


For “Custom” feature detection, also set the custom peak height and width triggers.



For “Multi λ” feature detection, set the number of wavelengths to extract the absorbance at to “1”, “2”, “3”, “4”, “5”, “6”, “7”, or “8”. Then set those wavelengths to between the previously defined minimum and maximum wavelengths, 190 – 1100nm

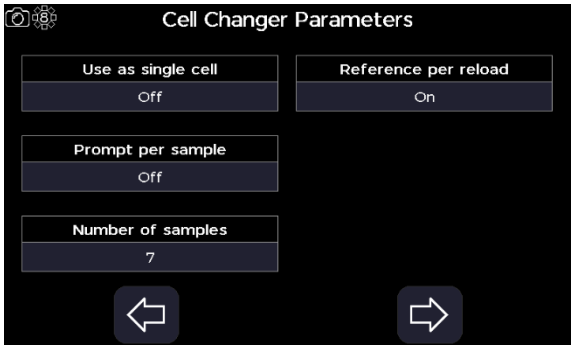
---



Step 12
Proceed to the next parameter screen using the right/forward arrow.

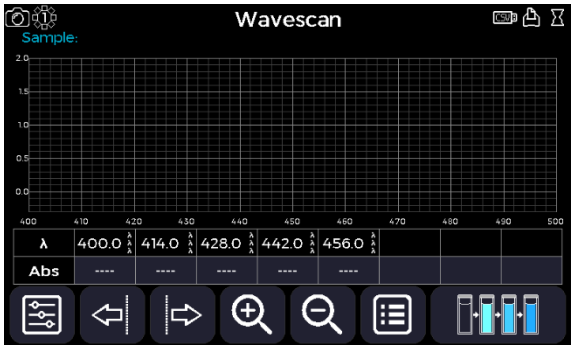
Step 13
Set auto print to “On” or “Off”. If auto print is set to “On”, select the print to hardware from “Internal Printer”, “PC via USB”, or “USB Mass Storage” depending on what hardware is connected to the instrument.

Step 14
Set auto save to “On” or “Off”. If auto save is set to “On”, select the save to hardware from “USB CSV”, “USB”, or “Internal” depending on what hardware is connected to the instrument.



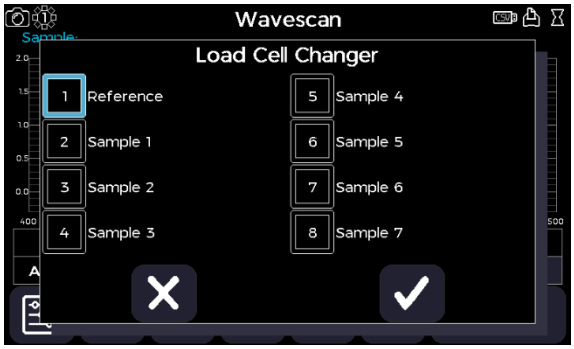
Step 15
Proceed to the next parameter screen using the right/forward arrow.

Step 16
Set whether to use as single cell to “On” or “Off”. If set to “Off”, set the position prompt per sample to “On” or “Off”, set the number of samples to between 2 and 100, and set whether to retake the reference between reload to “On” of “Off”.

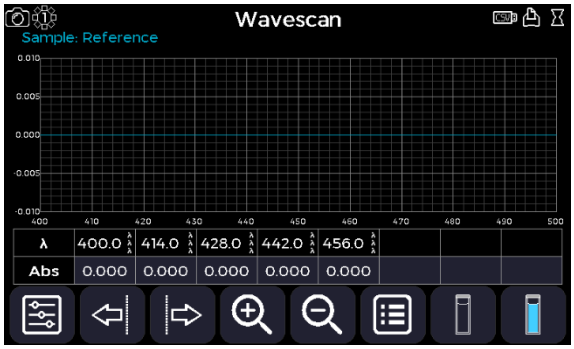


Step 17
Proceed to the measurement screen using the right/forward arrow.

If using a single cell holder, or a cell changer set to use as a single cell holder, skip step 18 and go straight to step 19.



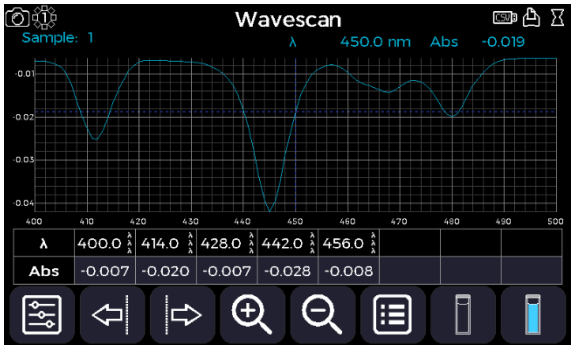
Step 18
Press the batch measurement icon, then load the cell changer according to the cell changer prompt. Then select the confirm icon.



If using a cell changer, skip steps 19 through 20 and go straight to step 21.

Step 19
Insert the reference sample then take a reference measurement using the reference measurement icon.

The acquired reference sample baseline will be applied to all subsequent sample measurements.



Step 20
Replace the previous sample with a test sample then take a sample measurement using the sample measurement icon.

Repeat for all samples.

Step 21
If sample overlays are active, the sample overlay tools will appear to the right of the displayed spectra.

	**Select data**	Select the sample overlay to be analysed
	**Display data**	Hide or show sample overlays

	**Delete data**	Delete sample overlays from the overlay tools list

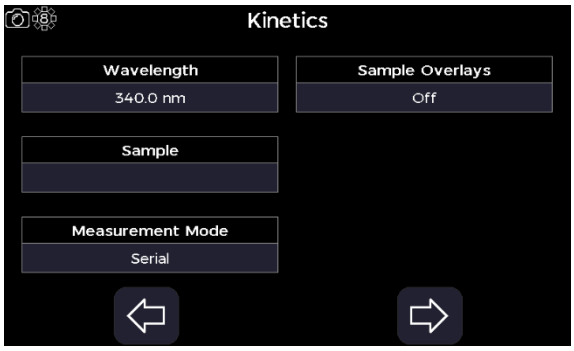
Additional viewing tools as available at the bottom of the screen.

	**Cursor left**	Move the x-axis cursor position left
	**Cursor right**	Move the x-axis cursor position right
	**Zoom in**	Zoon into the area around the x and y-axis cursor position
	**Zoom out**	Zoon out from the area around the x and y-axis cursor position

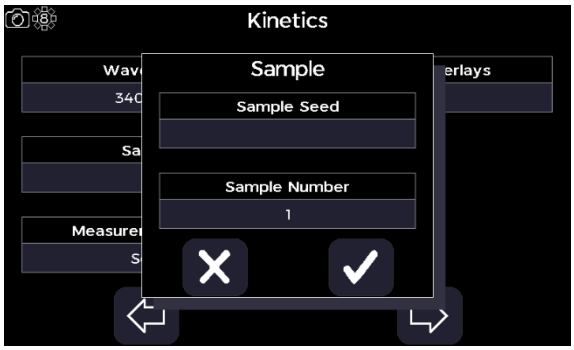
Step 22
Return to the Applications screen using the exit icon in the options menu **OR** use the additional options to save, print, and, or load previous measurements (see the Additional Options section).

Kinetics

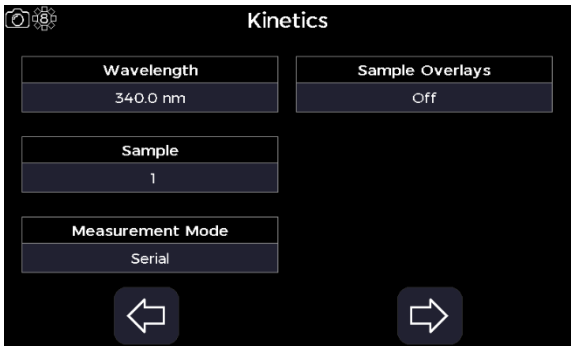
The Kinetics application is selected from the Applications screen. It can be used to perform a series of absorbance (A) measurements over a defined timeframe creating a time-course trace.



Step 1
Set the wavelength to between 190 and 1100 nm.

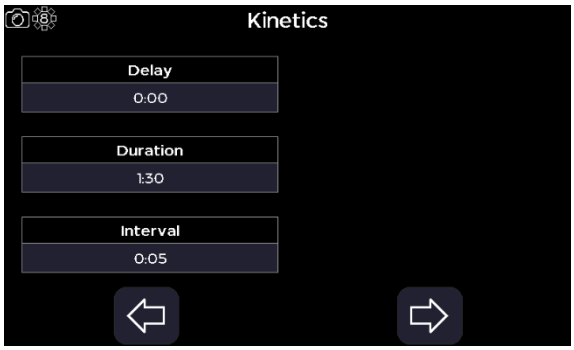


Step 2
Select the sample name to bring up the sample window, enter a sample seed prefix and the incremental sample number starting value. Confirm the settings using the confirm icon.



Step 3
Set the measurement mode to “Serial” or “Parallel”.

Step 4
Set the number of sample overlays limit to “Off”, “2”, “3”, “4”, “5”, “6”, “7”, or “8”.

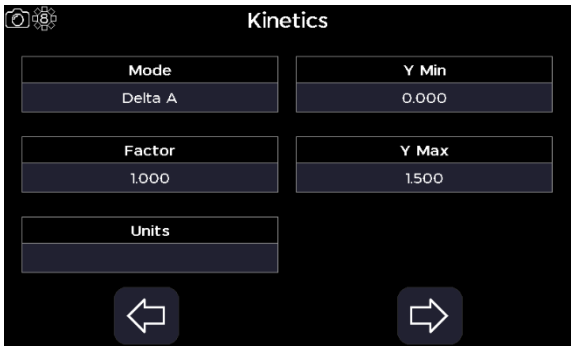


Step 6
Set the delay time before the first measurement to between 0 seconds and 99 minutes. Confirm the settings using the confirm icon.

Step 7
Set the duration of the observation to between 0 seconds and 180 minutes. Confirm the settings using the confirm icon.

Step 8
If the measurement mode is set to “Serial”, set the interval between individual measurements to between 2 seconds and 18 minutes. Confirm the settings using the confirm icon.

If the measurement mode is set to “Parallel”, select the interval between individual measurements from “10 Seconds”, “15 Seconds”, “20 Seconds”, “25 Seconds”, “30 Seconds”, “35 Seconds”, “40 Seconds”, “1 Minutes”, “2 Minutes”, “5 Minutes”, or “10 Minutes”.



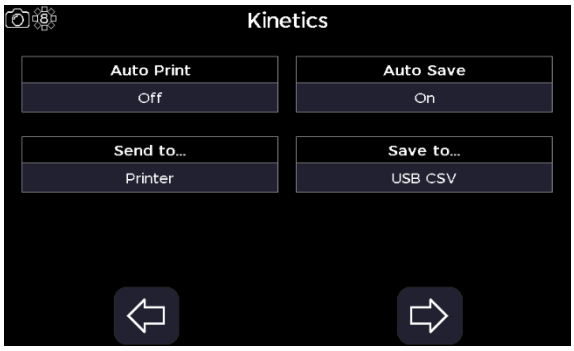
Step 9
Proceed to the next parameters screen using the right/forward arrow.

Step 10
Set the mode to define the desired result, “Delta A”, “Final A”, or “Slope”.

Step 11
Set the factor value to be applied to the result, between 0.000 and ±9999.

Step 12
Define the units that the result value will be reported in.

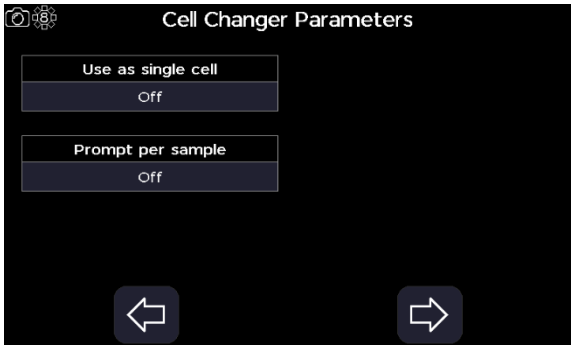
Step 13
Set the y-axis minimum and maximum to between -4 and 4.



Step 14
Proceed to the next parameter screen using the right/forward arrow.

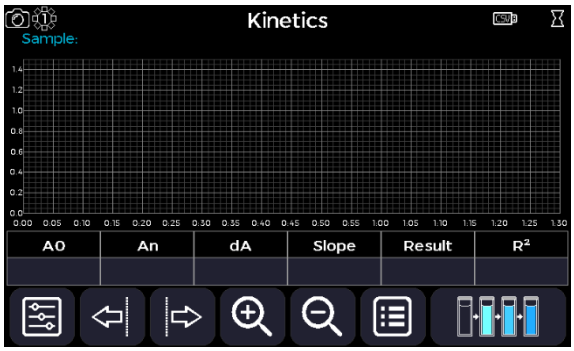
Step 15
Set auto print to “On” or “Off”. If auto print is set to “On”, select the print to hardware from “Internal Printer”, “PC via USB”, or “USB Mass Storage” depending on what hardware is connected to the instrument.

Step 16
Set auto save to “On” or “Off”. If auto save is set to “On”, select the save to hardware from “USB CSV”, “USB”, or “Internal” depending on what hardware is connected to the instrument.



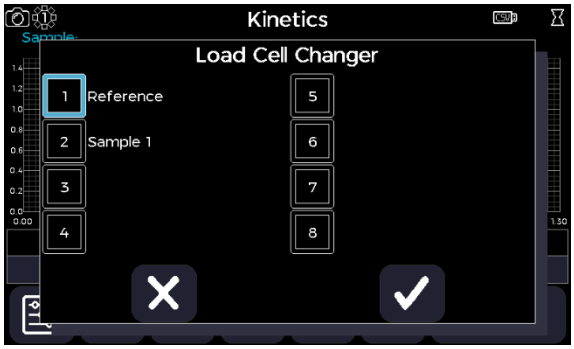
Step 17
Proceed to the next parameter screen using the right/forward arrow.

Step 18
Set whether to use as single cell to “On” or “Off”. If set to “Off”, set the position prompt per sample to “On” or “Off”.

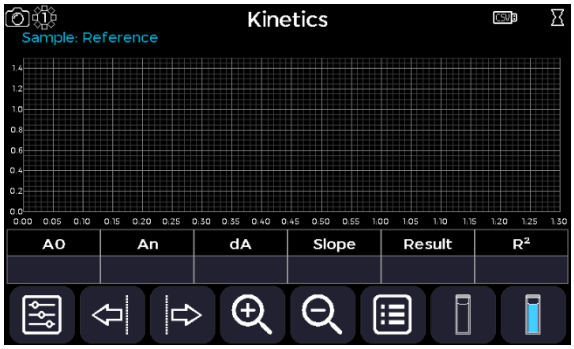


Step 18
Proceed to the measurement screen using the right/forward arrow.

If using a single cell holder, or a cell changer set to use as a single cell holder, skip step 19 and go straight to step 20.



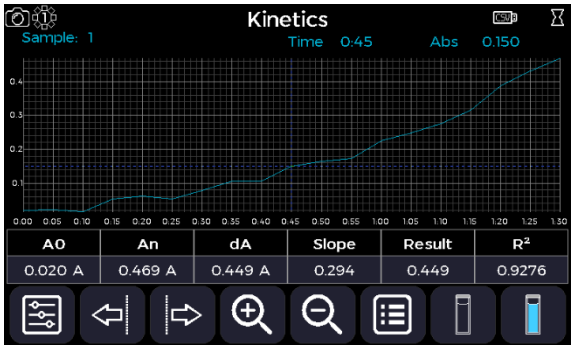
Step 19
Press the batch measurement icon, then load the cell changer according to the cell changer prompt. Then select the confirm icon.



If using a cell changer, skip steps 20 through 21 and go straight to step 22.

Step 20
Insert the reference sample then take a reference measurement using the reference measurement icon.

The acquired reference sample baseline will be applied to all subsequent sample measurements.



Step 21
Replace the previous sample with a test sample then take a sample measurement using the sample measurement icon.

Repeat for all samples.

Step 22
If sample overlays are active, the sample overlay tools will appear to the right of the displayed spectra.

	**Select data**	Select the sample overlay to be analysed
	**Display data**	Hide or show sample overlays
	**Delete data**	Delete sample overlays from the overlay tools list

Additional viewing tools as available at the bottom of the screen.

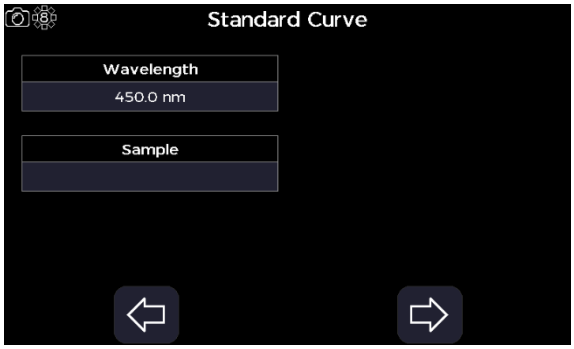
	**Cursor left**	Move the x-axis cursor position left
	**Cursor right**	Move the x-axis cursor position right
	**Zoom in**	Zoon in to the area around the x and y-axis cursor position

	**Zoom out**	Zoon out from the area around the x and y-axis cursor position

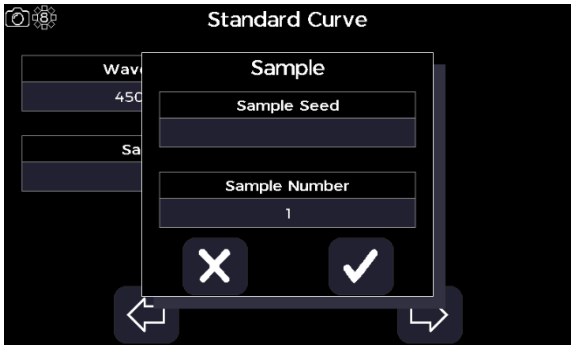
Step 23
Return to the Applications screen using the exit icon in the options menu **OR** use the additional options to save, print, and, or load previous measurements (see the Additional Options section).

Standard Curve

The Standard Curve application is selected from the Applications screen. It can be used to create a calibration curve from standard samples of known concentration. The curve fit equation is then applied to the absorbance (A) measurements of any subsequent test samples to determine their concentration.

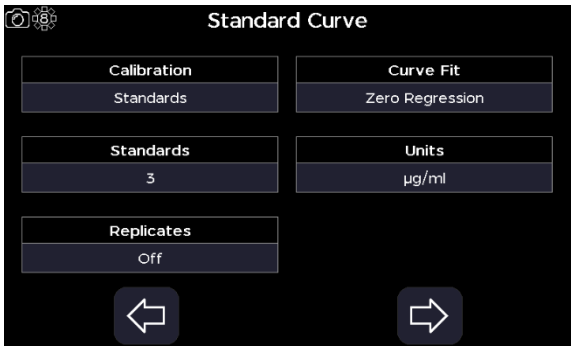


Step 1
Set the wavelength to between 190 and 1100 nm.



Step 2
Select the sample name to bring up the sample window, enter a sample seed prefix and the incremental sample number starting value. Confirm the settings using the confirm icon.

Step 3
Proceed to the next parameter screen using the right/forward arrow.

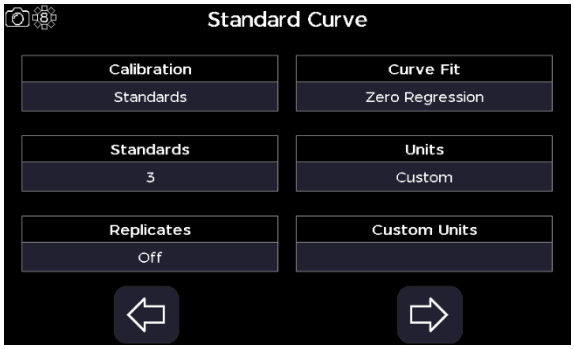


Step 4
Select the source of the calibration to “Standards” or “Manual”.

Step 5
Select the number of standard samples of known concentration to “1”, “2”, “3”, “4”, “5”, “6”, “7”, “8”, or “9”.

Step 6
If the source of the calibration is set to “Standards”, select the number of standard sample replicates to “Off”, “2”, or “3”.

Step 7
Select the curve fit, “Regression”, “Interpolation”, “Cubic Spline”, “Zero Regression”, or “2nd Order Polynomial”.



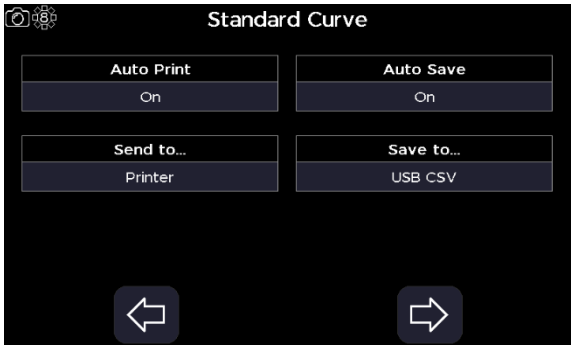
Step 8
Select one of the predefined units; “µg/ml”, “ng/µl”, or “µg/µl”.

OR

Select “Custom” and define the custom units that the concentration value will be reported in.

Step 9
Proceed to the next parameter screen using the right/forward arrow.

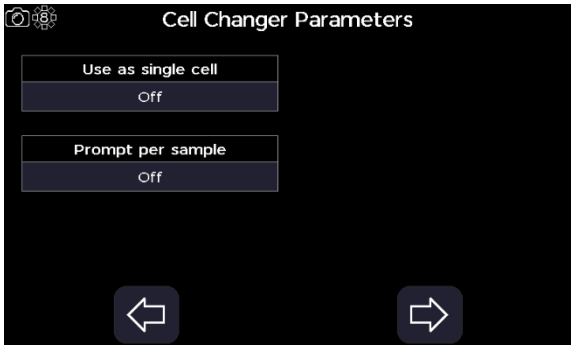
Step 10
Enter the concentration values of the standards samples between -9999 to 9999. Confirm the settings using the confirm icon.



Step 11
Proceed to the next parameter screen using the right/forward arrow.

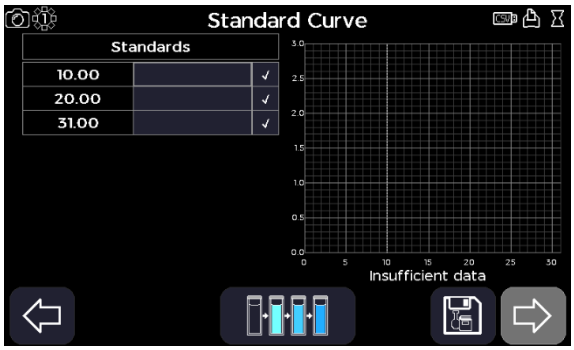
Step 12
Set auto print to “On” or “Off”. If auto print is set to “On”, select the print to hardware from “Internal Printer”, “PC via USB”, “USB Mass Storage” depending on what hardware is connected to the instrument.

Step 13
Set auto save to “On” or “Off”. If auto save is set to “On”, select the save to hardware from “USB CSV”, “USB”, or “Internal” depending on what hardware is connected to the instrument.



Step 14
Proceed to the next parameter screen using the right/forward arrow.

Step 15
Set whether to use as single cell to “On” or “Off”. If set to “Off”, set the position prompt per sample to “On” or “Off”.

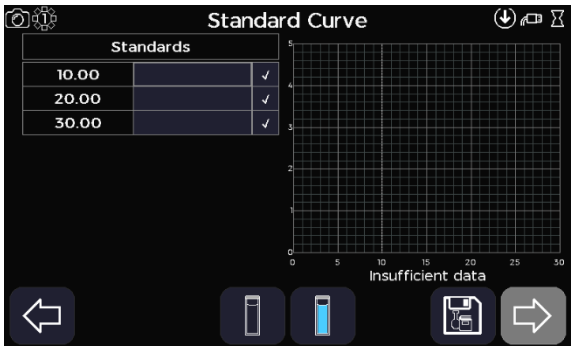


If the source of the calibration is set to “Manual”, skip steps 16 through 23 and go to step 25.

If using a single cell holder, or a cell changer set to use as a single cell holder, skip steps 16 through 17 and go straight to step 18.

Step 16
Proceed to the next parameter screen using the right/forward arrow.

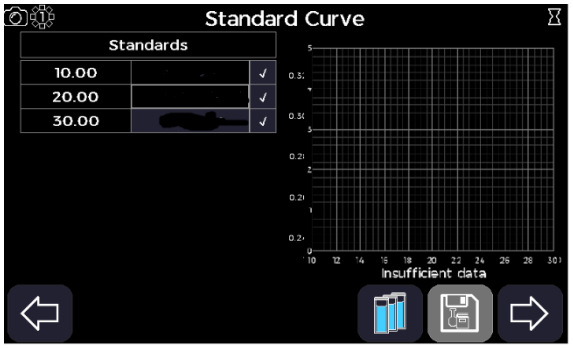
If using a cell changer, skip steps 18 through 23 and go straight to step 26.



Step 18
Proceed to the next parameter screen using the right/forward arrow.

Step 19
Take a reference using the reference icon, then insert standards and take each standard measurement by using the measurement icon

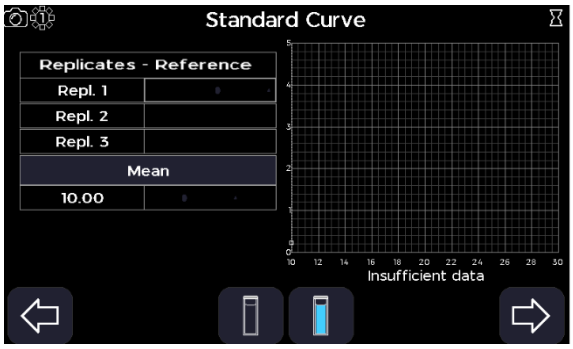
If not using replicates, ship to step 26



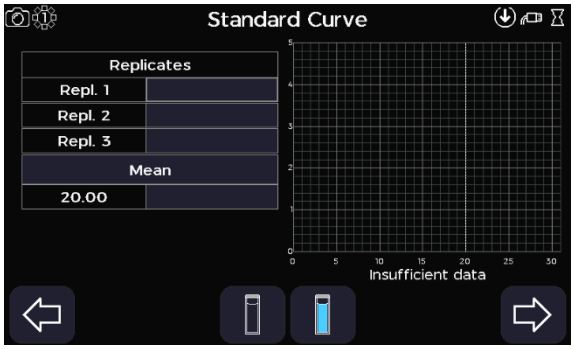
Step 20
If using replicate standards, run the standards by selecting the replicates icon.

Insert the reference sample then take a reference measurement using the reference measurement icon  .

The acquired reference sample baseline will be applied to all subsequent standard sample measurements.



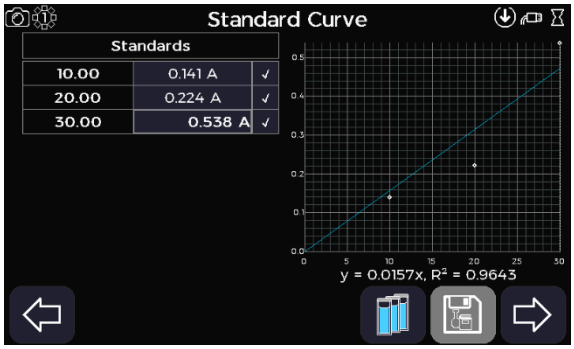
Step 21
Replace the reference sample with the first standard sample then take a sample measurement using the sample measurement icon for each replicate of that standard sample.



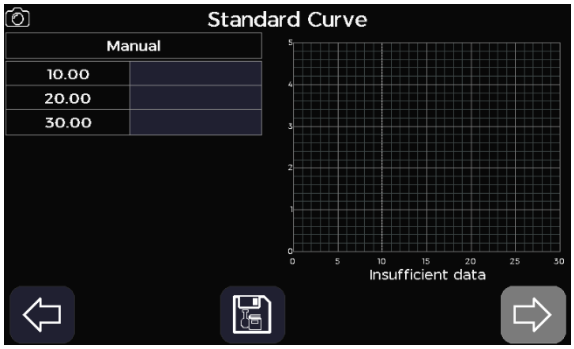
Step 22
Proceed to the next standard measurement screen using the right/forward arrow.

Step 23
Replace the previous standard sample with the next standard sample then take a sample measurement using the sample measurement icon for each replicate of that standard sample.

Repeat for all remaining standard samples if applicable.

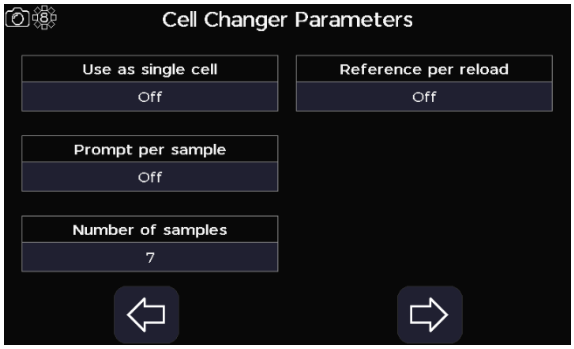


Step 24
Leave the replicates function using the left/backward arrow.



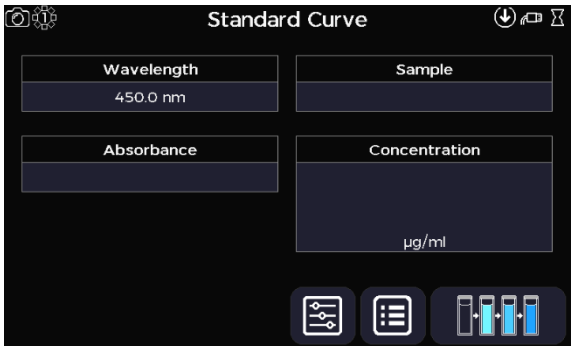
Step 25
If the source of the calibration is set to “Standards”, skip step 25 and go to step 26.

If source of the calibration is set to “Manual”. Define each standards absorbance value by selecting the appropriate text box and entering a value between -0.3 and 3.0 A.

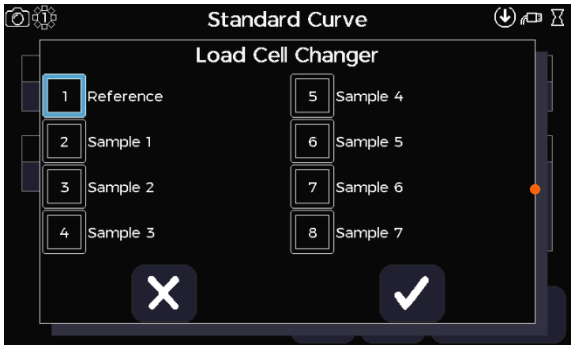


Step 26
Proceed to the next parameter screen using the right/forward arrow.

Step 27
Set whether to use as single cell to “On” or “Off”. If set to “Off”, set the position prompt per sample to “On” or “Off”, set the number of samples to between 2 and 100, and set whether to retake the reference between reload to “On” of “Off”.

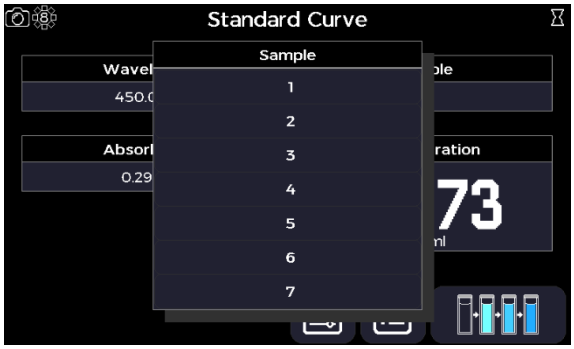


Step 28
Proceed to the measurement screen using the right/forward arrow.

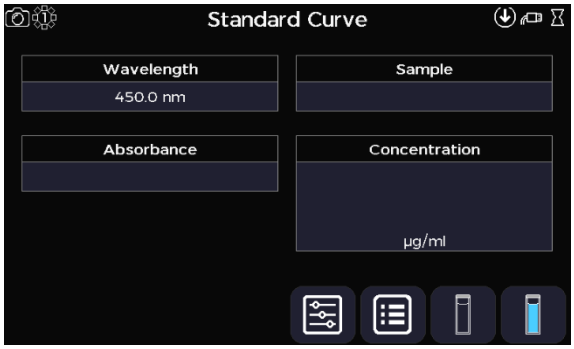


If using a single cell holder, or a cell changer set to use as a single cell holder, skip steps 29 through 30 and go straight to step 31.

Step 29
Press the batch measurement icon, then load the cell changer according to the cell changer prompt. Then select the confirm icon.



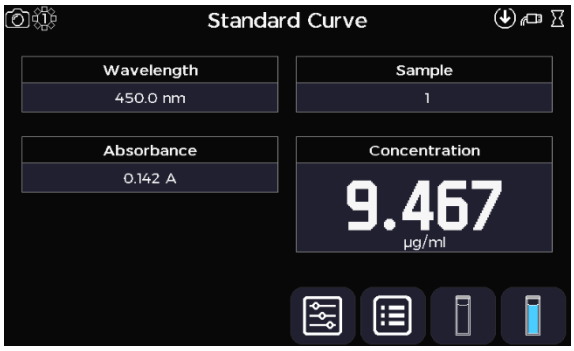
Step 30
The acquired reference sample baseline will be applied to all subsequent sample measurements. The sample measurements can be viewed by pressing the sample name test box and selecting the appropriate sample from the list.



If using a cell changer, skip steps 31 through 32 and go straight to step 33.

Step 31
Insert the reference sample then take a reference measurement using the reference measurement icon.

The acquired reference sample baseline will be applied to all subsequent sample measurements.



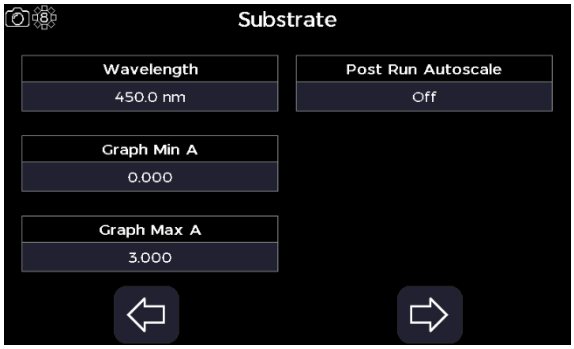
Step 32
Replace the previous sample with a test sample then take a sample measurement using the confirm key.

Repeat for all samples.

Step 33
Return to the Applications screen using the exit icon in the options menu **OR** use the additional options to save, print, and, or load previous measurements (see the Additional Options section).

Substrate

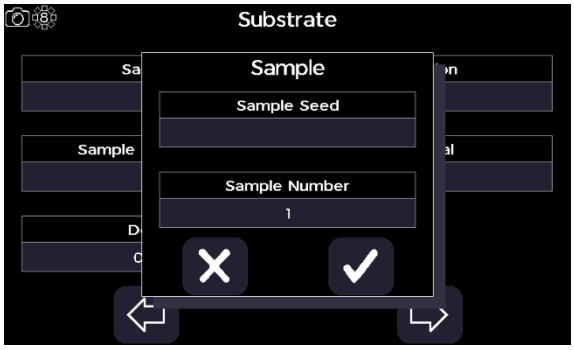
The Substrate application is selected from the Applications screen. It can be used to create a calibration curve from kinetic measurements of samples of known concentrations. The curve fit equation is then applied to the absorbance (A) measurements of any subsequent test samples to determine their concentration.



Step 1
Set the wavelength to between 190 and 1100 nm.

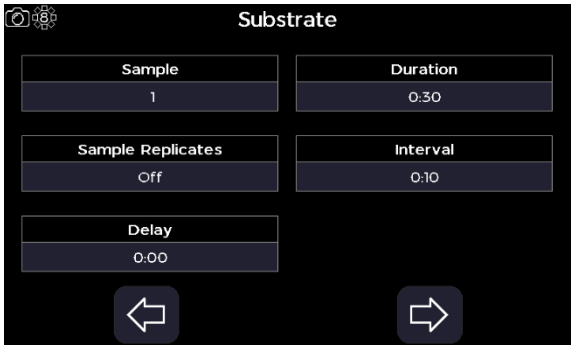
Step 2
Set the y-axis minimum and maximum to between -4 and 4.

Step 3
Set the post run autoscale to “On” or “Off”.



Step 4
Proceed to the next parameter screen using the right/forward arrow.

Step 5
Select the sample name to bring up the sample window, enter a sample seed prefix and the incremental sample number starting value. Confirm the settings using the confirm icon.

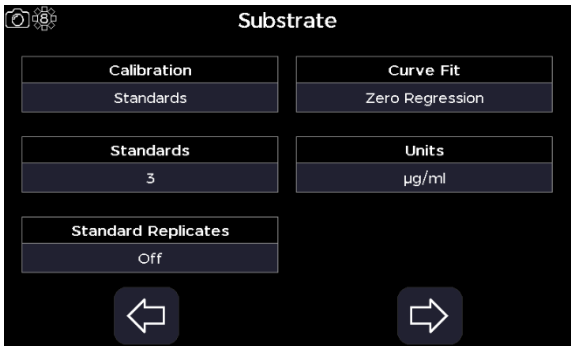


Step 6
Select the number of test sample replicates to “Off”, “2”, or “3”.

Step 7
Set the delay time before the first measurement to between 0 seconds and 99 minutes. Confirm the settings using the confirm icon.

Step 8
Set the duration of the observation to between 0 seconds and 180 minutes. Confirm the settings using the confirm icon.

Step 9
Set the interval between individual measurements to between 5 seconds and 18 minutes. Confirm the settings using the confirm icon.



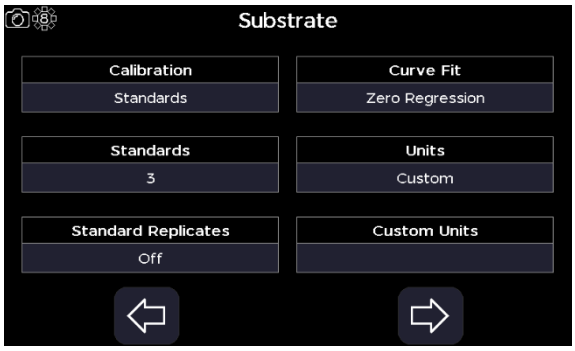
Step 10
Proceed to the next parameter screen using the right/forward arrow.

Step 11
Select the source of the calibration to “Standards” or “Manual”.

Step 12
Select the number of standard samples of known concentration to “1”, “2”, “3”, “4”, “5”, “6”, “7”, “8”, or “9”.

Step 13
If the source of the calibration is set to “Standards”, select the number of standard sample replicates to “Off”, “2”, or “3”.

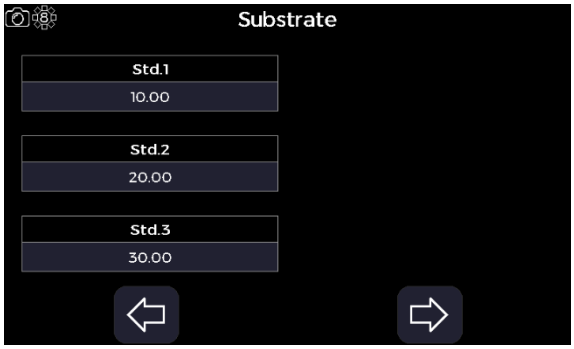
Step 14
Select the curve fit, “Regression”, “Interpolation”, “Cubic Spline”, “Zero Regression”, or “2nd Order Polynomial”.



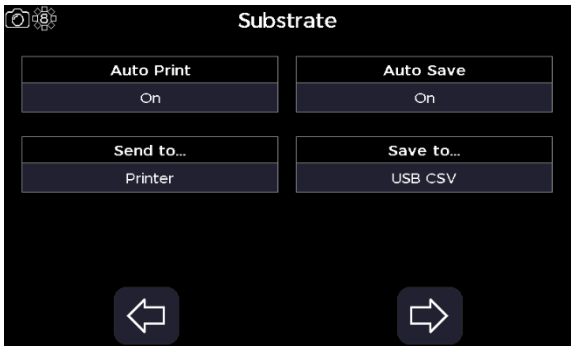
Step 15
Select one of the predefined units; “µg/ml”, “ng/µl”, or “µg/µl”.

OR

Select “Custom” and define the custom units that the concentration value will be reported in.



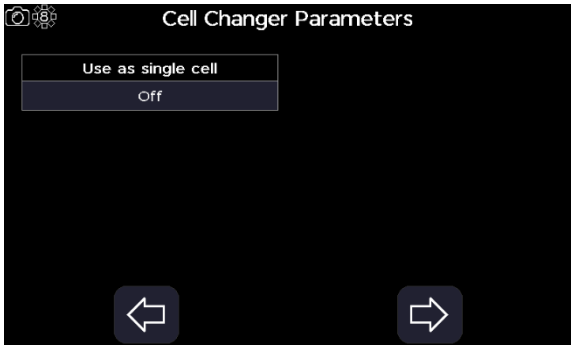
Step 16
Enter the concentration values of the standards samples between -9999 to 9999. Confirm the settings using the confirm icon.



Step 17
Proceed to the next parameter screen using the right/forward arrow.

Step 18
Set auto print to “On” or “Off”. If auto print is set to “On”, select the print to hardware from “Internal Printer”, “PC via USB”, or “USB Mass Storage” depending on what hardware is connected to the instrument.

Step 19
Set auto save to “On” or “Off”. If auto save is set to “On”, select the save to hardware from “USB CSV”, “USB”, or “Internal” depending on what hardware is connected to the instrument.

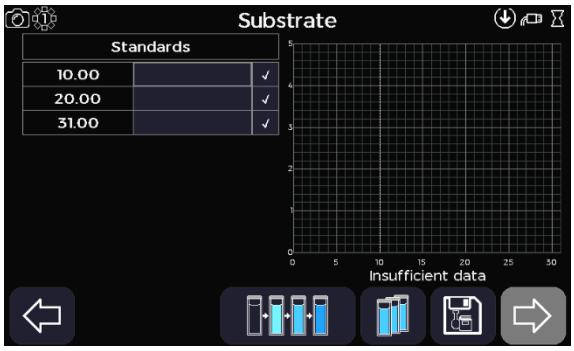


Step 20
Proceed to the next parameter screen using the right/forward arrow.

Step 21
Set whether to use as single cell to “On” or “Off”.

Step 22
Proceed to the next parameter screen using the right/forward arrow

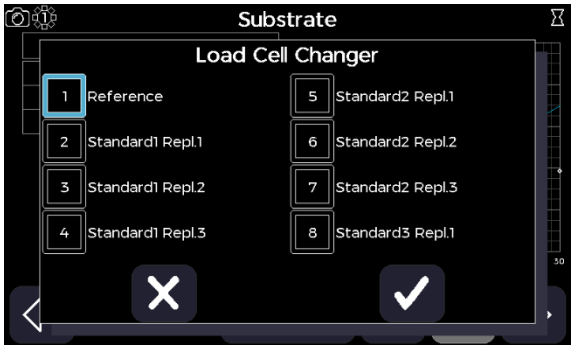
---



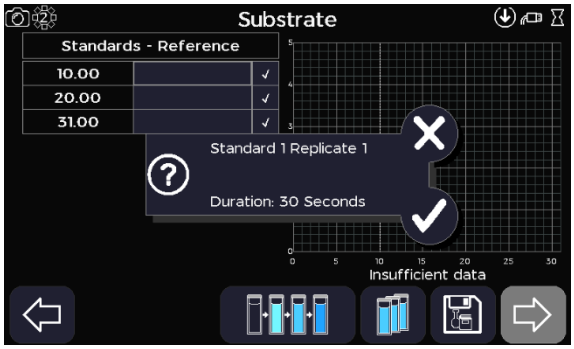
If the source of the calibration is set to “Manual”, skip steps 23 through 30 and go to step 31.

If using a single cell holder, or a cell changer set to use as a single cell holder, skip steps 23 through 24 and go straight to step 25.

Step 23
Press the batch measurement icon, then load the cell changer according to the cell changer prompt.

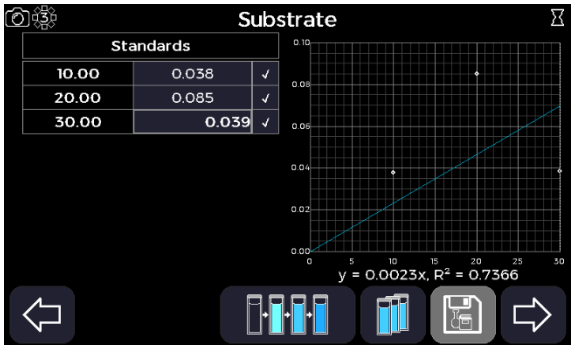


Step 24
Then select the confirm icon.



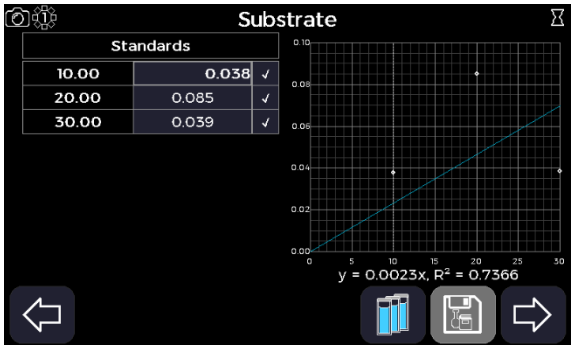
Step 25
Acknowledge the on-screen prompts by selecting the confirm icon.

Repeat for all replicate and standard samples.



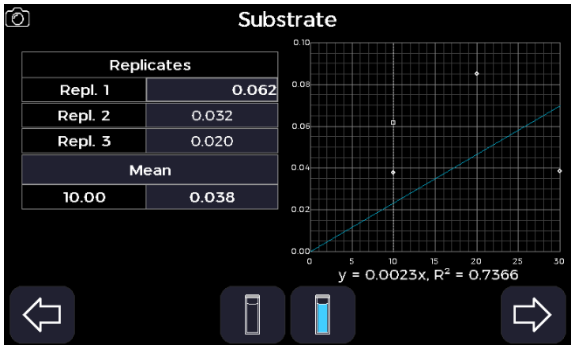
If using replicates, any of the standards or replicates can be re-run, by highlighting the appropriate standard and selecting the replicates icon.

Cycle through the standards using the right/forward arrow, and leave the replicates function by cycling back through the standards using the left/backward arrow.



If using a cell changer, skip steps 26 through 30 and go straight to step 31.

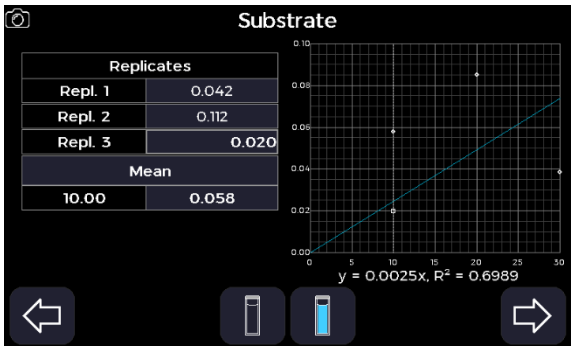
Step 26
Proceed to the next parameter screen using the right/forward arrow.



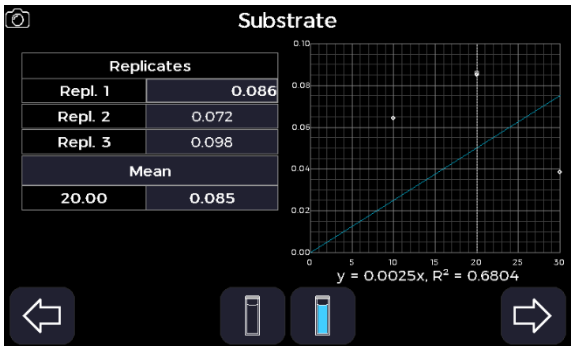
Step 27
Run the standards by selecting the replicates icon.

Insert the reference sample then take a reference measurement using the reference measurement icon.

The acquired reference sample baseline will be applied to all subsequent standard sample measurements.



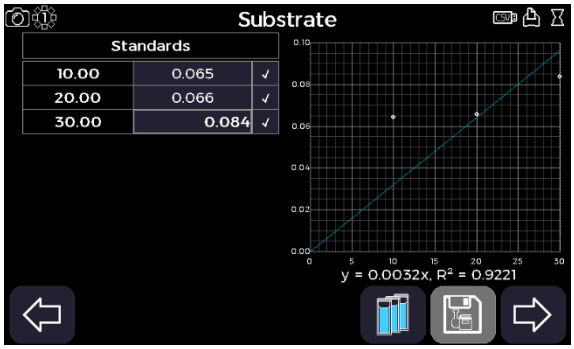
Step 28
Replace the reference sample with the first standard sample then take a sample measurement using the sample measurement icon for each replicate of that standard sample.



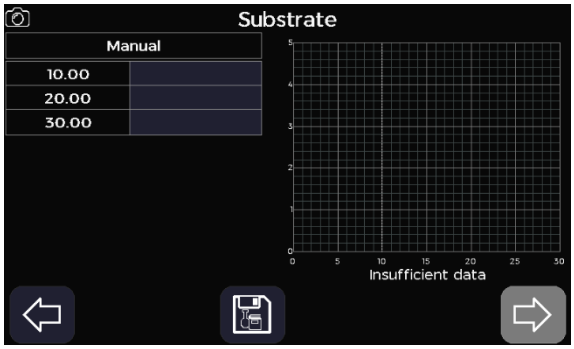
Step 29
Proceed to the next standard measurement screen using the right/forward arrow.

Step 30
Replace the previous standard sample with the next standard sample then take a sample measurement using the sample measurement icon for each replicate of that standard sample.

Repeat for all remaining standard samples if applicable.

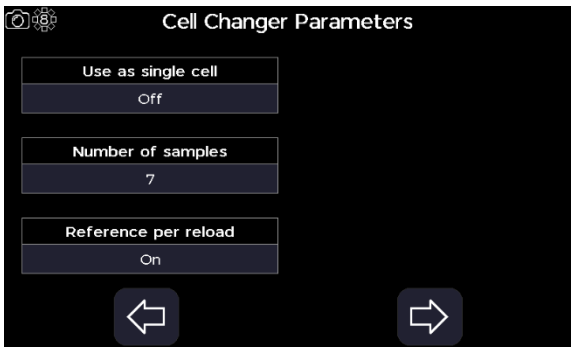


Step 31
Leave the replicates function using the left/backward arrow.



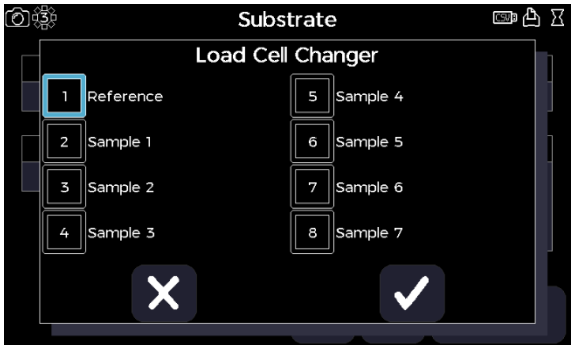
Step 32
If the source of the calibration is set to “Standards”, skip step 32 and go to step 33.

If source of the calibration is set to “Manual”. Define each standards absorbance value by selecting the appropriate text box and entering a value between -0.3 and 3.0 A.



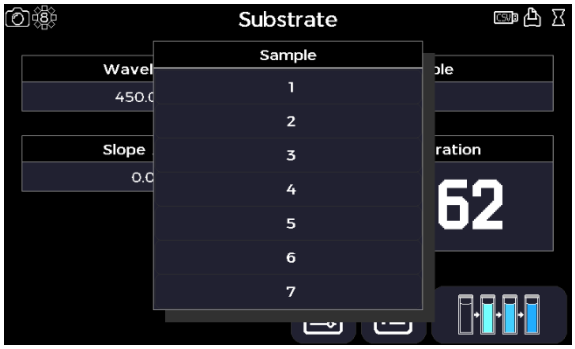
Step 33
Proceed to the next parameter screen using the right/forward arrow.

Step 34
Set whether to use as single cell to “On” or “Off”. If set to “Off”, set the position prompt per sample to “On” or “Off”, set the number of samples to between 2 and 100, and set whether to retake the reference between reload to “On” of “Off”.

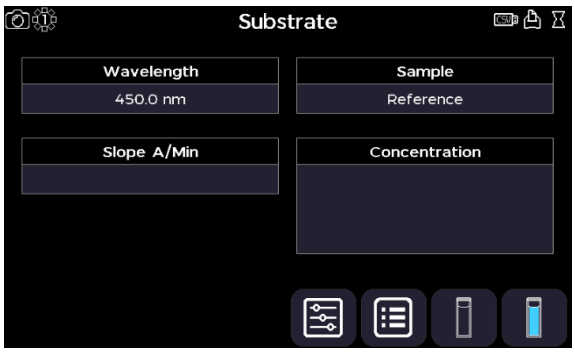


If using a single cell holder, or a cell changer set to use as a single cell holder, skip steps 35 through 36 and go straight to step 37.

Step 35
Press the batch measurement icon, then load the cell changer according to the cell changer prompt. Then select the confirm icon.



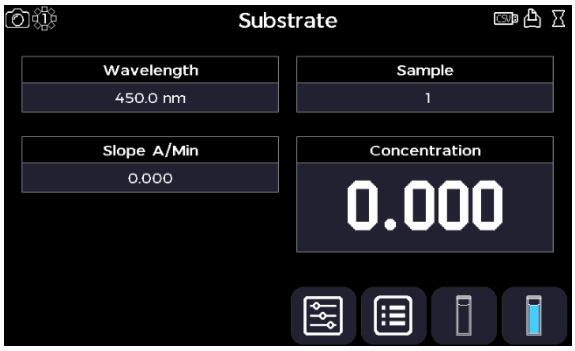
Step 35
The acquired reference sample baseline will be applied to all subsequent sample measurements. The sample measurements can be viewed by pressing the sample name test box and selecting the appropriate sample from the list.



If using a cell changer, skip steps 36 through 37 and go straight to step 38.

Step 36
Insert the reference sample then take a reference measurement using the reference measurement icon.

The acquired reference sample baseline will be applied to all subsequent sample measurements.



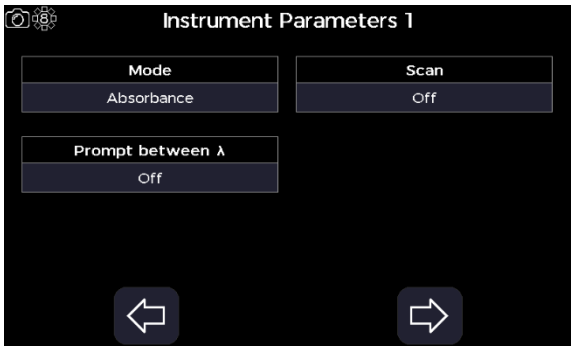
Step 37
Replace the previous sample with a test sample then take a sample measurement using the confirm key.

Repeat for all samples.

Step 38
Return to the Applications screen using the exit icon in the options menu **OR** use the additional options to save, print, and, or load previous measurements (see the Additional Options section).

Equation Editor

The Equation Editor application is selected from the Applications screen. It can be used to create more complex custom methods incorporating bespoke calculations.



Step 1
Select the mode from “Absorbance” or “%Transmission”.

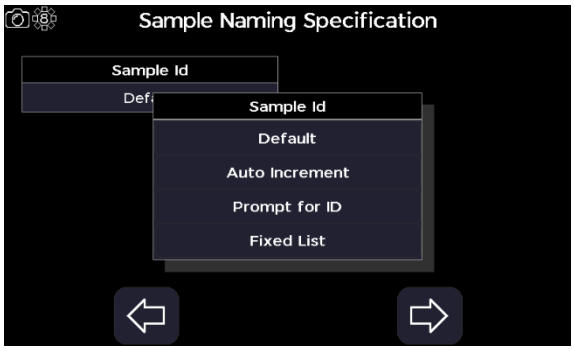
Step 2
Set sample message prompt between λ to “On” or “Off”.

Step 3
Set scan to “On” or “Off”.

Step 4
Proceed to the next parameter screen using the right/forward arrow.

Step 5
To define the measurements to be taken per sample:

- Under the “Name” column, allocate a name to the measurement.
- Under the “λ” column, set the wavelength to between 190 and 1100 nm, that the measurement will taken at.
- Under the “Function” column, select the data extracted by measurement from “Abs/%T at λ”, “Peak closest to λ”, or “Valley closest to λ”.
- For the peak and valley functions, under the “+/-“ column select the search tolerance from “1nm”, “2nm”, “5nm”, “10nm”, or “20nm”.

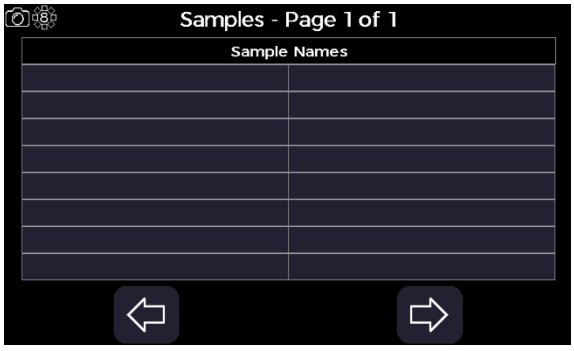


Step 6
Proceed to the next parameter screen using the right/forward arrow.

Step 7
Select the sample Id mode from “Default”, “Auto Increment”, “Prompt for ID”, or “Fixed List”.

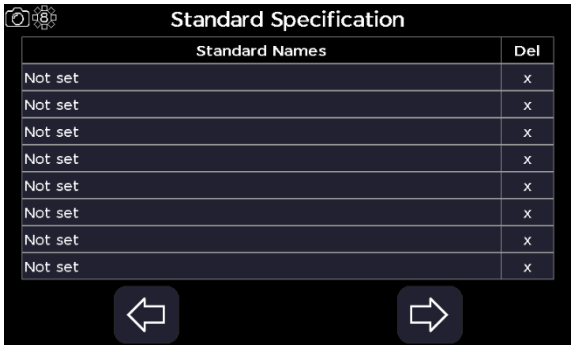
Step 8
If the sample Id is set to “Default”, “Auto Increment”, or “Prompt for ID” skip steps 8 through 10 and go to step 11.

If the sample Id is set to “Fixed List”, define the number of samples to between 1 and 99.



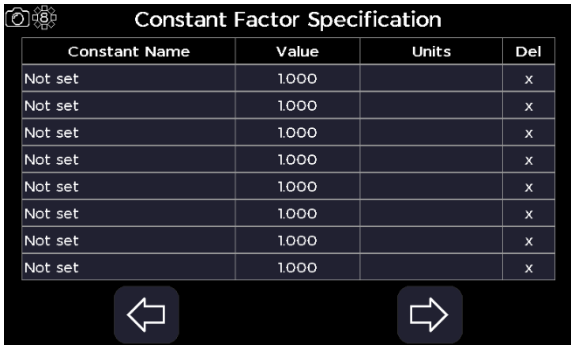
Step 9
Proceed to the next parameter screen using the right/forward arrow.

Step 10
Enter the sample Ids in the “Sample Names” table. If the list comprises multiple pages, us the up and down page arrows to change the page:



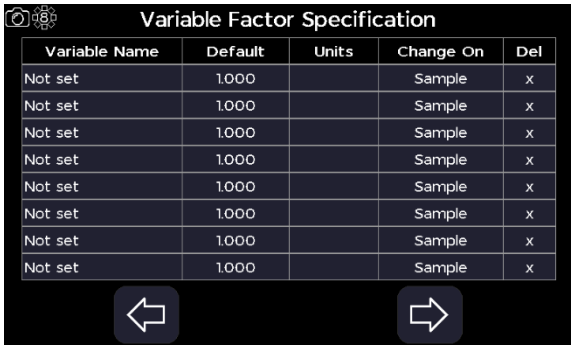
Step 11
Proceed to the next parameter screen using the right/forward arrow.

Step 12
Enter the standard names of any standard sample measurement data to be applied to the final equations.



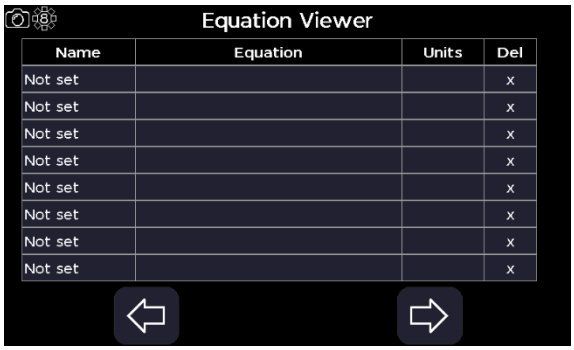
Step 13
Proceed to the next parameter screen using the right/forward arrow.

Step 14
Enter any constant factors to be applied to the final equations.



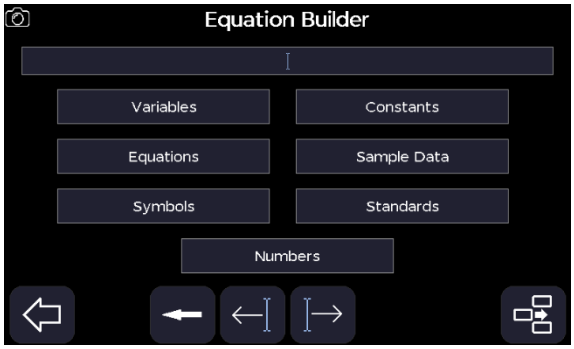
Step 15
Proceed to the next parameter screen using the right/forward arrow.

Step 16
Enter any variable factors to be applied to the final equations, select when the option to change the factor is presented in the “Change On” column to either between every “Sample” or between each “Batch”.



Step 17
Proceed to the next parameter screen using the right/forward arrow.

Step 18
Define the equation to be applied to the measurement.
Select the “Equation” column to open the equation builder screen, then select the features to incorporate into the equation being defined.



Variables	Select a defined variable factor.
Constants	Select a defined constant factor.
Equations	Select the results from a previous equation.
Sample Data	Select raw sample data.
Symbols	Select a mathematical operator.
Standards	Select a defined standard.
Numbers	Enter a fixed number.

Complete the current equation and close the equation builder screen using the left/backward arrow.

PLEASE NOTE

Only results generated from equations displayed, so even raw sample data needs defined as an equation.

---

n

Step 19

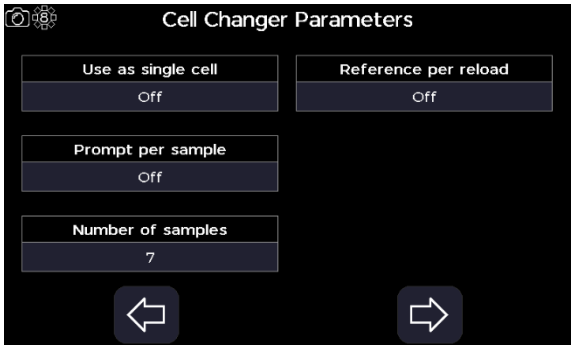
Proceed to the next parameter screen using the right/forward arrow.

Step 19

Set auto print to “On” or “Off”. If auto print is set to “On”, select the print to hardware from “Internal Printer”, “PC via USB”, or “USB Mass Storage” depending on what hardware is connected to the instrument.

Step 20

Set auto save to “On” or “Off”. If auto save is set to “On”, select the save to hardware from “USB CSV”, “USB”, or “Internal” depending on what hardware is connected to the instrument.

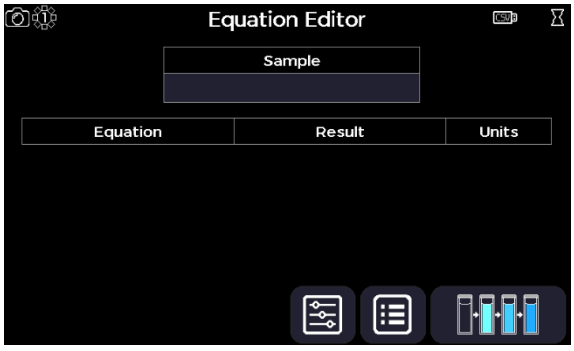


Step 21

Proceed to the next parameter screen using the right/forward arrow.

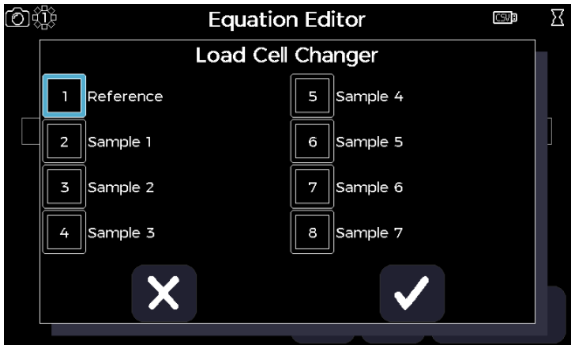
Step 22

Set whether to use as single cell to “On” or “Off”. If set to “Off”, set the position prompt per sample to “On” or “Off”, set the number of samples to between 2 and 100, and set whether to retake the reference between reload to “On” of “Off”.



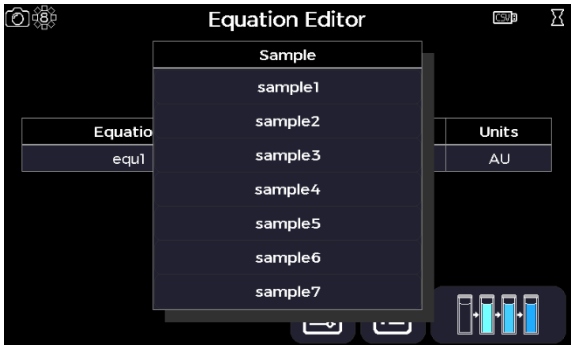
Step 23

Proceed to the measurement screen using the right/forward arrow.

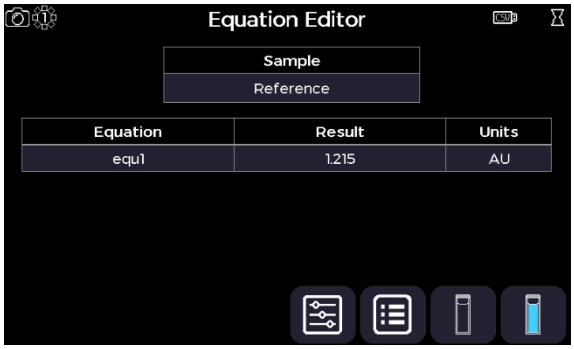


If using a single cell holder, or a cell changer set to use as a single cell holder, skip steps 24 through 25 and go straight to step 26.

Step 24
Press the batch measurement icon, then load the cell changer according to the cell changer prompt.



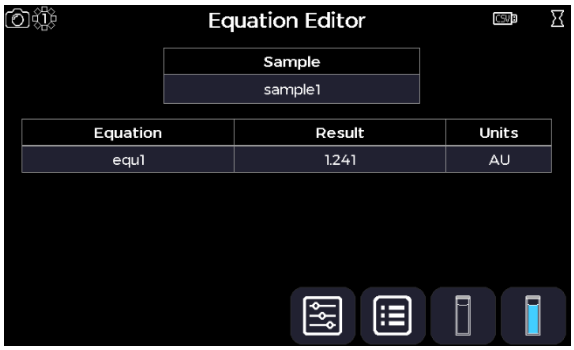
Step 25
The acquired reference sample baseline will have be applied to all subsequent sample measurements. The sample measurements can be viewed by pressing the sample name test box and selecting the appropriate sample from the list.



If using a cell changer, skip steps 26 through 27 and go straight to step 28.

Step 26
Insert the reference sample then take a reference measurement using the reference measurement icon.

The acquired reference sample baseline will be applied to all subsequent sample measurements.



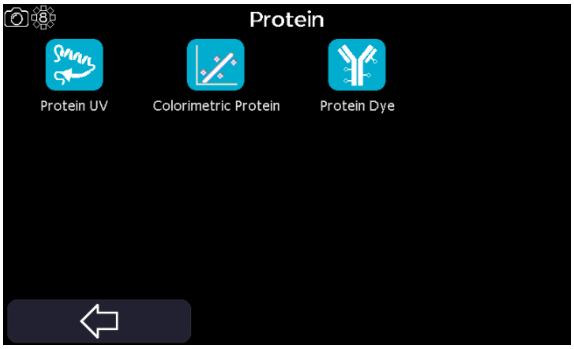
Step 27
Replace the previous sample with a test sample then take a sample measurement using the sample measurement icon.

Repeat for all samples.

Step 28
Return to the Applications screen using the exit icon in the options menu **OR** use the additional options to save, print, and, or load previous measurements (see the Additional Options section).

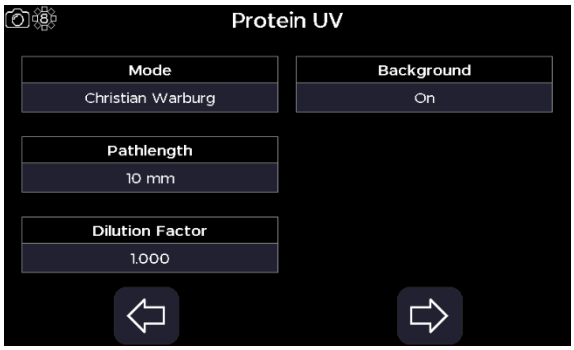
Protein

The Protein screen is accessed from the home screen. It contains predefined protein quantification methods and a protein dye application for fluorescent labelling efficiency of protein probes, based on the absorbance, prior to their use in microarrays. All calculations applied within the Protein applications are described in the Useful Calculation section.



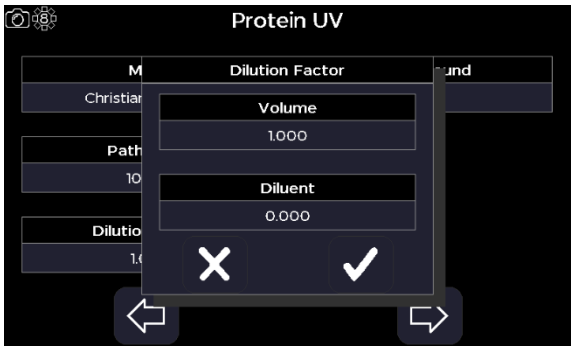
Protein UV

The Protein UV application is accessed from the Protein screen. It can be used to perform Protein quantification measurements at 280 nm and to assess sample quality using A260/A280 and A260/A230 absorbance (A) ratios.



Step 1
Set the mode; “Christian Warburg”, “BSA”, “IgG”, “Lysozyme”, “Molar extinction”, “Mass extinction”, “E 1%”, or “Custom”.

Step 2
Set the pathlength; “10 mm”, “Quantimate 500”, or, “Quantimate 200”.

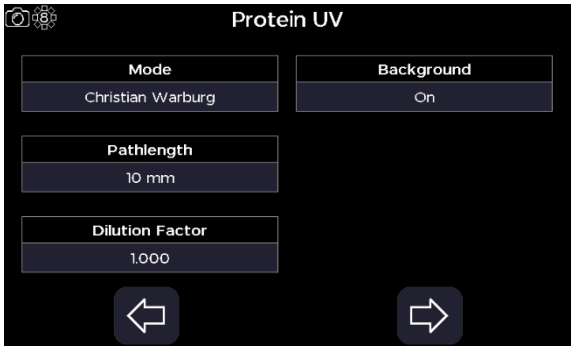


Step 3
Enter any dilution factor to be applied to the absorbance measurement.

Set the initial sample volume of a value of up to four significant figures.

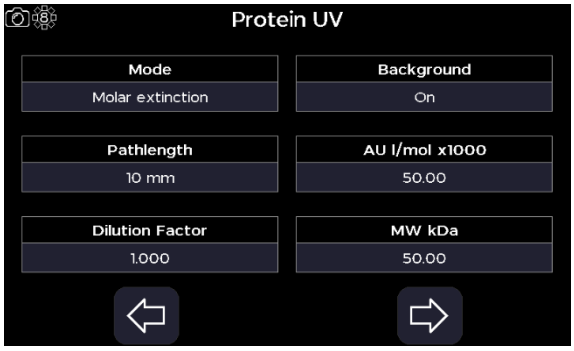
Then set the amount of diluent added to the initial volume of a value of up to four significant figures.

Implement the changes and return to the parameters screen by selecting the confirm icon.



Step 4
Set the background, “On” or “Off”.

If Christian Warburg”, “BSA”, “IgG”, or “Lysozyme” mode is selected, skip step 5 and go straight to step 6.



Step 5
For the "Molar Extinction" mode, define the molar extinction coefficient ("AU l/mol ×1000"), then the molecular weight ("MW kDa") of the protein of interest.

OR

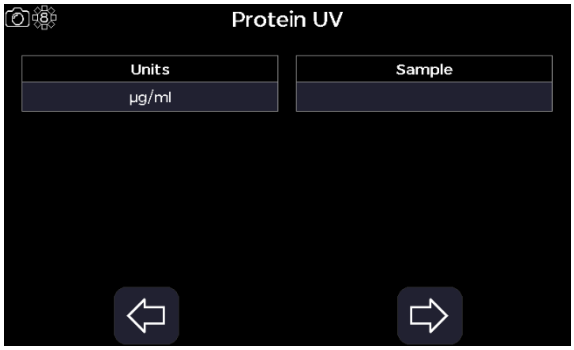
For the "Mass Extinction" mode, define the mass extinction coefficient ("AU l/g") of the protein of interest.

OR

For the "E 1%" mode, define the 1% w/v extinction coefficient ("E 1%") of the protein of interest.

OR

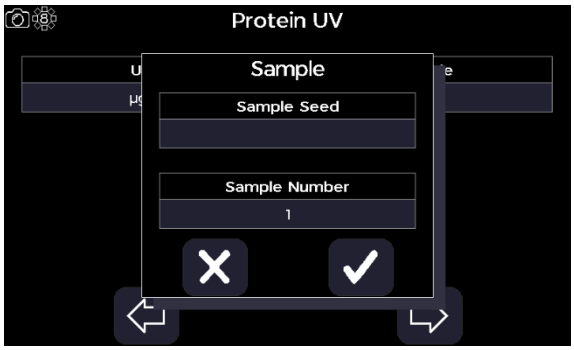
For the "Custom" mode, define the factors to apply to the absorbance measurements at 260 and 280 nm.



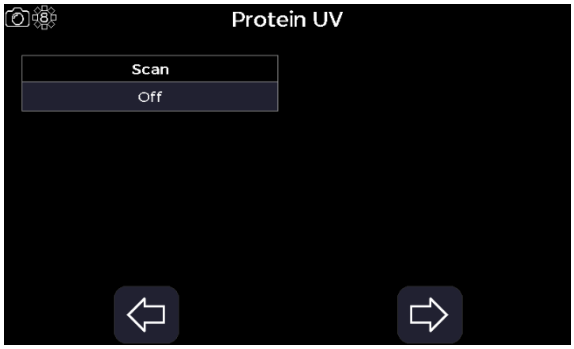
Step 6
Proceed to the next parameter screen using the right/forward arrow.

Step 7
Select one of the predefined units, "µg/ml", "ng/µl", "µg/µl", or "mg/ml".

Step 8
Select the integration time from "1 second", "2 seconds", or "5 seconds".

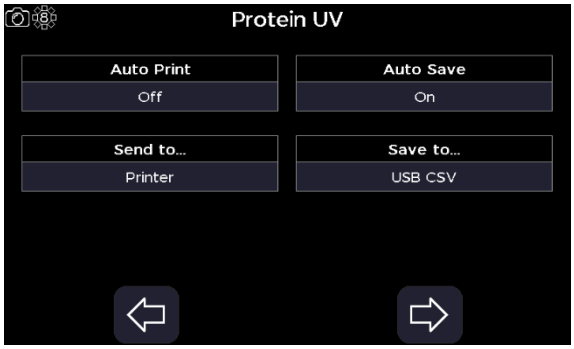


Step 9
Select the sample name to bring up the sample window, enter a sample seed prefix and the incremental sample number starting value. Confirm the settings using the confirm icon.



Step 10
Proceed to the next parameter screen using the right/forward arrow.

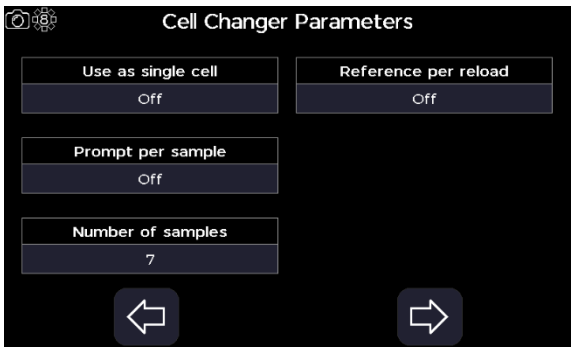
Step 11
Set display scan to "On" or "Off".



Step 12
Proceed to the next parameter screen using the right/forward arrow.

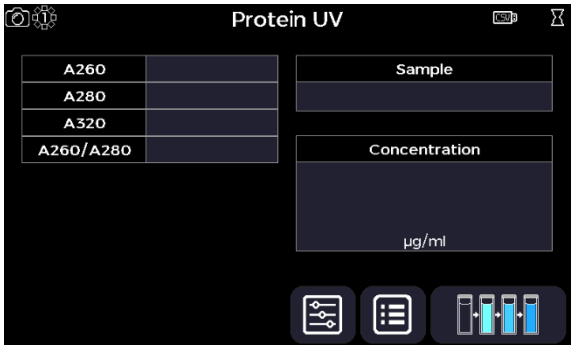
Step 13
Set auto print to “On” or “Off”. If auto print is set to “On”, select the print to hardware from “Internal Printer”, “PC via USB”, or “USB Mass Storage” depending on what hardware is connected to the instrument.

Step 14
Set auto save to “On” or “Off”. If auto save is set to “On”, select the save to hardware from “USB CSV”, “USB”, or “Internal” depending on what hardware is connected to the instrument.

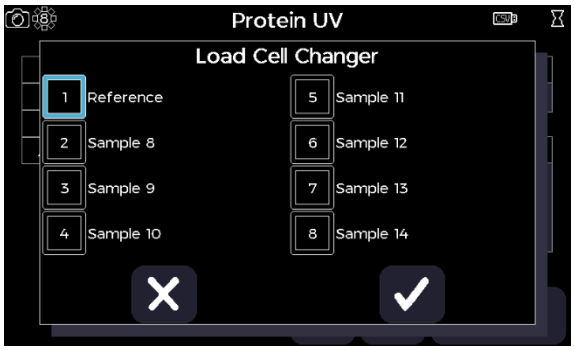


Step 15
Proceed to the next parameter screen using the right/forward arrow.

Step 16
Set whether to use as single cell to “On” or “Off”. If set to “Off”, set the position prompt per sample to “On” or “Off”, set the number of samples to between 2 and 100, and set whether to retake the reference between reload to “On” of “Off”.

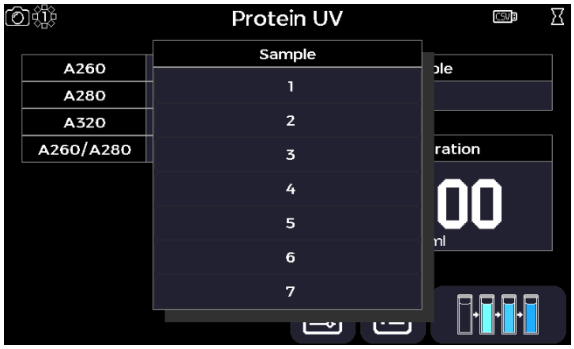


Step 17
Proceed to the measurement screen using the right/forward arrow.

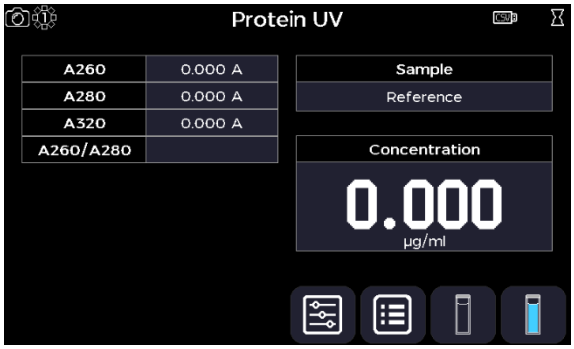


If using a single cell holder, or a cell changer set to use as a single cell holder, skip steps 18 through 19 and go straight to step 20.

Step 18
Press the batch measurement icon, then load the cell changer according to the cell changer prompt.



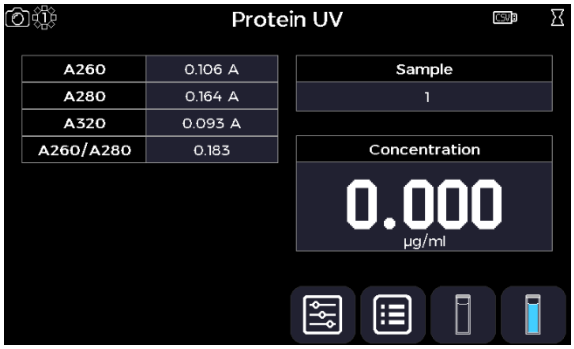
Step 19
The acquired reference sample baseline will be applied to all subsequent sample measurements. The sample measurements can be viewed by pressing the sample name test box and selecting the appropriate sample from the list.



If using a cell changer, skip steps 20 through 21 and go straight to step 22.

Step 20
Insert the reference sample then take a reference measurement using the reference measurement icon.

The acquired reference sample baseline will be applied to all subsequent sample measurements.



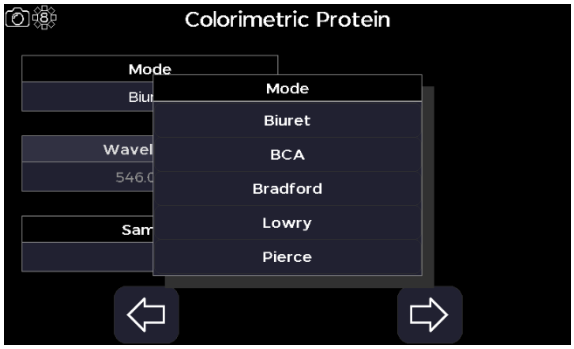
Step 21
Replace the previous sample with a test sample then take a sample measurement using the sample measurement icon.

Repeat for all samples.

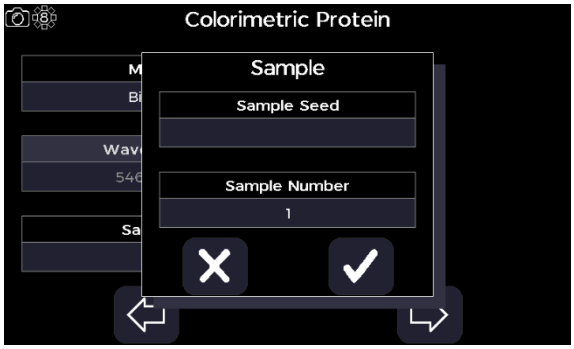
Step 22
Return to the Protein screen using the exit icon in the options menu **OR** use the additional options to save, print, and, or load previous measurements (see the Additional Options section).

Colorimetric Protein

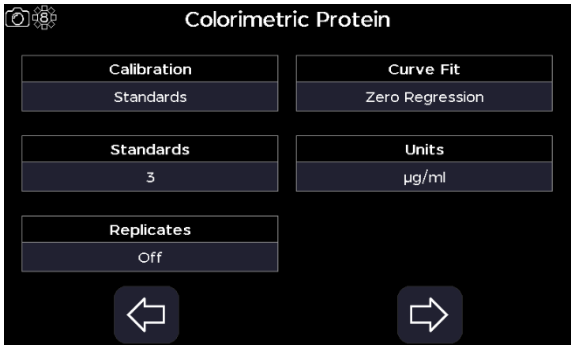
The Colorimetric Protein application is accessed from the Protein screen. It can be used to perform Biuret, BCA, Bradford, Lowry, and Pierce protein quantification assays.



Step 1
Select the mode from “Biuret”, “BCA”, “Bradford”, “Lowry”, or “Pierce”.



Step 2
Select the sample name to bring up the sample window, enter a sample seed prefix and the incremental sample number starting value. Confirm the settings using the confirm icon.



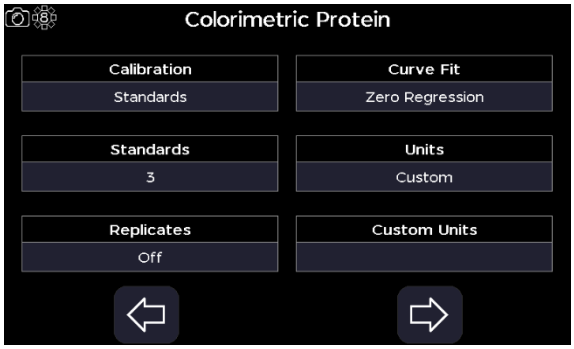
Step 3
Proceed to the next parameter screen using the right/forward arrow.

Step 4
Select the source of the calibration to “Standards” or “Manual”.

Step 5
Select the number of standard samples of known concentration to “1”, “2”, “3”, “4”, “5”, “6”, “7”, “8”, or “9”.

Step 6
If the source of the calibration is set to “Standards”, select the number of standard sample replicates to “Off”, “2”, or “3”.

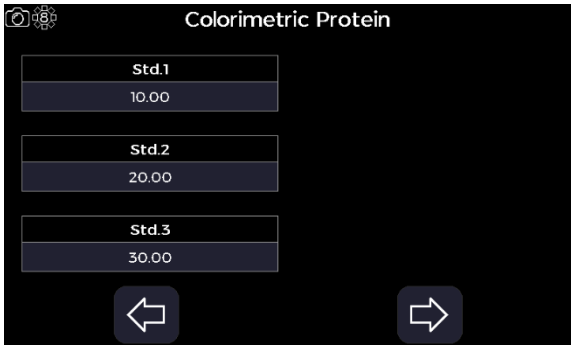
Step 7
Select the curve fit, “Regression”, “Interpolation”, “Cubic Spline”, “Zero Regression”, or “2nd Order Polynomial”.



Step 8
Select one of the predefined units; “µg/ml”, “ng/µl”, or “µg/µl”.

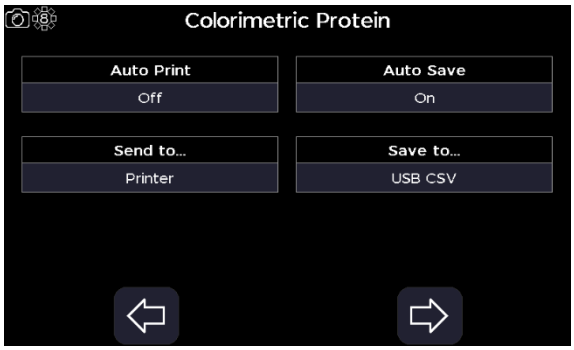
OR

Select “Custom” and define the custom units that the concentration value will be reported in.



Step 9
Proceed to the next parameter screen using the right/forward arrow.

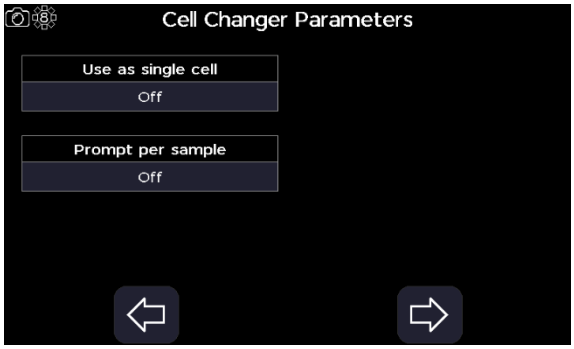
Step 10
Enter the concentration values of the standards samples between -9999 to 9999. Confirm the settings using the confirm icon.



Step 11
Proceed to the next parameter screen using the right/forward arrow.

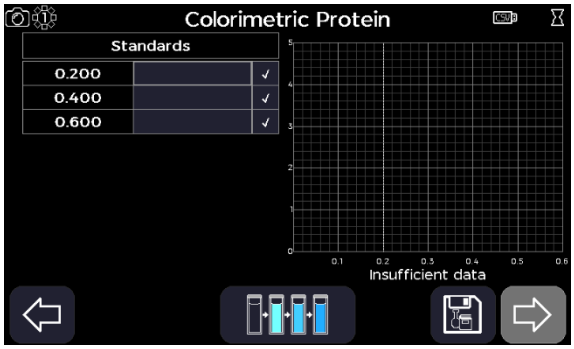
Step 12
Set auto print to “On” or “Off”. If auto print is set to “On”, select the print to hardware from “Internal Printer”, “PC via USB”, or “USB Mass Storage” depending on what hardware is connected to the instrument.

Step 13
Set auto save to “On” or “Off”. If auto save is set to “On”, select the save to hardware from “USB CSV”, “USB”, or “Internal” depending on what hardware is connected to the instrument.



Step 14
Proceed to the next parameter screen using the right/forward arrow.

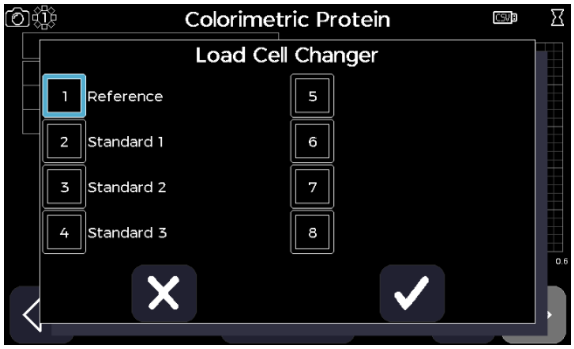
Step 15
Set whether to use as single cell to “On” or “Off”. If set to “Off”, set the position prompt per sample to “On” or “Off”.



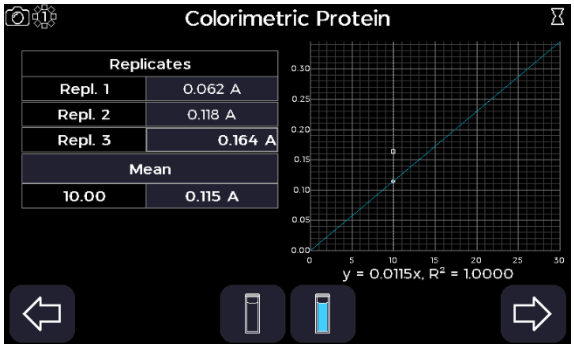
If the source of the calibration is set to “Manual”, skip steps 16 through 23 and go to step 24.

If using a single cell holder, or a cell changer set to use as a single cell holder, skip steps 16 through 17 and go straight to step 18.

Step 16
Proceed to the next parameter screen using the right/forward arrow.

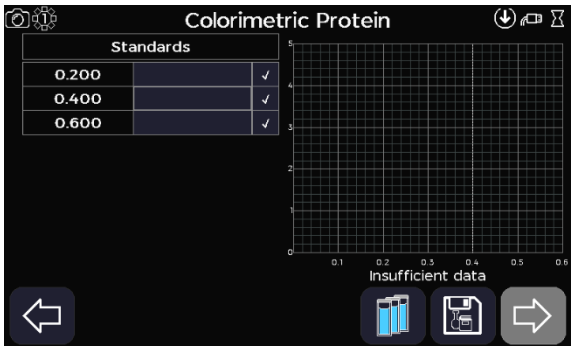


Step 17
Press the batch measurement icon, then load the cell changer according to the cell changer prompt. Then select the confirm icon.



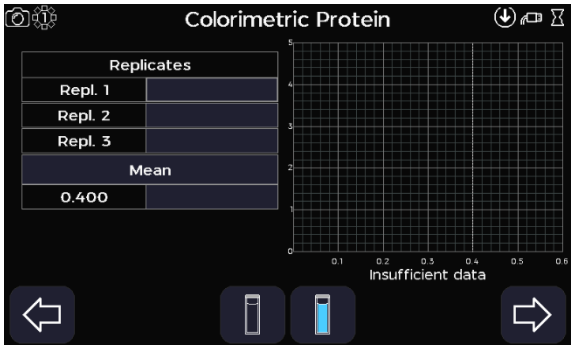
If necessary, any of the standards or replicates can be re-run, by highlighting the appropriate standard and selecting the replicates icon.

Cycle through the standards using the right/forward arrow, and leave the replicates function by cycling back through the standards using the left/backward arrow.



If using a cell changer, skip steps 18 through 23 and go straight to step 24.

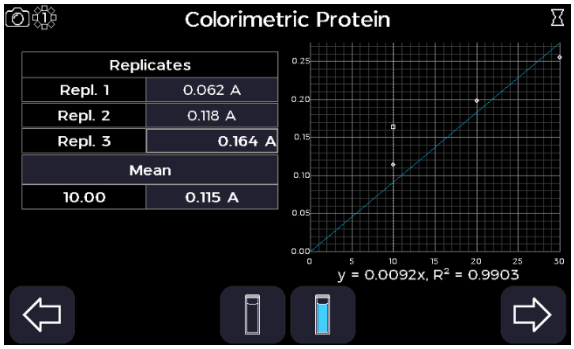
Step 18
Proceed to the next parameter screen using the right/forward arrow.



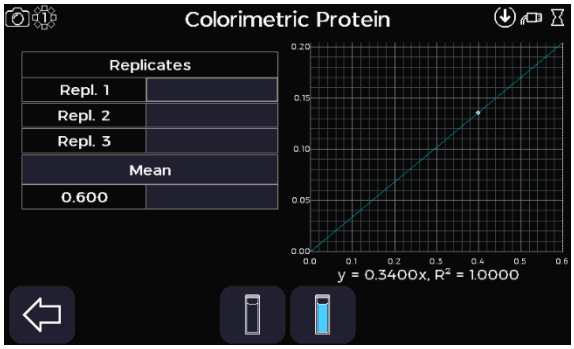
Step 19
Run the standards by selecting the replicates icon.

Insert the reference sample then take a reference measurement using the reference measurement icon.

The acquired reference sample baseline will be applied to all subsequent standard sample measurements.



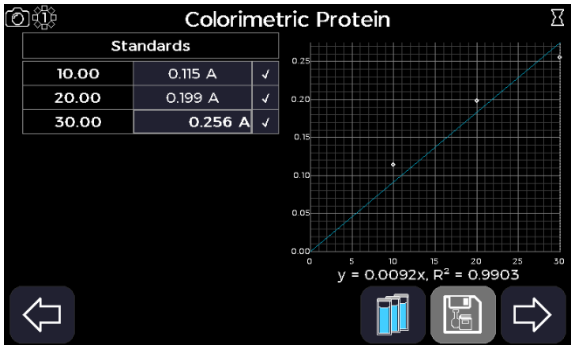
Step 20
Replace the reference sample with the first standard sample then take a sample measurement using the sample measurement icon for each replicate of that standard sample.



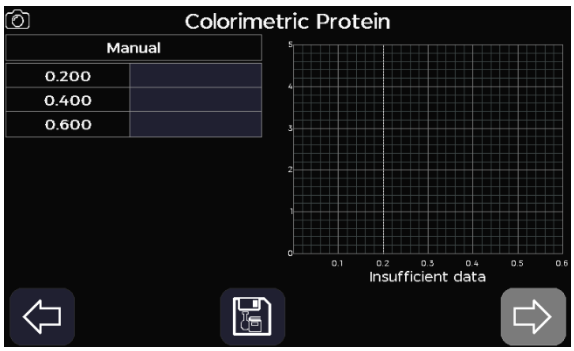
Step 21
Proceed to the next standard measurement screen using the right/forward arrow.

Step 22
Replace the previous standard sample with the next standard sample then take a sample measurement using the sample measurement icon for each replicate of that standard sample.

Repeat for all remaining standard samples if applicable.

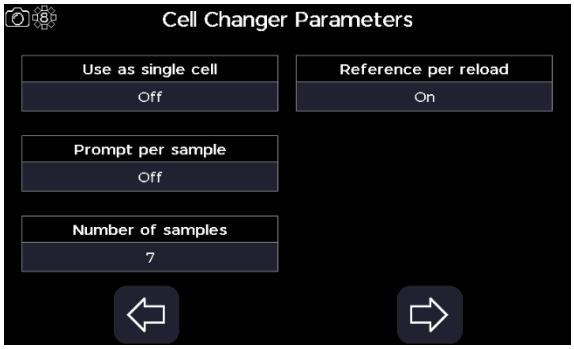


Step 23
Leave the replicates function using the left/backward arrow.



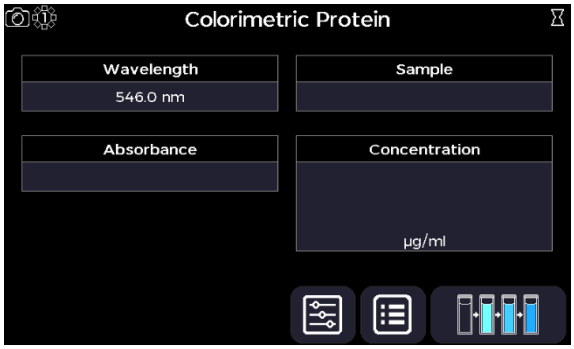
Step 24
If the source of the calibration is set to “Standards”, skip step 24 and go to step 25.

If source of the calibration is set to “Manual”. Define each standards absorbance value by selecting the appropriate text box and entering a value between -0.3 and 3.0 A.

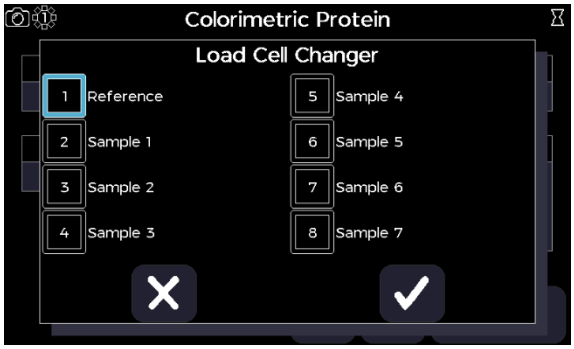


Step 25
Proceed to the next parameter screen using the right/forward arrow.

Step 26
Set whether to use as single cell to “On” or “Off”. If set to “Off”, set the position prompt per sample to “On” or “Off”, set the number of samples to between 2 and 100, and set whether to retake the reference between reload to “On” of “Off”.

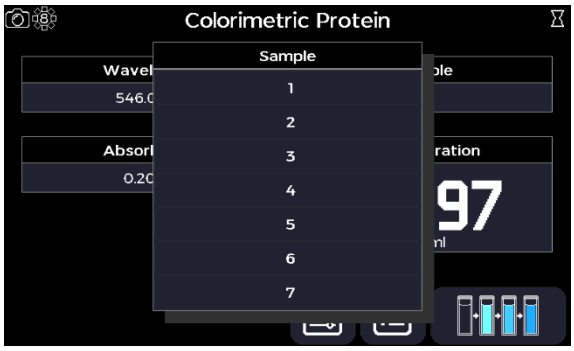


Step 27
Proceed to the measurement screen using the right/forward arrow.

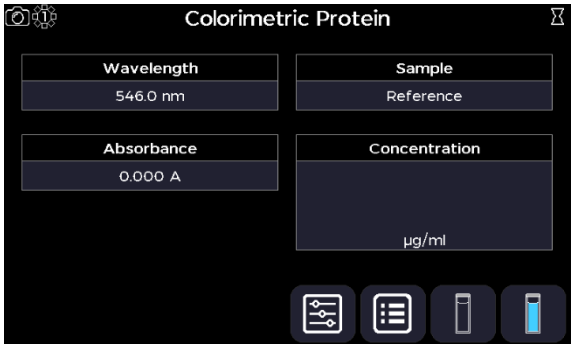


If using a single cell holder, or a cell changer set to use as a single cell holder, skip steps 28 through 29 and go straight to step 30.

Step 28
Press the batch measurement icon, then load the cell changer according to the cell changer prompt. Then select the confirm icon.



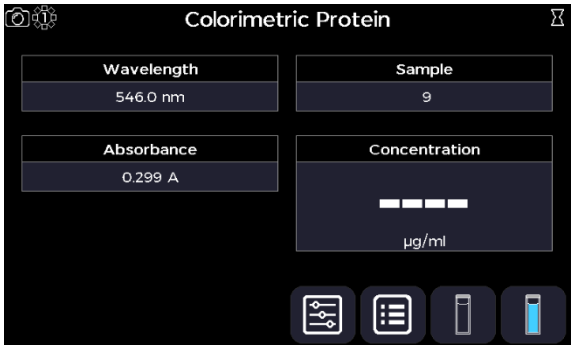
Step 29
The acquired reference sample baseline will have be applied to all subsequent sample measurements. The sample measurements can be viewed by pressing the sample name test box and selecting the appropriate sample from the list.



If using a cell changer, skip steps 30 through 31 and go straight to step 32.

Step 30
Insert the reference sample then take a reference measurement using the reference measurement icon.

The acquired reference sample baseline will be applied to all subsequent sample measurements.



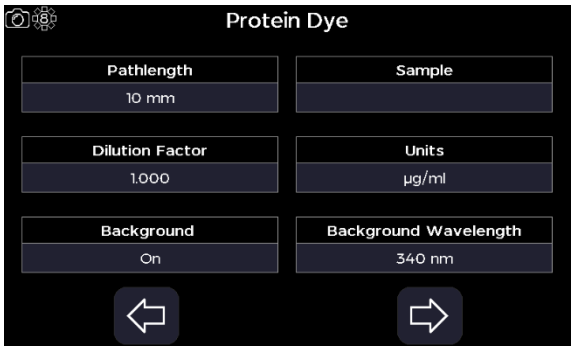
Step 31
Replace the previous sample with a test sample then take a sample measurement using the confirm key.

Repeat for all samples.

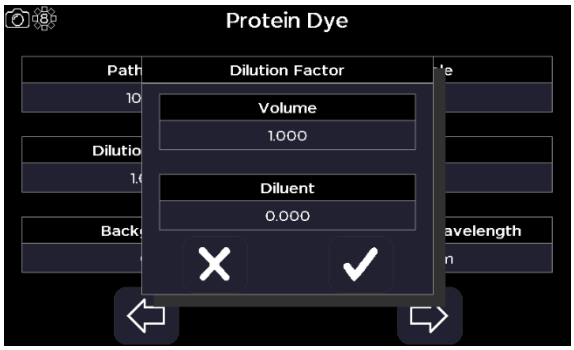
Step 32
Return to the Protein screen using the exit icon in the options menu **OR** use the additional options to save, print, and, or load previous measurements (see the Additional Options section).

Protein Dye

The Protein Dye application is selected from the Protein screen. It can be used to assess the fluorescent labelling efficiency of protein probes, based on the absorbance, prior to their use in microarrays. All calculations applied within the Protein Dye application are described in the Useful Calculations section.



Step 1
Select the pathlength; “10 mm”, “Quantimate 500”, or “Quantimate 200”.

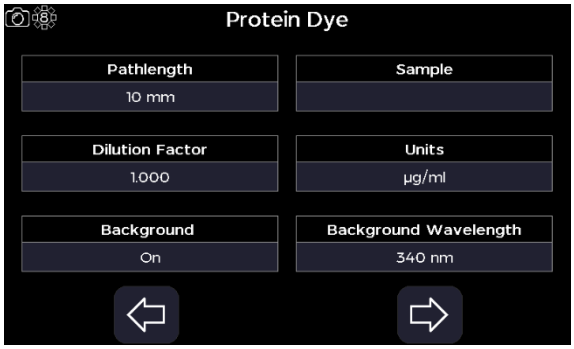


Step 2
Enter any dilution factor to be applied to the absorbance measurement.

Set the initial sample volume of a value of up to four significant figures.

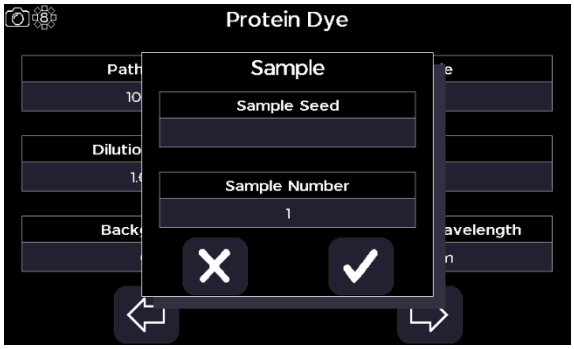
Then set the amount of diluent added to the initial volume of a value of up to four significant figures.

Implement the changes and return to the parameters screen by selecting the confirm icon.

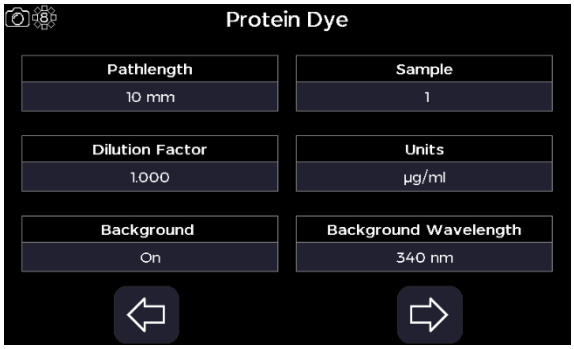


Step 3
Set the background, “On” or “Off”.

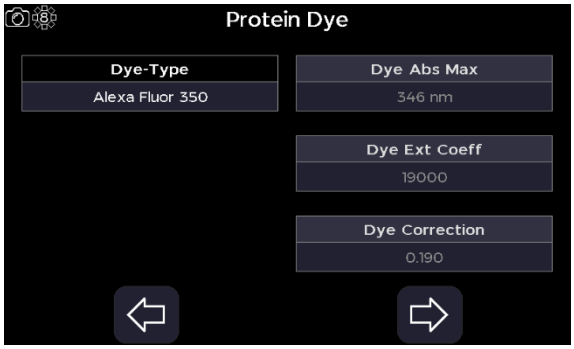
For background set to “On”, set the background wavelength to between 190 and 1100.



Step 4
Select the sample name to bring up the sample window, enter a sample seed prefix and the incremental sample number starting value. Confirm the settings using the confirm icon.



Step 5
Select one of the predefined units, “µg/ml”, “ng/µl”, “µg/µl”, or “mg/ml”.

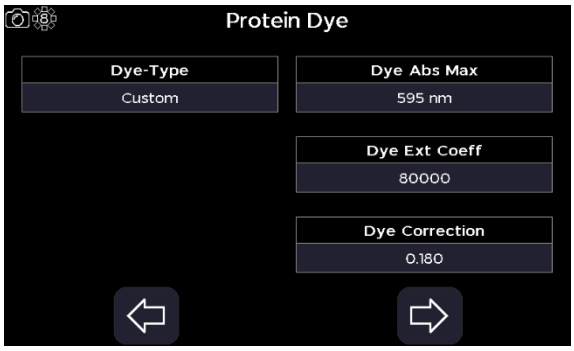


Step 6
Proceed to the next parameter screen using the right/forward arrow.

Step 7
Select the dye type from one of the predefined dyes.

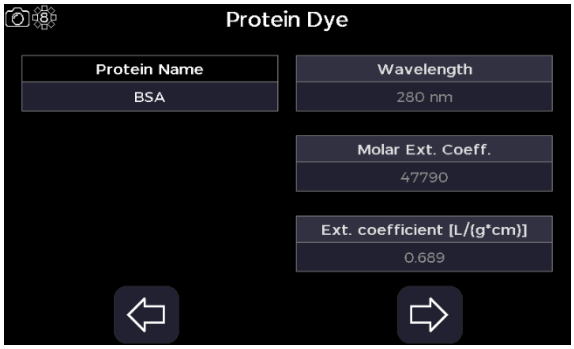
Each dye has fixed associated parameters:

Dye-Type	Dye Abs Max	Dye Ext Coeff	Dye Correction
Alexa Fluor 350	346 nm	19000	0.190
Alexa Fluor 405	401 nm	34000	0.700
Alexa Fluor 488	495 nm	71000	0.110
Alexa Fluor 647	650 nm	239000	0.030
Cy3	550 nm	150000	0.080
Cy5	649 nm	250000	0.050
DyLight 649	654 nm	250000	0.040
DyLight 488	493 nm	70000	0.150
FITC	495 nm	68000	0.300
Pacific Blue	416 nm	46000	0.200
r-PE	566 nm	200000	0.180
Texas Red	595 nm	80000	0.180



OR

Select the custom dye and define the dye absorbance max to between 300 and 950 nm, the dye extinction coefficient to between 10000 and 9999999, and the dye correction factor to between 0.001 and 0.999.

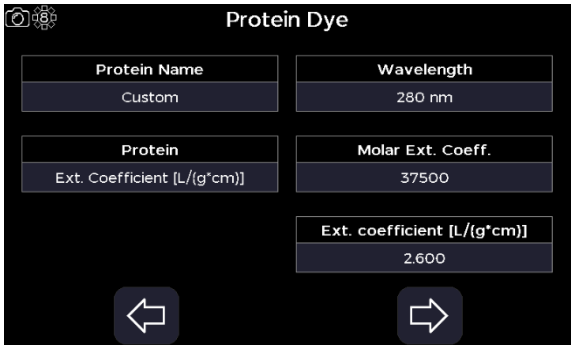


Step 8
Proceed to the next parameter screen using the right/forward arrow.

Step 9
Select the sample protein from one of the predefined protein names.

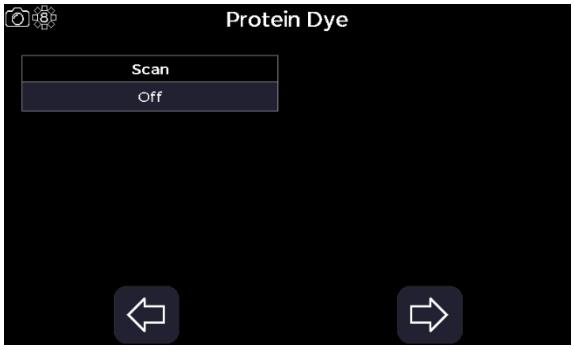
Each protein name has fixed associated parameters:

Protein Name	Wavelength	Molar Ext. Coeff.	Ext. coefficient [L/(g*cm)]
BSA	280 nm	47790	0.689
SA (mouse)	280 nm	43780	0.637
SA (human)	280 nm	39310	0.567
IgG	280 nm	210000	1.400
Lysozyme	280 nm	37500	2.600



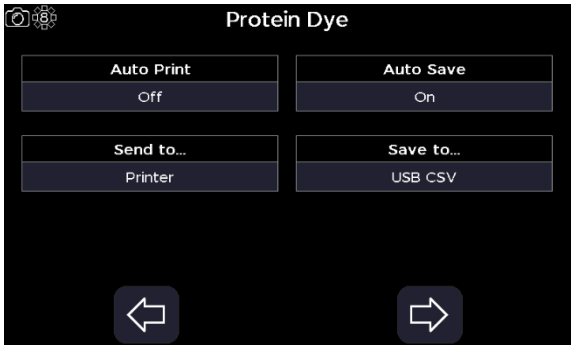
OR

Select the custom protein name, then set the protein quantification mode to “Ext. Coefficient [L/(g*cm)]” or “Molar Ext. Coeff.” and define the wavelength to between 200 and 340 nm, the molar extinction coefficient to between 10000 and 9999999, and the mass extinction coefficient, or molecular weight depending on the quantification mode, to between 0.001 and 9999999.



Step 10
Proceed to the next parameter screen using the right/forward arrow.

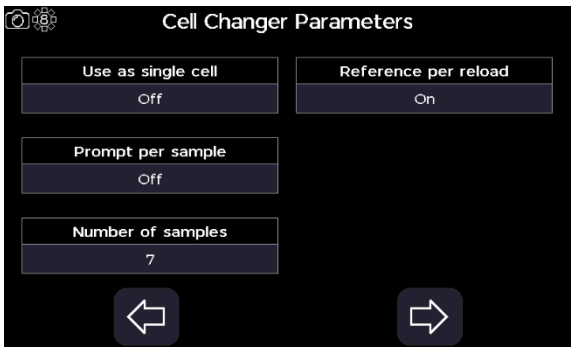
Step 11
Set display scan to “On” or “Off”.



Step 12
Proceed to the next parameter screen using the right/forward arrow.

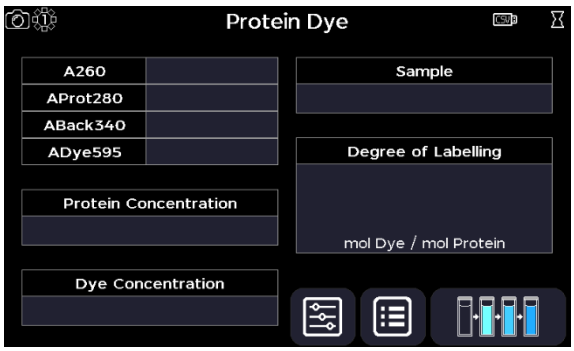
Step 13
Set auto print to “On” or “Off”. If auto print is set to “On”, select the print to hardware from “Internal Printer”, “PC via USB”, or “USB Mass Storage” depending on what hardware is connected to the instrument.

Step 14
Set auto save to “On” or “Off”. If auto save is set to “On”, select the save to hardware from “USB CSV”, “USB”, or “Internal” depending on what hardware is connected to the instrument.

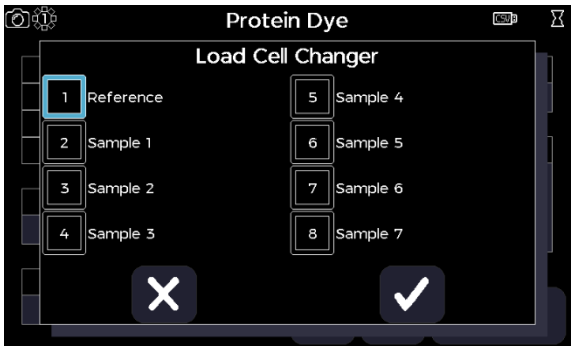


Step 15
Proceed to the next parameter screen using the right/forward arrow.

Step 16
Set whether to use as single cell to “On” or “Off”. If set to “Off”, set the position prompt per sample to “On” or “Off”, set the number of samples to between 2 and 100, and set whether to retake the reference between reload to “On” of “Off”.

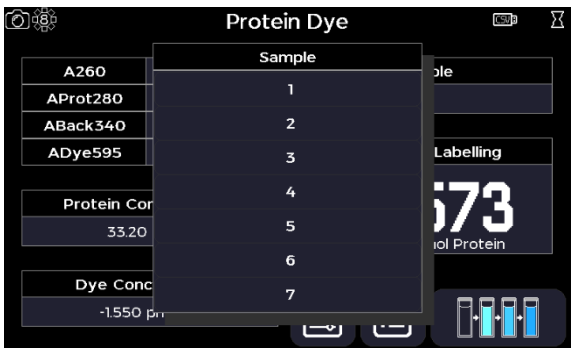


Step 17
Proceed to the measurement screen using the right/forward arrow.

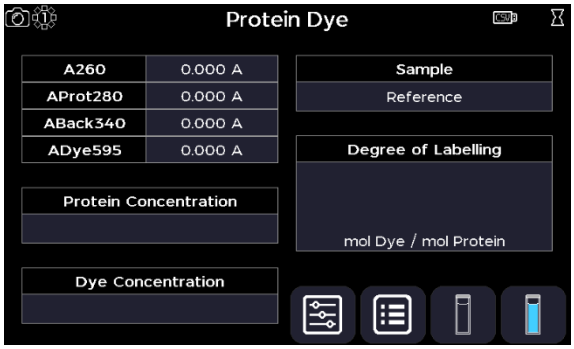


If using a single cell holder, or a cell changer set to use as a single cell holder, skip steps 18 through 19 and go straight to step 20.

Step 18
Press the batch measurement icon, then load the cell changer according to the cell changer prompt. Then select the confirm icon.



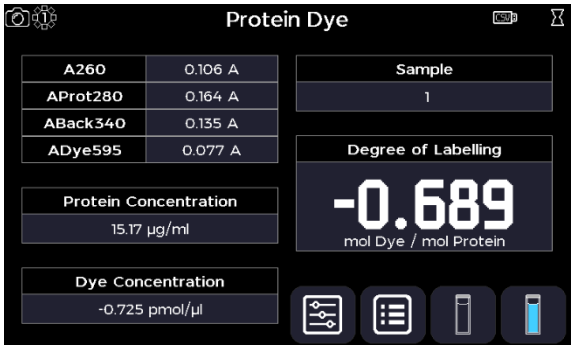
Step 19
The acquired reference sample baseline will have be applied to all subsequent sample measurements. The sample measurements can be viewed by pressing the sample name test box and selecting the appropriate sample from the list.



If using a cell changer, skip steps 20 through 21 and go straight to step 22.

Step 20
Insert the reference sample then take a reference measurement using the reference measurement icon.

The acquired reference sample baseline will be applied to all subsequent sample measurements.



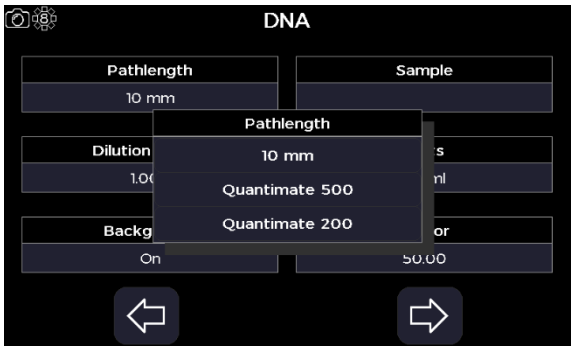
Step 21
Replace the previous sample with a test sample then take a sample measurement using the confirm key.

Repeat for all samples.

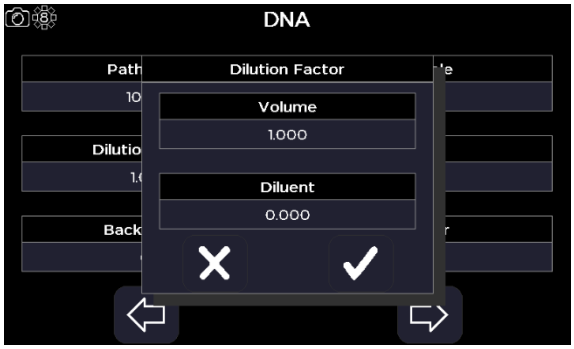
Step 21
Return to the Protein screen using the exit icon in the options menu **OR** use the additional options to save, print, and, or load previous measurements (see the Additional Options section).

DNA

The DNA application is accessed from the home screen. It can be used to perform DNA quantification measurements at 260 nm and to assess sample quality using A260/A280 and A260/A230 absorbance (A) ratios. All calculations applied within the DNA application are described in the Useful Calculations section.



Step 1
Set the pathlength; “10 mm”, “Quantimate 500”, or, “Quantimate 200”.

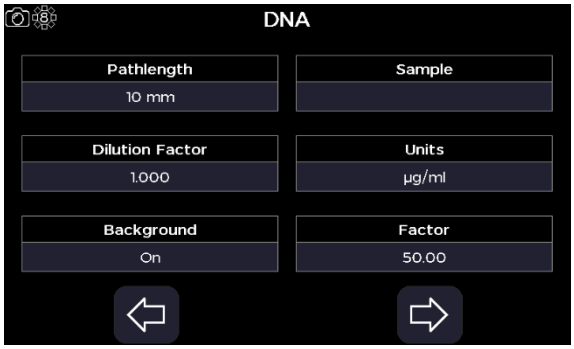


Step 2
Enter any dilution factor to be applied to the absorbance measurement.

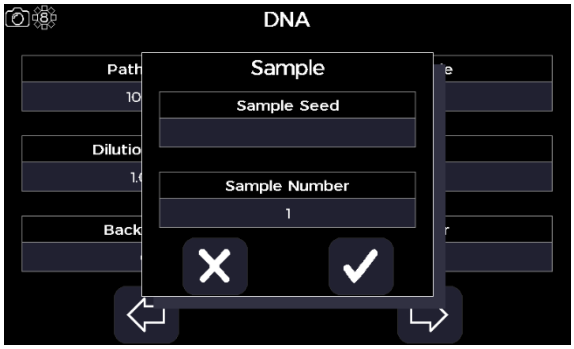
Set the initial sample volume of a value of up to four significant figures.

Then set the amount of diluent added to the initial volume of a value of up to four significant figures.

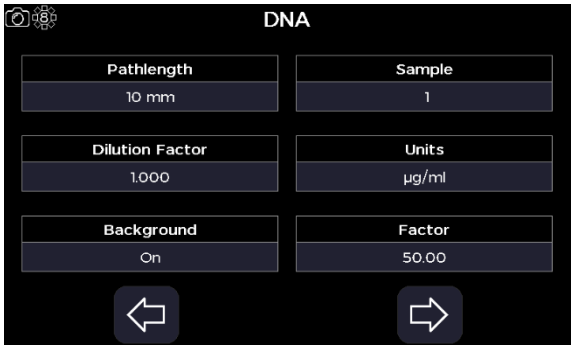
Implement the changes and return to the parameters screen by selecting the confirm icon.



Step 3
Set the background, “On” or “Off”.



Step 4
Select the sample name to bring up the sample window, enter a sample seed prefix and the incremental sample number starting value. Confirm the settings using the confirm icon.

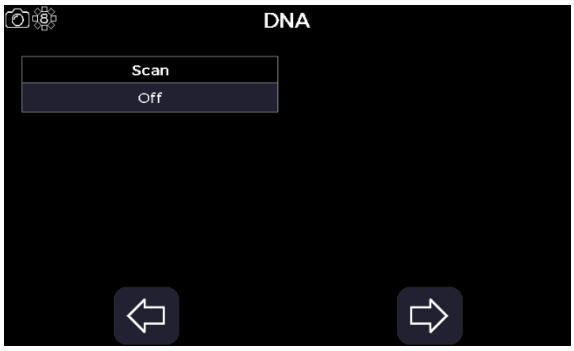


Step 5
Select one of the predefined units, “µg/ml”, “ng/µl”, or “µg/µl”.

For each predefined unit a default factor is applied, “50.00” for “µg/ml” and “ng/µl”, or “0.050” for “µg/µl”.

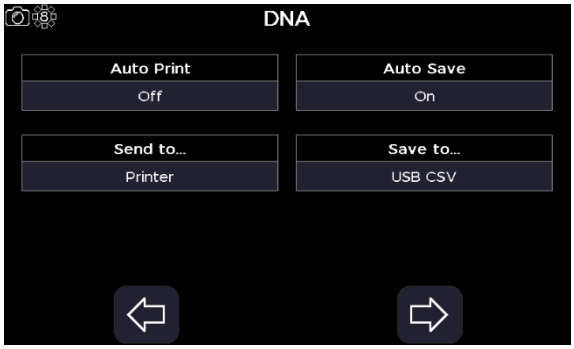
OR

Enter a custom value of up to four significant figures.



Step 6
Proceed to the next parameter screen using the right/forward arrow.

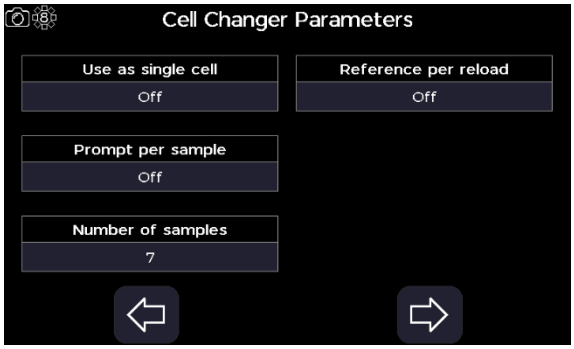
Step 7
Set display scan to “On” or “Off”.



Step 8
Proceed to the next parameter screen using the right/forward arrow.

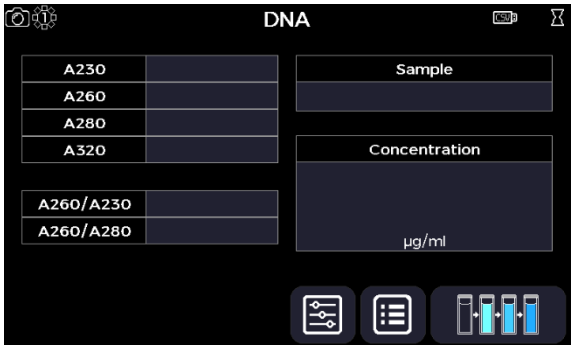
Step 9
Set auto print to “On” or “Off”. If auto print is set to “On”, select the print to hardware from “Internal Printer”, “PC via USB”, or “USB Mass Storage” depending on what hardware is connected to the instrument.

Step 10
Set auto save to “On” or “Off”. If auto save is set to “On”, select the save to hardware from “USB CSV”, “USB”, or “Internal” depending on what hardware is connected to the instrument.

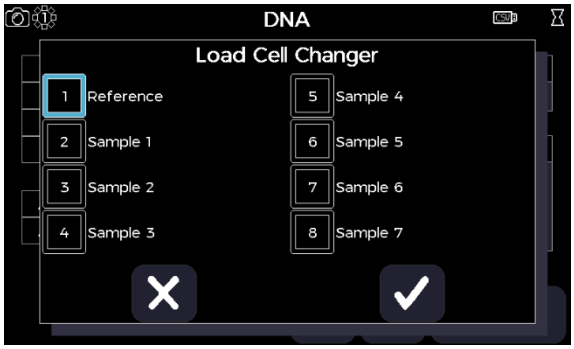


Step 11
Proceed to the next parameter screen using the right/forward arrow.

Step 12
Set whether to use as single cell to “On” or “Off”. If set to “Off”, set the position prompt per sample to “On” or “Off”, set the number of samples to between 2 and 100, and set whether to retake the reference between reload to “On” of “Off”.



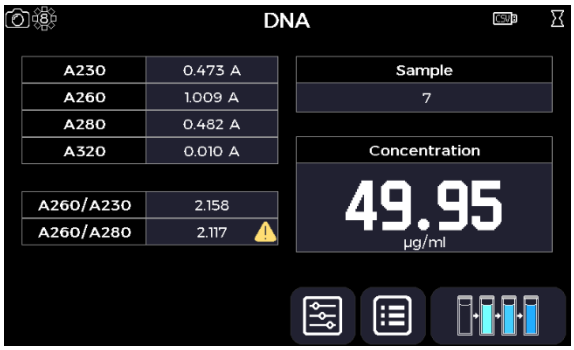
Step 13
Proceed to the measurement screen using the right/forward arrow.



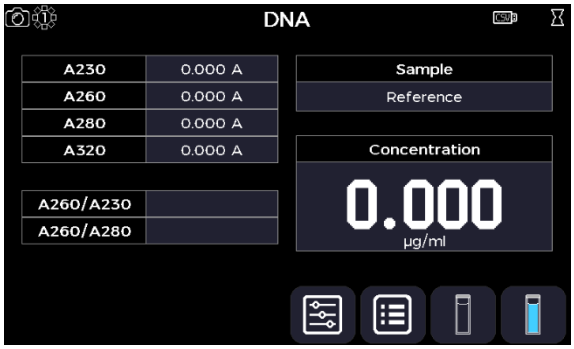
If using a single cell holder, or a cell changer set to use as a single cell holder, skip steps 14 through 15 and go straight to step 16.

Step 14
Press the batch measurement icon, then load the cell changer according to the cell changer prompt. Then select the confirm icon.

Step 15
The acquired reference sample baseline will have be applied to all subsequent sample measurements. The sample measurements can be viewed by pressing the sample name test box and selecting the appropriate sample from the list.



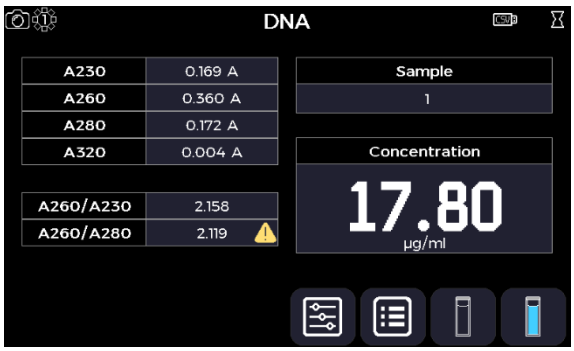
The exclamation icon (⚠) appearing beside the purity ratios, indicates that they are outside the ideal Warburg and Christian range.



If using a cell changer, skip steps 16 through 17 and go straight to step 18.

Step 16
Insert the reference sample then take a reference measurement using the reference measurement icon.

The acquired reference sample baseline will be applied to all subsequent sample measurements.



Step 17
Replace the previous sample with a test sample then take a sample measurement using the confirm key.

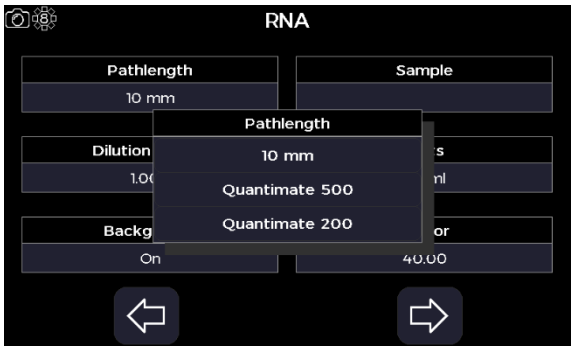
The exclamation icon (⚠) appearing beside the purity ratios, indicates that they are outside the ideal Warburg and Christian range.

Repeat for all samples.

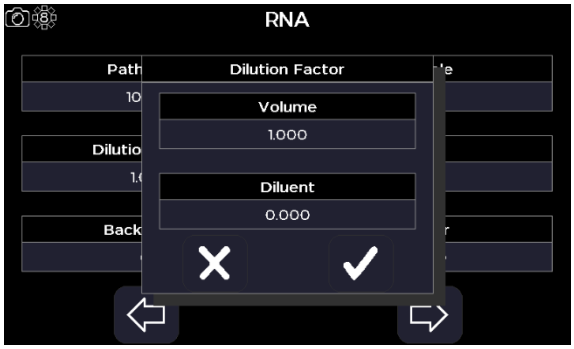
Step 18
Return to the home screen using the exit icon in the options menu **OR** use the additional options to save, print, and, or load previous measurements (see the Additional Options section).

RNA

The RNA application is selected from the home screen. It can be used to perform RNA quantification measurements at 260 nm and to assess sample quality using A260/A280 and A260/A230 absorbance (A) ratios. All calculation applied within the RNA application are described in the Useful Calculation section.



Step 1
Set the pathlength; “10 mm”, “Quantimate 500”, or, “Quantimate 200”.

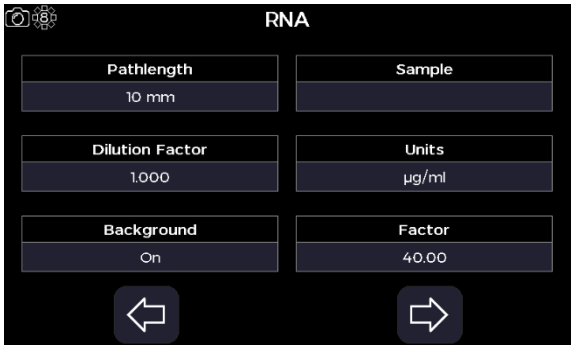


Step 2
Enter any dilution factor to be applied to the absorbance measurement.

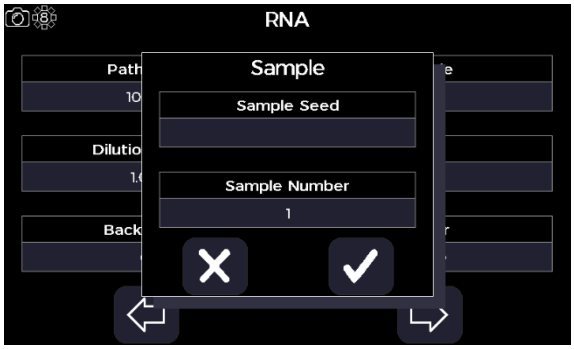
Set the initial sample volume of a value of up to four significant figures.

Then set the amount of diluent added to the initial volume of a value of up to four significant figures.

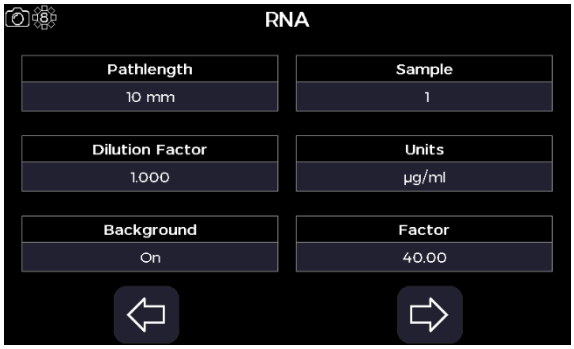
Implement the changes and return to the parameters screen by selecting the confirm icon.



Step 3
Set the background, “On” or “Off”.



Step 4
Select the sample name to bring up the sample window, enter a sample seed prefix and the incremental sample number starting value. Confirm the settings using the confirm icon.

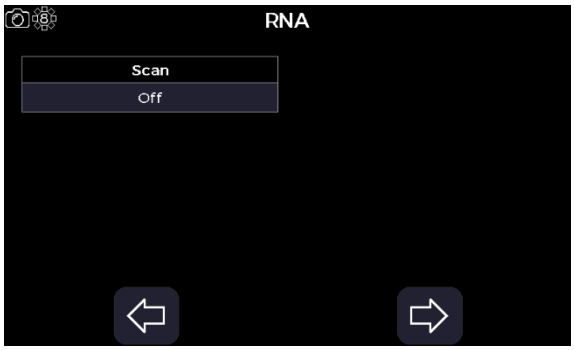


Step 5
Select one of the predefined units, “µg/ml”, “ng/µl”, or “µg/µl”.

For each predefined unit a default factor is applied, “50.00” for “µg/ml” and “ng/µl”, or “0.050” for “µg/µl”.

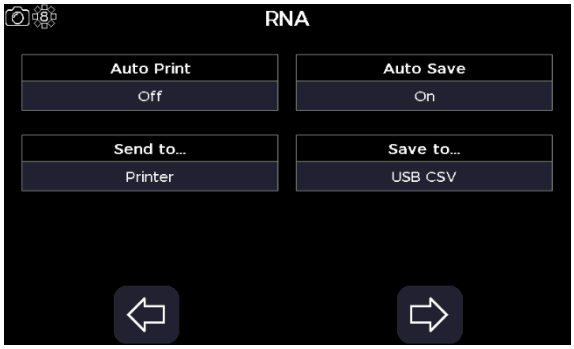
OR

Enter a custom value of up to four significant figures.



Step 6
Proceed to the next parameter screen using the right/forward arrow.

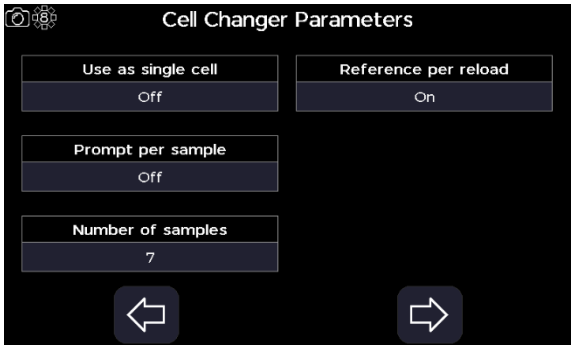
Step 7
Set display scan to “On” or “Off”.



Step 8
Proceed to the next parameter screen using the right/forward arrow.

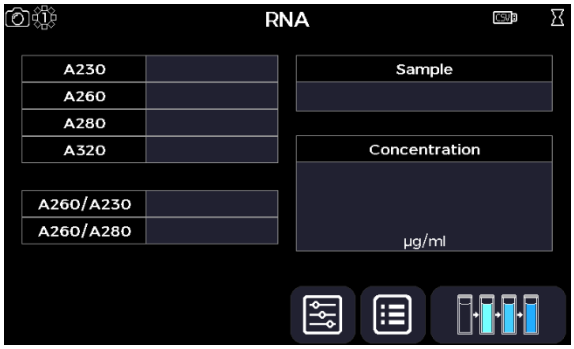
Step 9
Set auto print to “On” or “Off”. If auto print is set to “On”, select the print to hardware from “Internal Printer”, “PC via USB”, or “USB Mass Storage” depending on what hardware is connected to the instrument.

Step 10
Set auto save to “On” or “Off”. If auto save is set to “On”, select the save to hardware from “USB CSV”, “USB”, or “Internal” depending on what hardware is connected to the instrument.

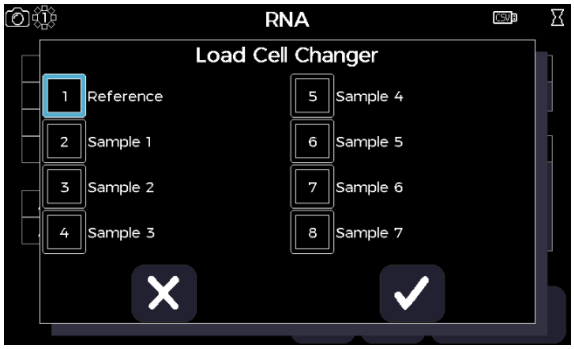


Step 11
Proceed to the next parameter screen using the right/forward arrow.

Step 12
Set whether to use as single cell to “On” or “Off”. If set to “Off”, set the position prompt per sample to “On” or “Off”, set the number of samples to between 2 and 100, and set whether to retake the reference between reload to “On” of “Off”.



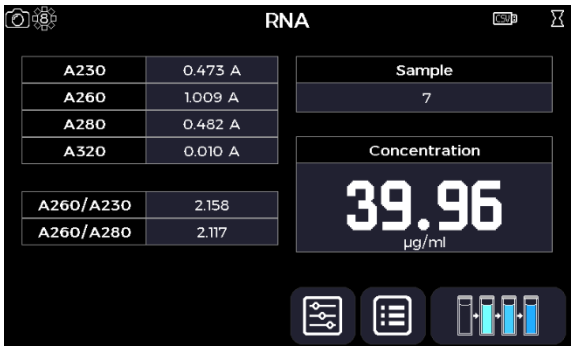
Step 13
Proceed to the measurement screen using the right/forward arrow.



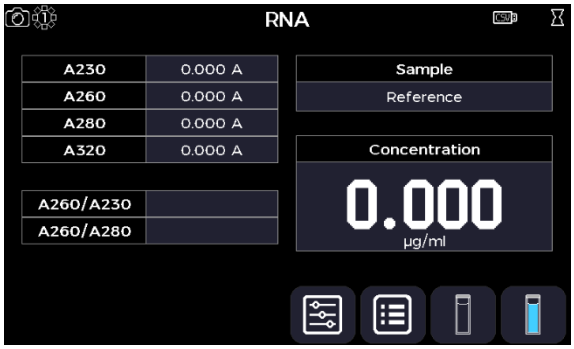
If using a single cell holder, or a cell changer set to use as a single cell holder, skip steps 14 through 15 and go straight to step 16.

Step 14
Press the batch measurement icon, then load the cell changer according to the cell changer prompt. Then select the confirm icon.

Step 15
The acquired reference sample baseline will have be applied to all subsequent sample measurements. The sample measurements can be viewed by pressing the sample name test box and selecting the appropriate sample from the list.



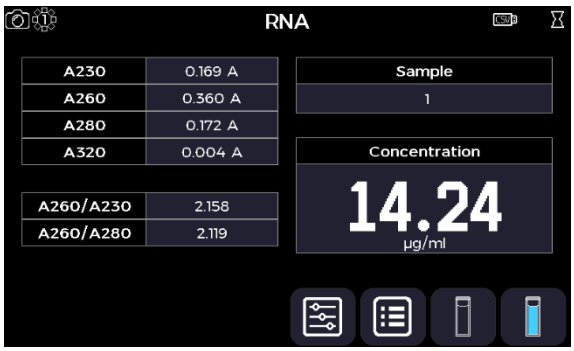
The exclamation icon (⚠) appearing beside the purity ratios, indicates that they are outside the ideal Warburg and Christian range.



If using a cell changer, skip steps 16 through 17 and go straight to step 18.

Step 16
Insert the reference sample then take a reference measurement using the reference measurement icon.

The acquired reference sample baseline will be applied to all subsequent sample measurements.



Step 17
Replace the previous sample with a test sample then take a sample measurement using the confirm key.

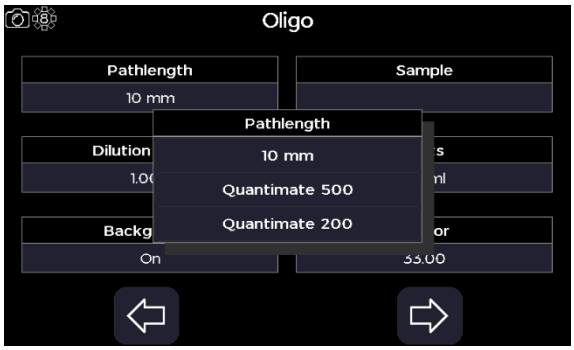
The exclamation icon (⚠) appearing beside the purity ratios, indicates that they are outside the ideal Warburg and Christian range.

Repeat for all samples.

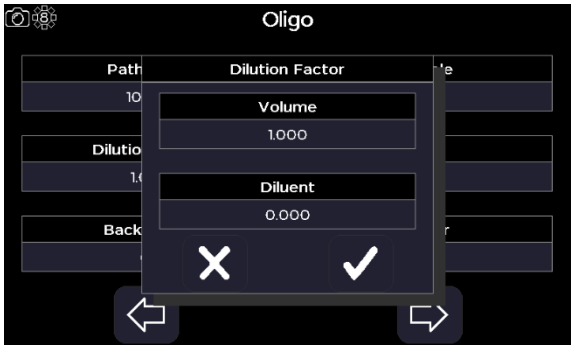
Step 18
Return to the home screen using the exit icon in the options menu **OR** use the additional options to save, print, and, or load previous measurements (see the Additional Options section).

Oligo

The Oligo application is selected from the home screen. It can be used to perform Oligonucleotide quantification measurements at 260 nm and to assess sample quality using A260/A280 and A260/A230 absorbance (A) ratios. All calculations applied within the Oligo application are described in the Useful Calculations section.



Step 1
Set the pathlength; “10 mm”, “Quantimate 500”, or, “Quantimate 200”.

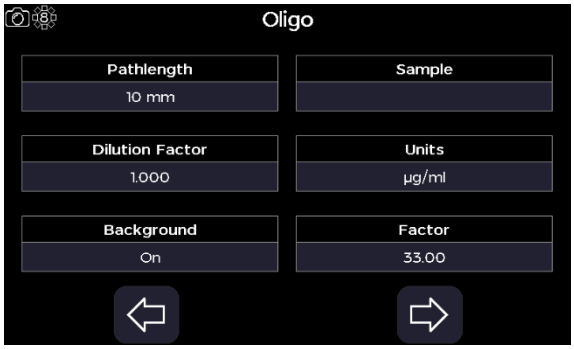


Step 2
Enter any dilution factor to be applied to the absorbance measurement.

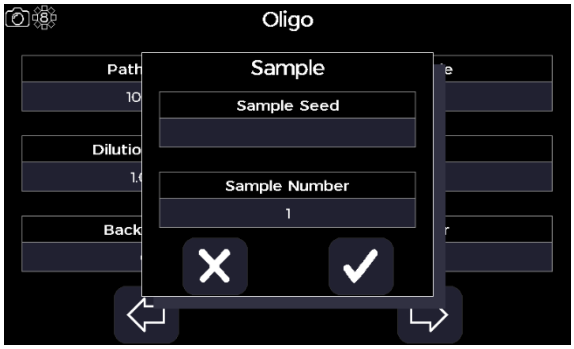
Set the initial sample volume of a value of up to four significant figures.

Then set the amount of diluent added to the initial volume of a value of up to four significant figures.

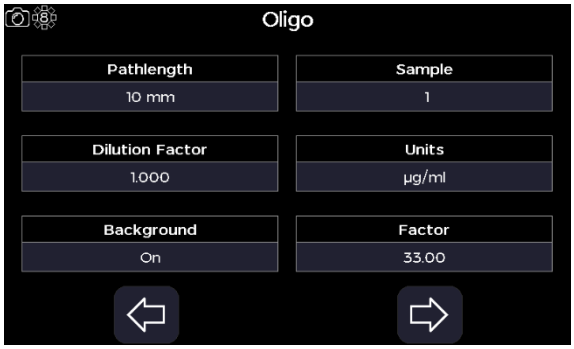
Implement the changes and return to the parameters screen by selecting the confirm icon.



Step 3
Set the background, “On” or “Off”.



Step 4
Select the sample name to bring up the sample window, enter a sample seed prefix and the incremental sample number starting value. Confirm the settings using the confirm icon.



Step 5
Select one of the predefined units, “µg/ml”, “ng/µl”, or “µg/µl”.

For each predefined unit a default factor is applied, “50.00” for “µg/ml” and “ng/µl”, or “0.050” for “µg/µl”.

OR

Enter a custom value of up to four significant figures.

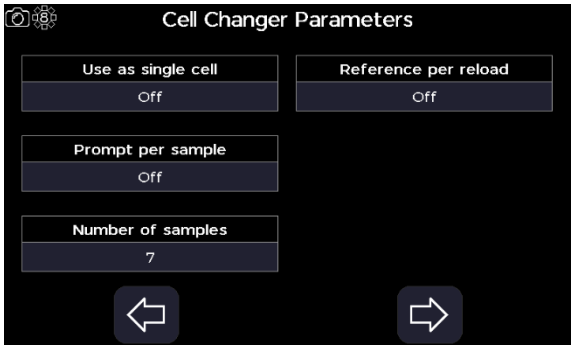
Step 6
Proceed to the next parameter screen using the right/forward arrow.

Step 7
Set display scan to “On” or “Off”.

Step 8
Proceed to the next parameter screen using the right/forward arrow.

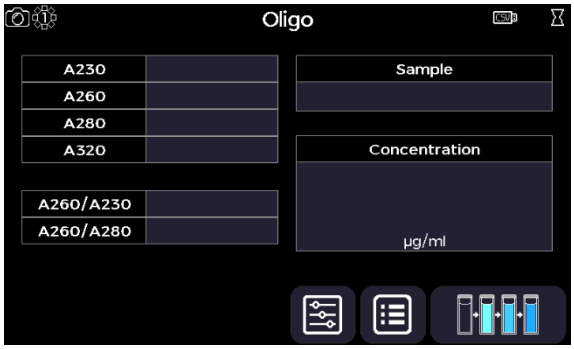
Step 9
Set auto print to “On” or “Off”. If auto print is set to “On”, select the print to hardware from “Internal Printer”, “PC via USB”, or “USB Mass Storage” depending on what hardware is connected to the instrument.

Step 10
Set auto save to “On” or “Off”. If auto save is set to “On”, select the save to hardware from “USB CSV”, “USB”, or “Internal” depending on what hardware is connected to the instrument.

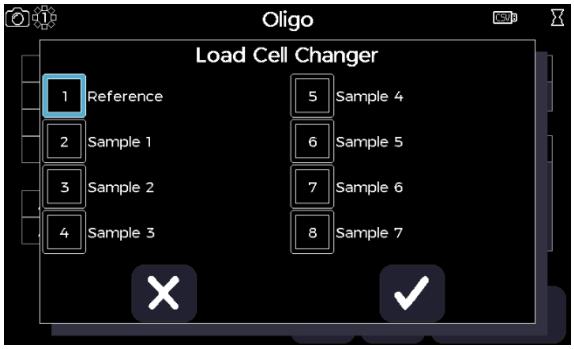


Step 11
Proceed to the next parameter screen using the right/forward arrow.

Step 12
Set whether to use as single cell to “On” or “Off”. If set to “Off”, set the position prompt per sample to “On” or “Off”, set the number of samples to between 2 and 100, and set whether to retake the reference between reload to “On” of “Off”.



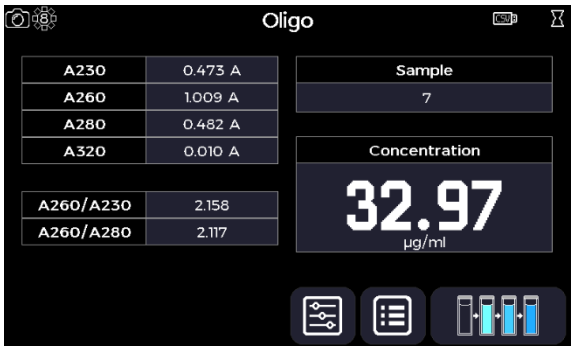
Step 13
Proceed to the measurement screen using the right/forward arrow.



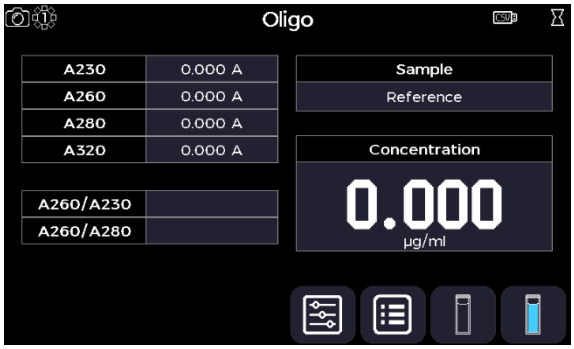
If using a single cell holder, or a cell changer set to use as a single cell holder, skip steps 14 through 15 and go straight to step 16.

Step 14
Press the batch measurement icon, then load the cell changer according to the cell changer prompt. Then select the confirm icon.

Step 15
The acquired reference sample baseline will have be applied to all subsequent sample measurements. The sample measurements can be viewed by pressing the sample name test box and selecting the appropriate sample from the list.



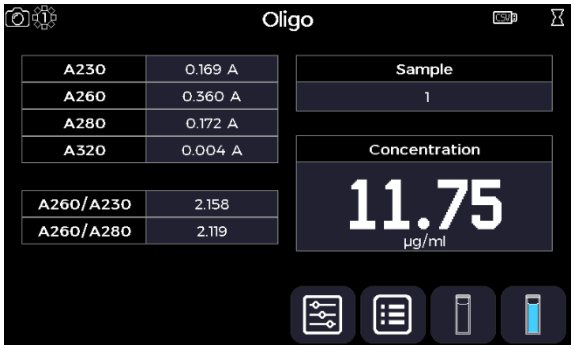
The exclamation icon (⚠) appearing beside the purity ratios, indicates that they are outside the ideal Warburg and Christian range.



If using a cell changer, skip steps 16 through 17 and go straight to step 18.

Step 16
Insert the reference sample then take a reference measurement using the reference measurement icon.

The acquired reference sample baseline will be applied to all subsequent sample measurements.



Step 17
Replace the previous sample with a test sample then take a sample measurement using the confirm key.

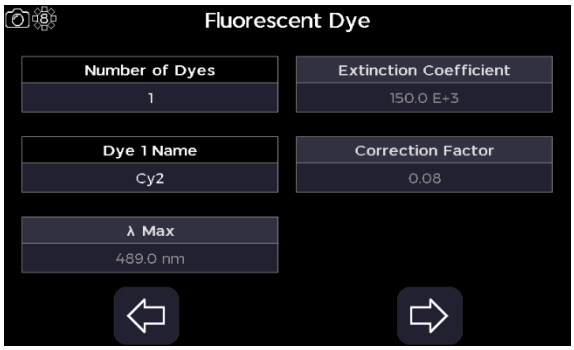
The exclamation icon (⚠) appearing beside the purity ratios, indicates that they are outside the ideal Warburg and Christian range.

Repeat for all samples.

Step 18
Return to the home screen using the exit icon in the options menu **OR** use the additional options to save, print, and, or load previous measurements (see the Additional Options section).

Fluorescent Dye

The Fluorescent Dye application is selected from the home screen. It can be used to assess the fluorescent labelling efficiency of nucleic acid probes, based on the absorbance, prior to their use in microarrays. All calculations applied within the Fluorescent Dye application are described in the Useful Calculations section.

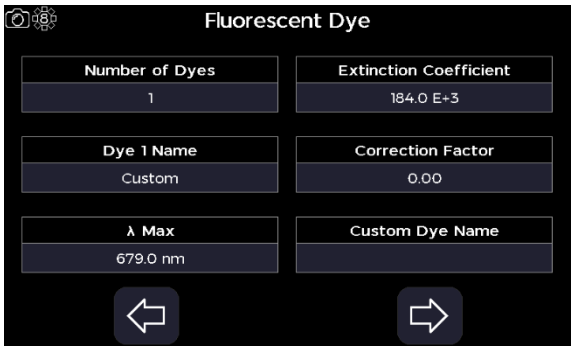


Step 1
Set the number of dyes to “1” or “2”.

Step 2
Select the dye 1 name from one of the predefined dyes.

Each dye has fixed associated parameters:

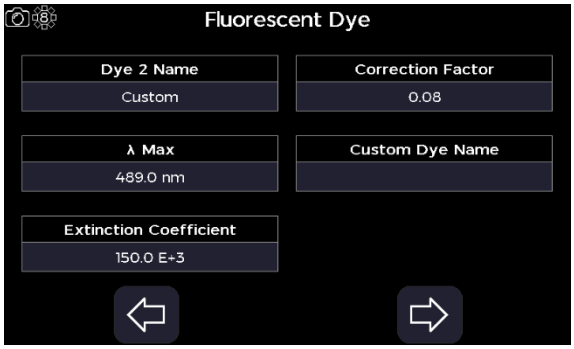
Dye 1 Name	λ Max	Extinction Coefficient	Correction Factor
Cy2	489 nm	150 E+3	0.08
Cy3	550 nm	150 E+3	0.08
Cy3B	558 nm	130 E+3	0.06
Cy3.5	581 nm	150 E+3	0.14
Cy5	649 nm	250 E+3	0.05
Cy5.5	675 nm	190 E+3	0.15
Cy7	747 nm	200 E+3	0.04
Hyper5	660 nm	110 E+3	0.25
Fluorescein	494 nm	92.3 E+3	0.32
Alexa Fluor 350	346 nm	19.0 E+3	0.25
Alexa Fluor 488	495 nm	71.0 E+3	0.30
Alexa Fluor 532	532 nm	81.0 E+3	0.24
Alexa Fluor 546	554 nm	112 E+3	0.21
Alexa Fluor 555	555 nm	150 E+3	0.08
Alexa Fluor 568	578 nm	91.3 E+3	0.45
Alexa Fluor 594	590 nm	90.0 E+3	0.43
Alexa Fluor 647	650 nm	239 E+3	0.00
Alexa Fluor 660	663 nm	132 E+3	0.00
Alexa Fluor 680	679 nm	184 E+3	0.00



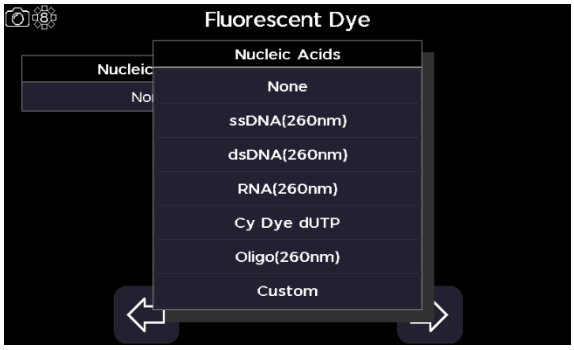
OR

Select the custom dye and define the λ max to between 200 and 999 nm, the Molar extinction coefficient to between 0.001 and 9999 E+3, the A260 correction factor to between 0.01 and 9999, and a15-digit dye name.

Step 3
Proceed to the next parameter screen using the right/forward arrow.

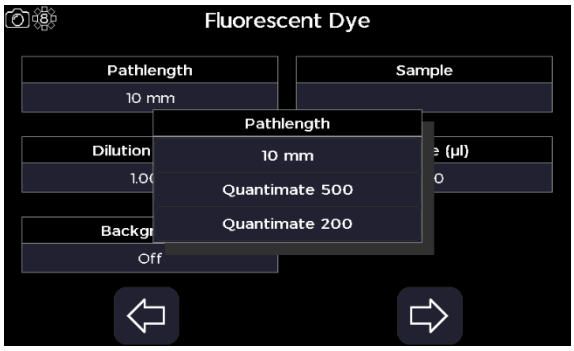


Step 4
If two dyes are being measured, repeat step 2 for the second dye. If one dye is being measured go straight to step 5.



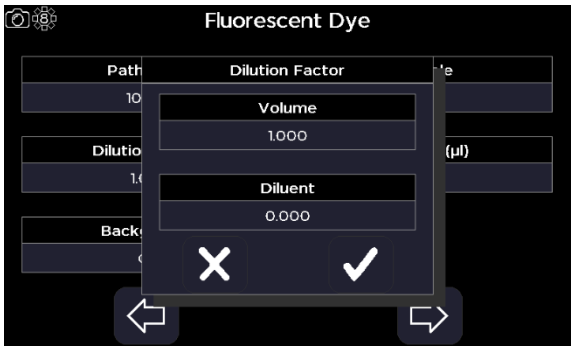
Step 5
Select the nucleic acid target to "none", "ssDNA (260nm)", "dsDNA (260nm)", "RNA (260nm)" "Cy Dye dUTP", "Oligo (260nm)", or "Custom".

For the "Custom" target selection, press the down arrow and enter a factor of up to four significant figures.



Step 6
Proceed to the next parameter screen using the right/forward arrow.

Step 7
Select the pathlength; “10 mm”, “Quantimate 500”, or “Quantimate 200”.

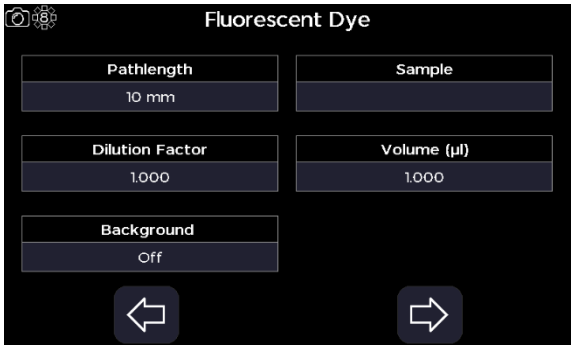


Step 8
Enter any dilution factor to be applied to the absorbance measurement.

Set the initial sample volume of a value of up to four significant figures.

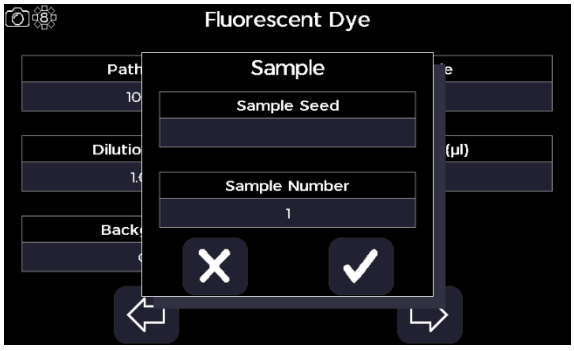
Then set the amount of diluent added to the initial volume of a value of up to four significant figures.

Implement the changes and return to the parameters screen by selecting the confirm icon.

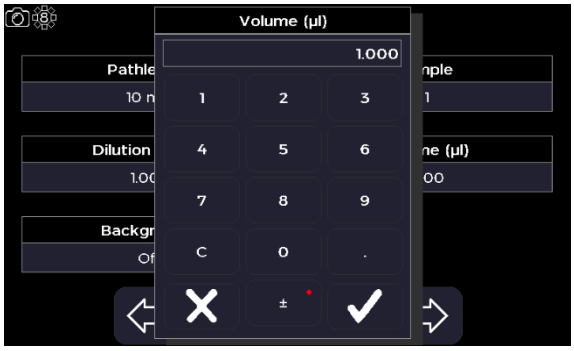


Step 9
Set the background, “On” or “Off”.

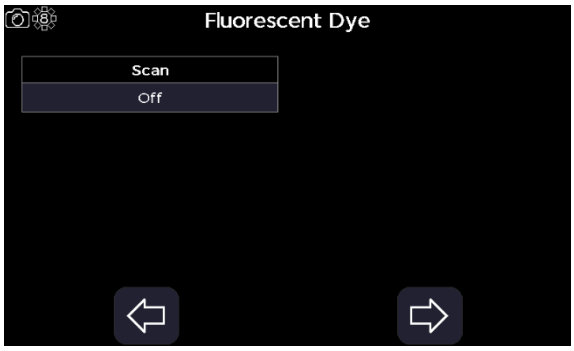
For background set to “On”, set the background wavelength to between 202 and 997.



Step 10
Select the sample name to bring up the sample window, enter a sample seed prefix and the incremental sample number starting value. Confirm the settings using the confirm icon.

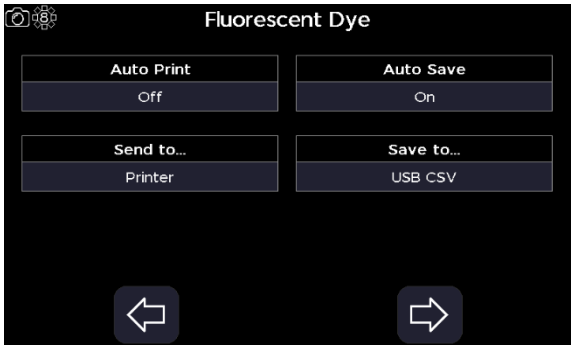


Step 11
Set the volume to a value of up to four significant figures.



Step 12
Proceed to the next parameter screen using the right/forward arrow.

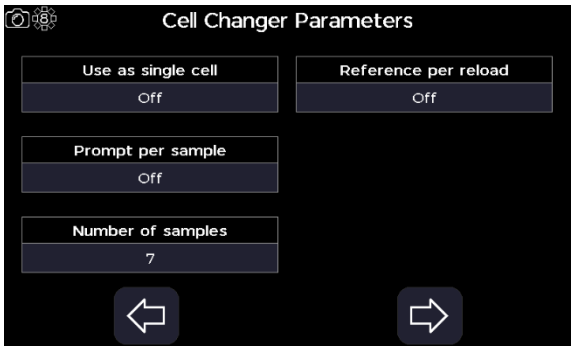
Step 13
Set display scan to “On” or “Off”.



Step 14
Proceed to the next parameter screen using the right/forward arrow.

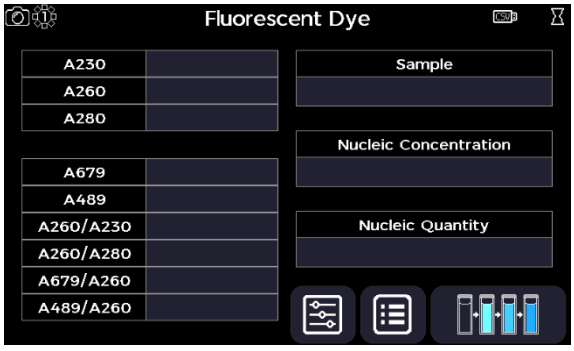
Step 15
Set auto print to “On” or “Off”. If auto print is set to “On”, select the print to hardware from “Internal Printer”, “PC via USB”, or “USB Mass Storage” depending on what hardware is connected to the instrument.

Step 16
Set auto save to “On” or “Off”. If auto save is set to “On”, select the save to hardware from “USB CSV”, “USB”, or “Internal” depending on what hardware is connected to the instrument.

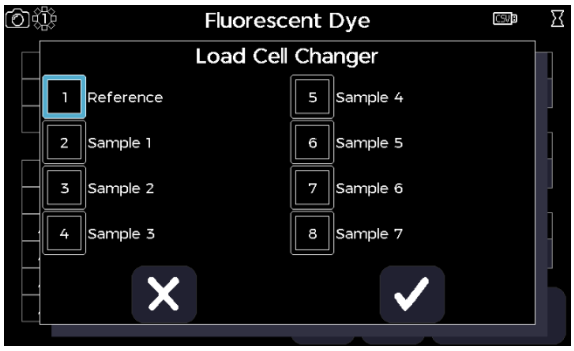


Step 17
Proceed to the next parameter screen using the right/forward arrow.

Step 18
Set whether to use as single cell to “On” or “Off”. If set to “Off”, set the position prompt per sample to “On” or “Off”, set the number of samples to between 2 and 100, and set whether to retake the reference between reload to “On” of “Off”.

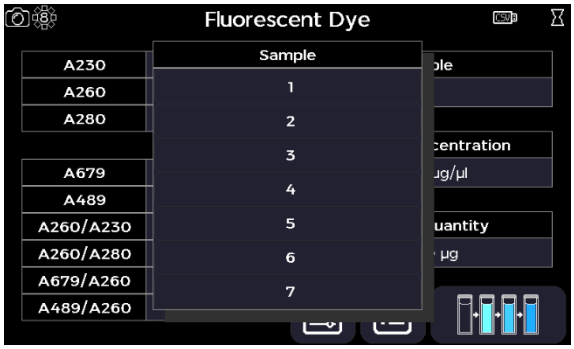


Step 19
Proceed to the measurement screen using the right/forward arrow.

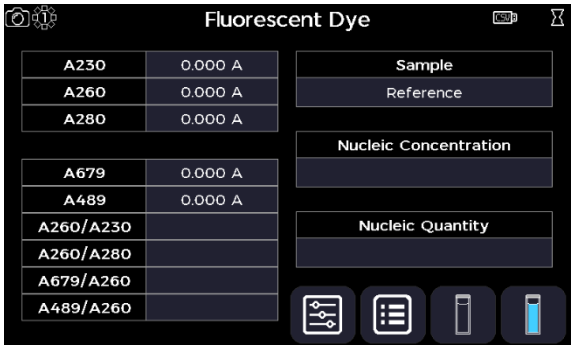


If using a single cell holder, or a cell changer set to use as a single cell holder, skip steps 20 through 21 and go straight to step 22.

Step 20
Press the batch measurement icon, then load the cell changer according to the cell changer prompt. Then select the confirm icon.



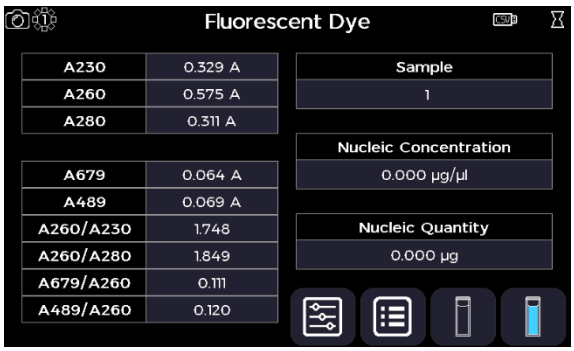
Step 21
The acquired reference sample baseline will have be applied to all subsequent sample measurements. The sample measurements can be viewed by pressing the sample name test box and selecting the appropriate sample from the list.



If using a cell changer, skip steps 22 through 23 and go straight to step 24.

Step 22
Insert the reference sample then take a reference measurement using the reference measurement icon.

The acquired reference sample baseline will be applied to all subsequent sample measurements.



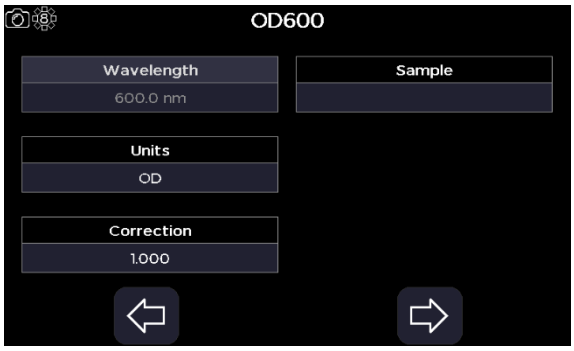
Step 23
Replace the previous sample with a test sample then take a sample measurement using the confirm key.

Repeat for all samples.

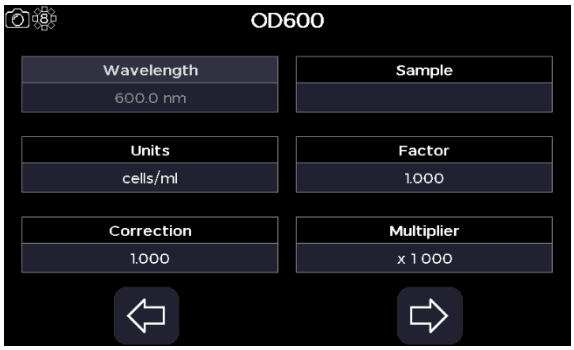
Step 24
Return to the home screen using the exit icon in the options menu **OR** use the additional options to save, print, and, or load previous measurements (see the Additional Options section).

OD 600

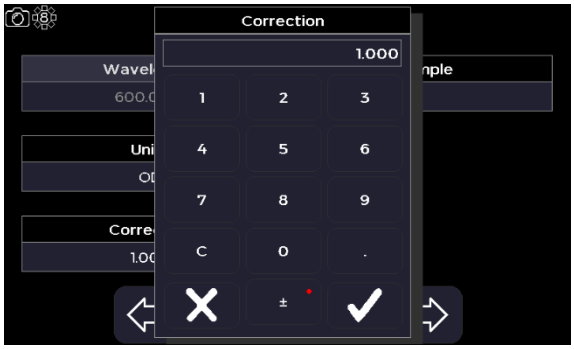
The OD 600 application is accessed from the home screen. It can be used to performed simple optical density measurements of microbial growth cultures.



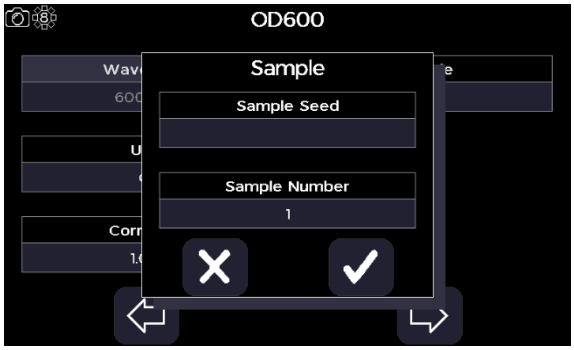
Step 1
Set the units, “OD” or “cells/ml”.



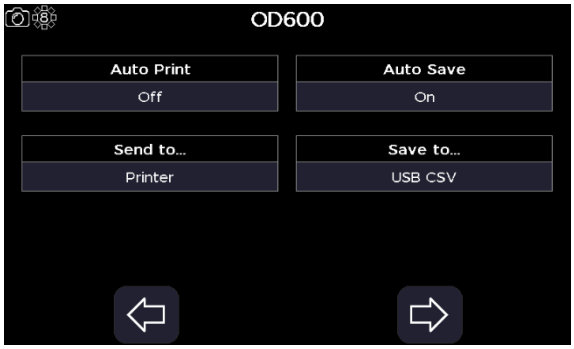
For “cells/ml” units, define the multiplication factor of a value of up to four significant figures, and set the multiplier, “×1 000” or “×1 000 000”.



Step 2
Enter any correction factor to be applied to the absorbance measurement of a value of up to four significant figures.



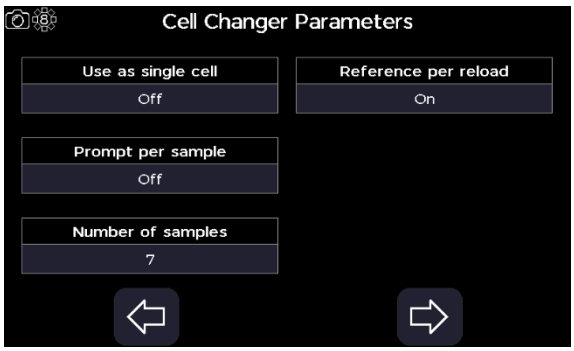
Step 3
Select the sample name to bring up the sample window, enter a sample seed prefix and the incremental sample number starting value. Confirm the settings using the confirm icon.



Step 4
Proceed to the next parameter screen using the right/forward arrow.

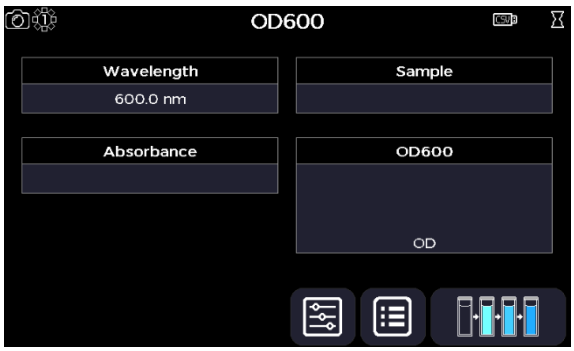
Step 5
Set auto print to “On” or “Off”. If auto print is set to “On”, select the print to hardware from “Internal Printer”, “PC via USB”, or “USB Mass Storage” depending on what hardware is connected to the instrument.

Step 6
Set auto save to “On” or “Off”. If auto save is set to “On”, select the save to hardware from “USB CSV”, “USB”, or “Internal” depending on what hardware is connected to the instrument.

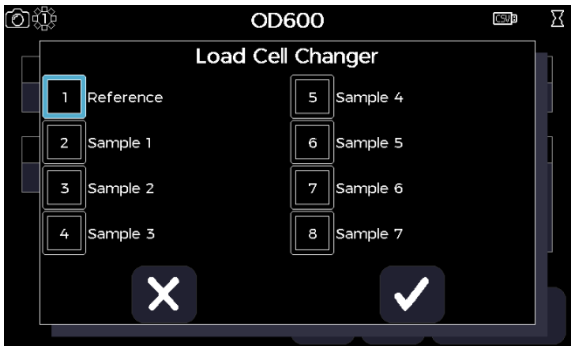


Step 7
Proceed to the next parameter screen using the right/forward arrow.

Step 8
Set whether to use as single cell to “On” or “Off”. If set to “Off”, set the position prompt per sample to “On” or “Off”, set the number of samples to between 2 and 100, and set whether to retake the reference between reload to “On” of “Off”.



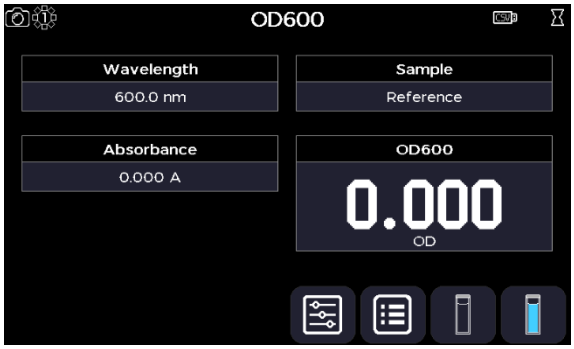
Step 9
Proceed to the measurement screen using the right/forward arrow.



If using a single cell holder, or a cell changer set to use as a single cell holder, skip steps 10 through 11 and go straight to step 12.

Step 10
Press the batch measurement icon, then load the cell changer according to the cell changer prompt. Then select the confirm icon.

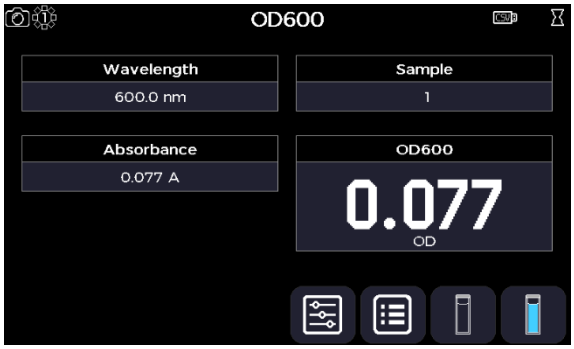
Step 11
The acquired reference sample baseline will have be applied to all subsequent sample measurements. The sample measurements can be viewed by pressing the sample name test box and selecting the appropriate sample from the list.



If using a cell changer, skip steps 12 through 13 and go straight to step 14.

Step 12
Insert the reference sample then take a reference measurement using the reference measurement icon.

The acquired reference sample baseline will be applied to all subsequent sample measurements.



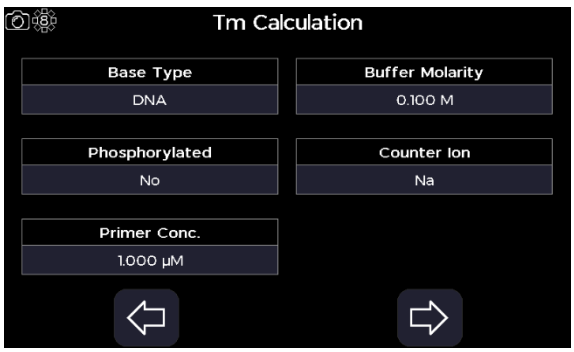
Step 13
Replace the previous sample with a test sample then take a sample measurement using the confirm key.

Repeat for all samples.

Step 14
Return to the home screen using the exit icon in the options menu **OR** use the additional options to save, print, and, or load previous measurements (see the Additional Options section).

Tm Calculation

The melting temperature (Tm) Calculation application is selected from the home screen. It can be used to determine the theoretical melting point of a PCR primer at the measured concentration from its nucleotide base sequence, using the nearest-neighbour model. All calculations applied within the Tm Calculation application are described in the Useful Calculations section.



Step 1
Select the base type to “DNA” or “RNA”.

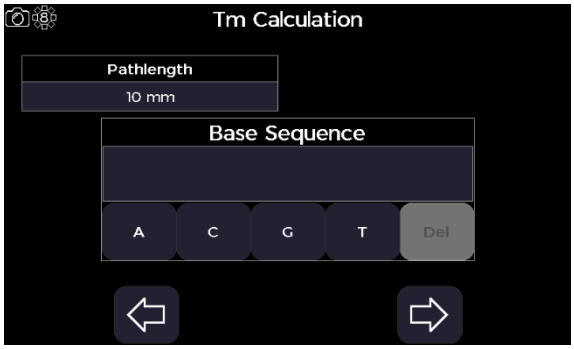
Step 2
Select if the nucleotides are phosphorylated to “No” or “Yes”.

Step 3
Enter the PCR primer concentration a value of up to four significant figures.

Step 4
Enter the buffer molarity (µM) to a value of up to four significant figures.

Step 5
Select the counter ion present in the buffer to “Na”, “K”, “TEA”. “TEOA”, or “Other”.

For the “Other” counter ion selection, enter a molecular weight (“Other MW”) of up to four significant figures.

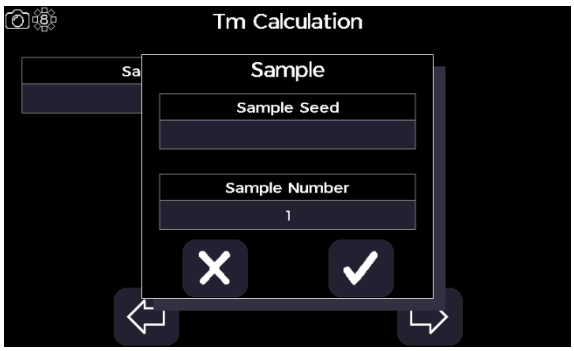


Step 6
Proceed to the next parameter screen using the right/forward arrow.

Step 7
Select the pathlength to “10 mm”, “Quantimate 500”, or “Quantimate 200”.

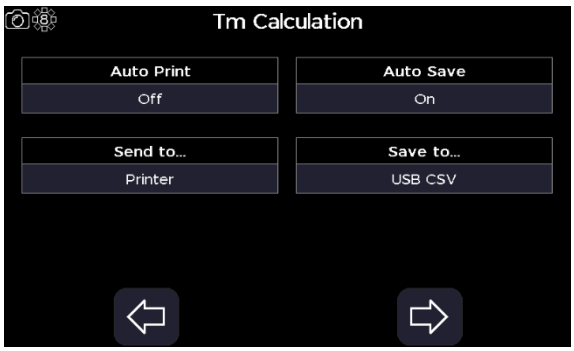
Step 8
Select the integration time from “1 second”, “2 seconds”, or “5 seconds”.

Step 9
Enter the primer nucleotide base sequence of up to 60 bases.



Step 10
Proceed to the next parameter screen using the right/forward arrow.

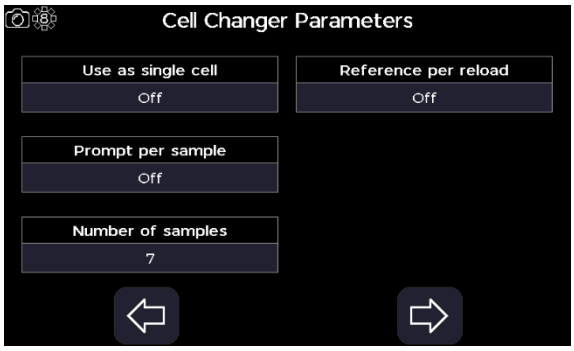
Step 11
Select the sample name to bring up the sample window, enter a sample seed prefix and the incremental sample number starting value. Confirm the settings using the confirm icon.



Step 12
Proceed to the next parameter screen using the right/forward arrow.

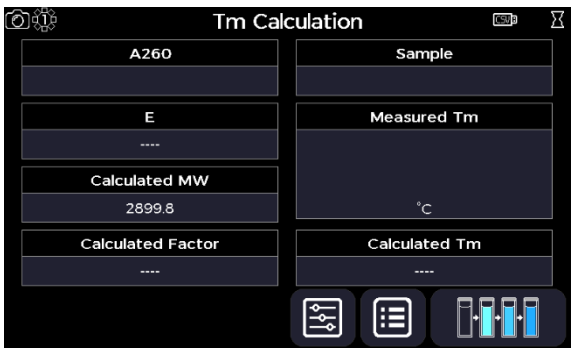
Step 13
Set auto print to “On” or “Off”. If auto print is set to “On”, select the print to hardware from “Internal Printer”, “PC via USB”, or “USB Mass Storage” depending on what hardware is connected to the instrument.

Step 14
Set auto save to “On” or “Off”. If auto save is set to “On”, select the save to hardware from “USB CSV”, “USB”, or “Internal” depending on what hardware is connected to the instrument.

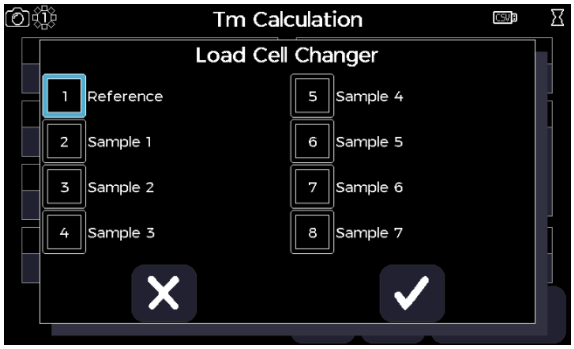


Step 15
Proceed to the next parameter screen using the right/forward arrow.

Step 16
Set whether to use as single cell to “On” or “Off”. If set to “Off”, set the position prompt per sample to “On” or “Off”, set the number of samples to between 2 and 100, and set whether to retake the reference between reload to “On” of “Off”.

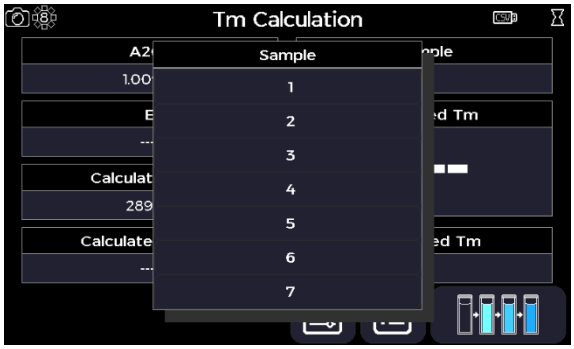


Step 17
Proceed to the measurement screen using the right/forward arrow.

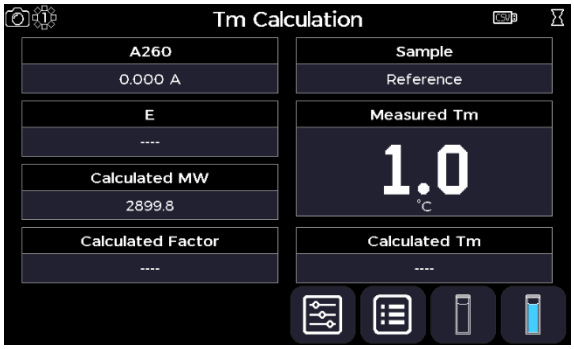


If using a single cell holder, or a cell changer set to use as a single cell holder, skip steps 18 through 19 and go straight to step 20.

Step 18
Press the batch measurement icon, then load the cell changer according to the cell changer prompt. Then select the confirm icon.



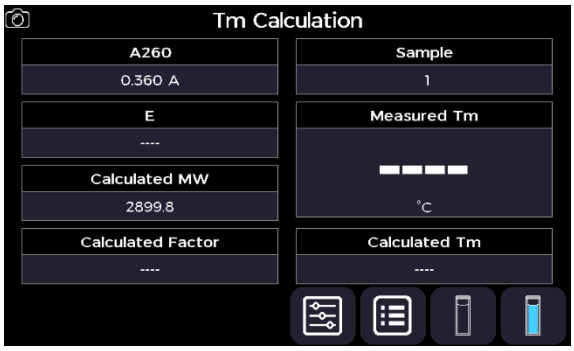
Step 19
The acquired reference sample baseline will have be applied to all subsequent sample measurements. The sample measurements can be viewed by pressing the sample name test box and selecting the appropriate sample from the list.



If using a cell changer, skip steps 20 through 21 and go straight to step 22.

Step 20
Insert the reference sample then take a reference measurement using the reference measurement icon.

The acquired reference sample baseline will be applied to all subsequent sample measurements.



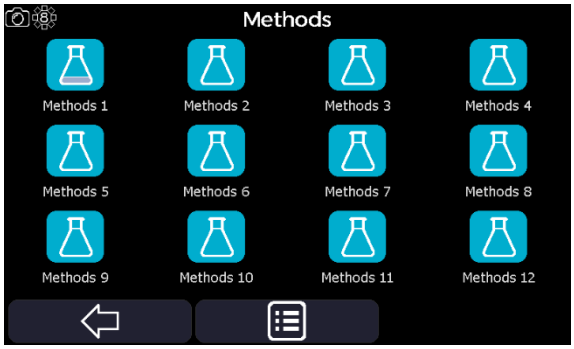
Step 21
Replace the previous sample with a test sample then take a sample measurement using the confirm key.

Repeat for all samples.

Step 22
Return to the home screen using the exit icon in the options menu **OR** use the additional options to save, print, and, or load previous measurements (see the Additional Options section).

Methods

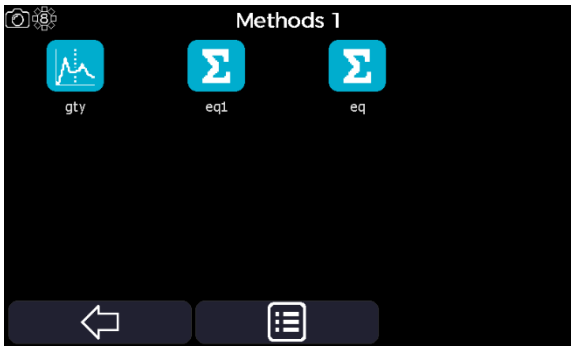
The Methods, Favourites, and USB Methods screens are accessed from the home screen. They are directories to save custom methods to, using the options menu from the results screen (see the Additional Options section).



Select the methods subdirectory, where the custom method is saved.

PLEASE NOTE

This only applies to the Methods screen. When selected from the home screen, the Favourites and Memory Stick screens directly display the saved custom method applications.



Once the directory containing the saved custom method application is accessed, there are several options available.

1. To open a saved custom method, simply select the application.

2. To delete a saved custom method application, use the options icon, the bin icon, and then select the method name to delete.

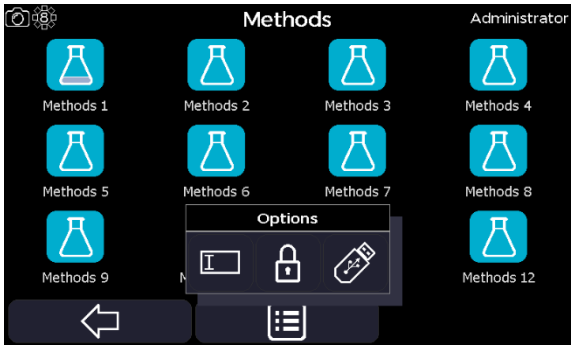
   Confirm the selection using the confirm icon.

3. To lock a saved custom method application, use the options icon, the lock icon, and then select the method name to lock and enter a pass code.

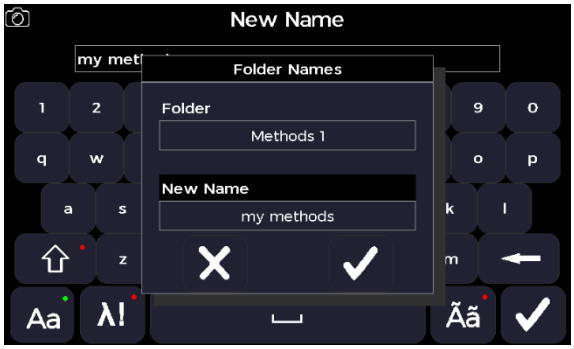
   Confirm the selection using the confirm icon.

4. To unlock a protected saved custom method application, use the options icon, the unlock icon, and then select the method name to unlock and enter a pass code.

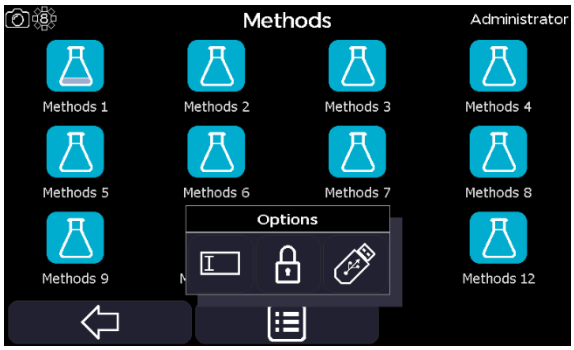
   Confirm the selection using the confirm icon.



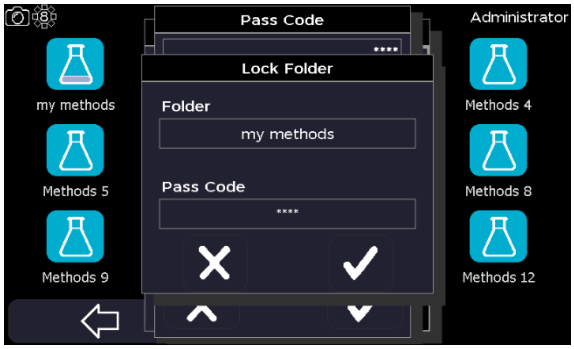
5. By clicking on the Options icon , the method folders can be renamed using the Rename icon



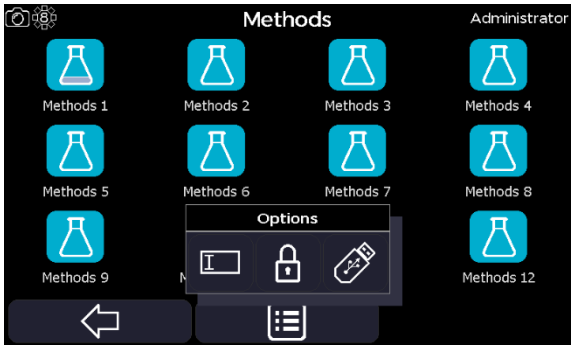
6. On the New Name screen, select the folder you want to rename, then in the New Name box, type in the new name and touch the Accept icon.



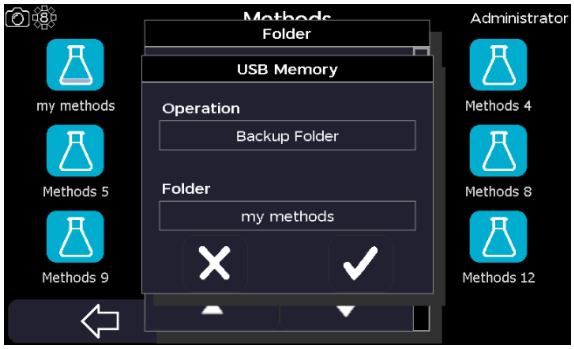
7. A method folder can be locked with a passcode by selecting the lock icon .



8. Then select the folder you want to protect in the drop down list, enter a passcode and touch the Accept icon.



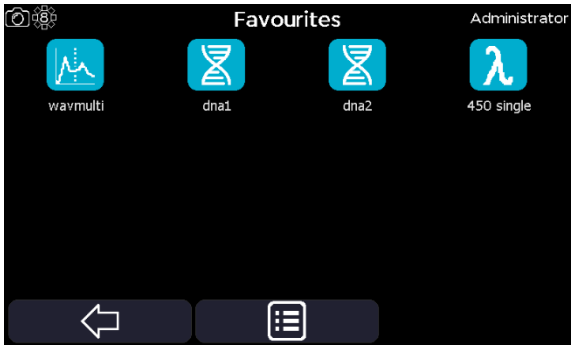
9. The USB stick icon allow backing up or restoring one or all method folders to/from the USB memory stick



10. Select the operation required in the Operation drop down menu and select which folder this operation will be applied to and touch the Accept icon. In this example, the “my methods” folder will be backed up onto the USB memory stick.

Favourites

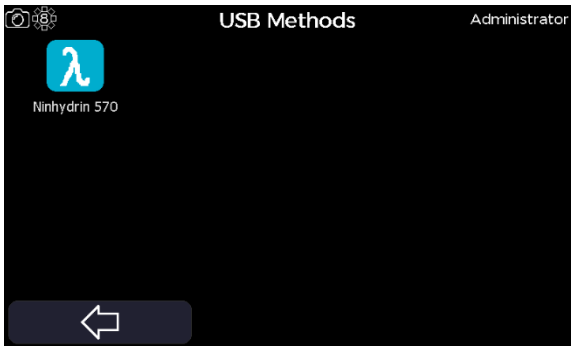
The Favourites folder contains all the methods that have been saved in the Favourites directory, when selected from any other applications screens.



1. View of the Favorites folder. When the “Favourites” folder has been selected when saving a method, this method will be stored here. Methods can also be deleted, locked or unlocked with a passcode using the Options icon .

USB Methods

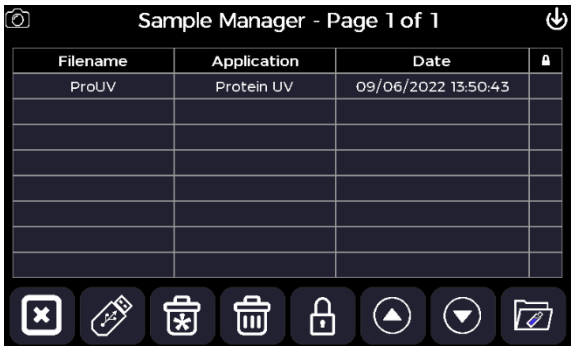
The USB Methods folder contains all the methods that have been saved on the USB memory stick, when selected from any other applications screens.



1. View of the USB Methods folder. When the “USB Methods” folder has been selected when saving a method, this method will be stored here. In this example, the “Ninhydrin 570” single wavelength method has been saved onto the USB memory stick.

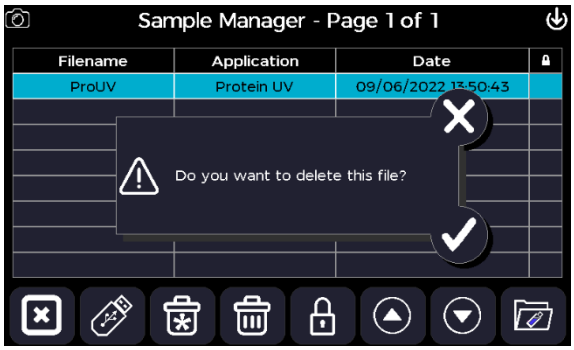
Sample Manager

The Sample Manager screen is accessed from the home screen. It is a directory to save result data to, using the options menu from the results screen (see the Additional Options section).



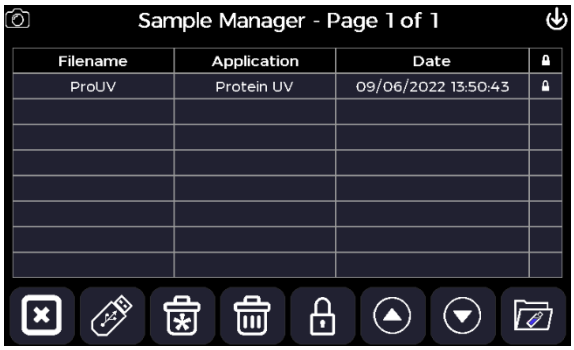
Once the sample manager directory application is accessed, there are several options available

1. To open saved results, simply highlight the applicable saved data entries then select the load sample icon.



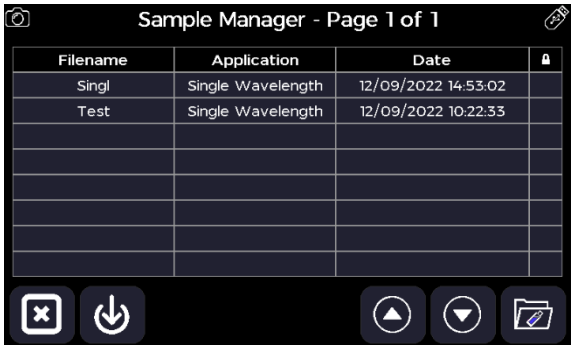
2. To delete saved results, simply highlight the applicable saved data entries then select the delete icon.

Confirm the selection using the confirm icon.



3. To lock or unlock saved results, simply highlight the applicable saved data entries then select the lock icon.

4. To view the results saved onto the USB memory stick, click on the USB stick icon.



5. To revert to the results saved onto the internal memory, click on the  icon

Additional Options

Additional options are available from the measurement screen using the options icon. The available options, in addition to those described in the 'Common Icons on the Option Menu' of the 'Frequently Used Icons' section, vary between applications.

Options Menu Icons

Icon	Name	Function	Application
	**Exit**	Exit the application and return to the application menu	All
	**Save data**	Save the sample data	All
	**Save method**	Save the method with the current parameters settings	All
	**Print**	Print the sample data from the specified printer	All
	**Auto-print**	Toggle auto print on (green) or off (red)	All
	**Load sample**	Open saved sample data	All
	**Trace Manager**	Open the trace manager to load samples	Wavescan, Kinetics
	**Vertical Cursor**	Toggle on or off the vertical cursor	Wavescan, Kinetics
	**Horizontal Cursor**	Toggle on or off the horizontal cursor	Wavescan, Kinetics
	**Trend Line**	Insert a trend line across section	Kinetics
	**Section Break**	Insert section break at cursor position	Kinetics
	**Standard Curve**	View the standard curve	Standard Curve, Colorimetric Protein
	**Spectrum**	View the sample spectrum	Substrate, DNA, RNA, Oligo, Fluorescent Dye, Protein UV, Protein Dye
	**Turn Page**	Toggle between multiple measurement screens	Fluorescent Dye

Status Bar Icons

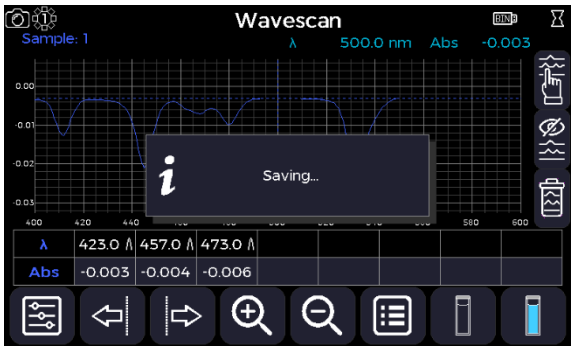
During the measurement process, various status icons are displayed in the status bar at the top of the screen. The icons displayed depend on the current process being undertaken and the defined settings.

Auto-print to external printer is active.	Printing to external printer.
Auto-print to computer via USB cable connection.	Printing to computer via the USB cable is active.
Auto-print to binary file to USB flash drive.	Printing to binary file to USB flash drive.
Auto-print to comma separate value file to USB flash drive.	Printing to comma separate value file to USB flash drive.
Auto-save to internal memory.	Saving to internal memory.
Screenshot icon.	Taking Screenshot and exporting as a bitmap file to USB flash drive.
Taking measurement.	Instrument failed initial calibration.

Taking and Saving Screenshots

It is possible to take a screenshot of any screen from the unit, providing a USB memory stick has been plugged in. When a screenshot is taken, it will automatically be saved onto the memory stick.

To take a screenshot, touch the camera icon on the top left corner of the screen



1. An Information pop-up message is displayed when taking a screenshot. Please note this information pop-up will NOT be seen on the actual screenshot. The screenshot can then be retrieved from the memory stick as an image in .BMP format.

USEFUL CALCULATIONS

Beer-Lambert Law

$$A = c\varepsilon l$$

A is the absorbance, which although unit-less is usually described as A or AU (absorbance units).

C is the concentration in molar units (M).

ε is the molar extinction coefficient in per molar unit per cm ($M^{-1}cm^{-1}$).

l is the pathlength in centimetres (cm).

As the absorbance value is the known quantity, the Beer-Lambert equation can be rearranged to make concentration (c) the product:

$$c = \frac{A}{\varepsilon \times l}$$

Alternative extinction coefficients can be applied to calculate the concentration in alternative units

ε		c
Molar extinction coefficient ($M^{-1}cm^{-1}$)	➡	Molar, or moles per litre, concentration (M, mol L^{-1})
Mass extinction coefficient ($g^{-1}cm^{-1}$)	➡	0.1 % Mass per volume concentration (g L^{-1})
E1% extinction coefficient ($mg^{-1}mL^{-1}cm^{-1}$)	➡	1 % Mass per volume concentration (10g L^{-1})

Conversions between molar, mass, E1% extinction coefficients:

$$\frac{\text{Molar Extinction Coefficient}}{\text{Molecular Weight (g mol}^{-1})} = \text{Mass Extinction Coefficient}$$

Mass Extinction Coefficient × 10 = E1% Extinction Coefficient

When E1% extinction coefficient are used, the absorbance is multiplied by 10 to present the concentration as a 0.1 % weight per volume (w/v) unit in keeping with convention:

$$c = \frac{A \times 10}{E1\% \times l}$$

References:

Beer, A. (1852). Bestimmung der Absorption des roten Lichts in farbigen Flüssigkeiten. 1st ed. Leipzig: Johann Ambbosius Barth.

Lambert, J. (1760). Photometrie. Photometria sive de ensura et gradibus luminis, colorum et umbrae. 1st ed. Augsburg: Eberhardt Klett, p.391.

Nucleic Acid Concentrations

Concentration = (A260 - A320) × Factor × Pathlength Factor × Units Factor × Dilution Factor

A260 is the absorbance at 260 nm.

A320 is the optional background absorbance at 320 nm.

Factor is the value defined within the application method parameters.

Pathlength Factor is based on the pathlength selected:

Selected Pathlength	Pathlength Factor
10 mm	1
5 mm	2
1 mm	10
0.5 mm	20
0.2 mm	50
0.125 mm	80

Units Factor is based on the units selected:

Selected Units	Units Factor
µg/ml	1
ng/µl	1
µg/µl	0.001
pmol/µl ACGT	Calculated from nucleotide sequence*
pmol/µl	User Defined+

* Calculated using method described by Ahnert and Patel (197, p. 272), specifically:

Molar Extinction Coefficient ($M^{-1}cm^{-1}$) = 15 200 × Number of A + 7 050 × Number of C+ 12 010 × Number of G + 8 400 × Number of T

The units factor is 1 000 000 ÷ Molar Extinction Coefficient ($M^{-1}cm^{-1}$).

+ The user defined coefficient for pmol/µl has to be 1 000 000 ÷ Molar Extinction Coefficient ($M^{-1}cm^{-1}$).

Dilution Factor is the value defined within the application method parameters.

References:

Ahnert, P. and Patel, S. (1997). Asymmetric Interactions of Hexameric Bacteriophage T7 DNA Helicase with the 5'- and 3'-Tails of the Forked DNA Substrate. Journal of Biological Chemistry, 272(51), pp.32267-32273.

Protein Concentrations

$$\text{Concentration} = (((A280 - A320) \times F280) - ((A260 - A320) \times F260)) \times \text{Pathlength Factor} \times \text{Units Factor} \times \text{Dilution Factor}$$

A280 is the absorbance at 280 nm.

A320 is the optional background absorbance at 320 nm.

F280 and F260 are the factors associated with the mode selected:

Mode	F280	F260
Christian Warburg	1.55	0.76
BSA	1.49	N/A
IgG	0.73	N/A
Lysozyme	0.38	N/A
Molar Extinction*	$\frac{\text{Molecular Weight}}{\text{Molar Extinction}}$	N/A
Mass Extinction*	$\frac{1}{\text{Mass Extinction}}$	N/A
E 1%*	$\frac{10}{\text{E 1\%}}$	N/A
Custom	Custom	Custom

* Molecular weight, molar extinction, mass extinction, and E 1% are the respective values defined within the application method parameters.

Pathlength Factor is based on the pathlength selected:

Selected Pathlength	Pathlength Factor
10 mm	1
5 mm	2
1 mm	10
0.5 mm	20
0.2 mm	50
0.125 mm	80

Units Factor is based on the units selected:

Selected Units	Units Factor
µg/ml	1000
ng/µl	1000
µg/µl	1
mg/ml	1

Dilution Factor is the value defined within the application method parameters.

References:

Ahnert, P. and Patel, S. (1997). Asymmetric Interactions of Hexameric Bacteriophage T7 DNA Helicase with the 5'- and 3'-Tails of the Forked DNA Substrate. Journal of Biological Chemistry, 272(51), pp.32267-32273.

Nucleic Acid and Protein Purity Ratios

$$\text{A260/A280} = \frac{\text{A260} - \text{A320}}{\text{A280} - \text{A320}}$$

$$\text{A260/A230} = \frac{\text{A260} - \text{A320}}{\text{A230} - \text{A320}}$$

A260 is the absorbance at 260 nm.

A280 is the absorbance at 280 nm.

A230 is the absorbance at 230 nm.

A320 is the optional background absorbance at 320 nm.

References:

Measuring protein concentration in the presence of nucleic acids by A280/A260: The method of Warburg and Christian. (2006). Cold Spring Harbor Protocols (1).

Fluorescent Dye Quantity

$$\text{Quantity (pmol)} = (\text{Adye - A320}) \times [\text{Pathlength Factor}] \times \text{Volume} \times \text{Dilution Factor} \times \frac{1\,000\,000}{\text{Extinction Coefficient}}$$

Adye is the absorbance value at the dye λ_{max}.

A320 is the optional background absorbance at 320 nm.

Pathlength Factor is based on the pathlength selected:

Selected Pathlength	Pathlength Factor
10 mm	1
5 mm	2
1 mm	10
0.5 mm	20
0.2 mm	50
0.125 mm	80

Volume is the value defined within the application method parameters.

Dilution Factor is the value defined within the application method parameters.

Extinction Coefficient is the value defined within the application method parameters.

Fluorescent Dye Concentration

$$\text{Concentration (pmol/µl)} = \frac{\text{Quantity}}{\text{Volume}}$$

Quantity is the calculated fluorescent dye quantity.

Volume is the value defined within the application method parameters.

Fluorescent Frequency of Incorporation (FOI)

$$\text{FOI (dye/kb)} = \frac{(\text{Adye} \times 1\,000\,000 \times \text{Molecular Weight})}{(\text{Extinction Coefficient} \times \text{A260} \times \text{Factor})}$$

Adye is the absorbance value at the dye λ_{max}.

Molecular Weight is fixed 324.5 g mol^{-1} which is an average molecular weight of the nucleotides

Extinction Coefficient is the value defined within the application method parameters.

A260 is the dye corrected absorbance at 260 nm.

Factor is the value defined within the application method parameters.

Fluorescent Dye Incorporation

$$\text{Dye Incorporation (pmol/µg)} = \frac{\text{FOI} \times 1\,000}{\text{Molecular Weight}}$$

FOI is the calculated Frequency of Incorporation.

Molecular Weight is fixed 324.5 g mol^{-1} which is an average molecular weight of the nucleotides

Melting Temperature (T_m)

$$T_m\ (°C) = \frac{\Delta H}{(16.6 \times \log_{10}[Buffer]) + (\alpha + \Delta S + (R \times \ln[c \div 4])) - 273.15}$$

ΔH is the change in enthalpy (kcal mol^{-1}) and ΔS is the change in entropy (kcal K^{-1} mol^{-1}), and are the sum values of their nearest-neighbour pair values, specifically:

Molecule	DNA		RNA	
Pair*	**ΔH**	**ΔS**	**ΔH**	**ΔS**
AA:TT/UU	-9.1	-0.0240	-6.6	-0.0184
AT/AU:TA/UA	-8.6	-0.0239	-5.7	-0.0155
TA/UA:AT/AU	-6.0	-0.0169	-8.1	-0.0226
CA:GT/GU	-5.8	-0.0129	-10.5	-0.0278
GT/GU:CA	-6.5	-0.0173	-10.2	-0.0262
CT/CU:CG	-7.8	-0.0208	-7.6	-0.0192
GA:CT/CU	-5.6	-0.0135	-13.3	-0.0355
CG:GC	-11.9	-0.0278	-8.0	-0.0194
GC:CG	-11.1	-0.0267	-14.2	-0.0349
GG:CC	-11.0	-0.0266	-12.2	-0.0297

* The nucleotide pair on the left of the colon is the 5’ to 3’ sequence while the nucleotide pair on the right of the colon is the 3’ to 5’ sequence.

The nucleotide pair on the left of the forward-slash is the DNA sequence while the nucleotide pair on the right of the forward-slash is the RNA sequence.

Buffer is the buffer concentration (M).

α is the helix initiation factor fixed at -0.0108 kcal K^{-1} mol^{-1}.

R is the gas constant fixed at 0.001987 kcal K^{-1} mol^{-1}.

c is the calculated nucleic acid concentration (M), specifically:

$$c = A260 \times \text{Calculated Factor}$$

A260 is the absorbance at 260 nm.
Calculated Factor is calculated from the molecular weight and molar extinction coefficient (E).

$$\text{Calculated Factor} = \frac{\text{Molecular Weight}}{\text{Molar Extinction Coefficient}}$$

Molecular Weight is calculated for the base sequence defined within the application method parameters.

Molecular Weight of DNA = 312.2 × number of A + 288.2 × number of C + 328.2 × number of G + 303.2 × number of T + K

Molecular Weight of RNA = 312.2 × number of A + 288.2 × number of C + 328.2 × number of G + 289.2 × number of U + K

K is phosphorylation constant, specifically

$$K_{(\text{Phosphorylated})} = 17 + (N+2) \times \text{Counter Ion}$$

$$K_{(\text{Non-phosphorylated})} = -61 + (N+1) \times \text{Counter Ion}$$

N is the base sequence length
Counter ion is the molecular weight of the counter ion selected:

Counter Ion	Pathlength Factor
Na	23.00
K	39.10
TEA[1]	102.2
TEOA[2]	149.19
Other*	Custom

[1] Triethylamine
[2] Triethanolamine
* Other is values defined within the application method parameters.

Molar Extinction Coefficient is calculated for the base sequence defined within the application method parameters, and is the sum values of their nearest-neighbour pair values, specifically:

	A	C	G	T/U*
A	13.7	10.6	12.5	11.4
C	10.6	7.3	9.0	7.6
G	12.6	8.8	10.8	10.0
T/U*	11.7/12.3	8.1/8.6	9.5/10.0	8.4/9.8

* The nucleotide on the left of the forward-slash is the DNA sequence while the nucleotide on the right of the forward-slash is the RNA sequence.

References:

Breslauer, K., Frank, R., Blocker, H. and Marky, L. (1986). Predicting DNA duplex stability from the base sequence. Proceedings of the National Academy of Sciences, 83(11), pp.3746-3750.

SantaLucia, J. (1998). A unified view of polymer, dumbbell, and oligonucleotide DNA nearest-neighbor thermodynamics. Proceedings of the National Academy of Sciences, 95(4), pp.1460-1465.

OD 600

$$OD = A600 \times Correction$$

$$Cell/ml = A600 \times Correction \times Factor$$

A600 is the absorbance at 600 nm.

Correction is value defined within the application method parameters.

Factor is the value defined within the application method parameters.

TROUBLESHOOTING

Negative absorbance readings	• Sample measurements will be negative absorbance reading if the absorbance value of the reference is higher than the sample. • Negative readings can also result if reference and sample are interchanged or if the sample is very dilute and close to the absorbance of the reference.
Unexpected results	• Bubbles or contamination in the sample or reference can result in considerable errors. • Incorrect cuvette orientation. Rotate by 90˚ and repeat. • Incorrect cuvette material for UV measurement wavelengths. • Wrong pathlength selected in software. • Contact your supplier for advice on the minimum concentrations that can be measured.
Absorbance higher than expected	• Incorrect sample reference. • Incorrect cuvette orientation. • Incorrect cuvette material for measurement wavelengths. • Wrong pathlength selected in software. • Contamination in sample or on cuvette. • Check baseline, if greater than 0 A toggle background correction or us an appropriate reference sample. • Possible incorrect optical alignment. Contact technical support.
Absorbance lower than expected	• Incorrect sample reference. • Check sample and reference for contamination. • Check sample and reference samples are not the same. • Incorrect cuvette material for measurement wavelengths. • Wrong pathlength selected in software. • Check the beam height and buffer sample volume. • Check baseline, if greater than 0 A toggle background correction or us an appropriate reference sample. • Possible stray light issue. Contact technical support.
Poor reproducibility	• Insufficient sample in cuvette. • Cuvette in wrong orientation. • Cuvette material unsuitable for wavelengths used. • Concentration of sample too low or too high. For best results, the measured sample absorbance using a 10 mm pathlength cuvette should ideally be between 0.1 and 1.0 A. If absorbance is >1 A, measurement is no longer in the most linear range. • Particulates in sample. Absorbance measurements will not be accurate with turbid samples. • Possible noise or measurement stability issue. Contact technical support.
Instrument start up reported failure	• Check the cell holder is empty. • Check original 19V dc supply is connected and is fully engaged. • Report persistent failures to technical support.

Printing Sample Data

The Ultrospec 7500 allows users to print sample data in one of two ways:

Note: Only available printers will be shown in the *Print to…* options box.

External Printer

Data can be printed to an external printer when fitted. Data is printed with method header, instrument serial number, time/date and all sample results. If numerical data is being shown on the display only this data will be printed, if graphics are displayed on the screen these will be printed as well as numerical data.

The external printer is available as an accessory, part number: 80-2140-62 - U7500 Serial Printer Kit, and can easily be fitted to existing instruments – see instructions at the end of this section.

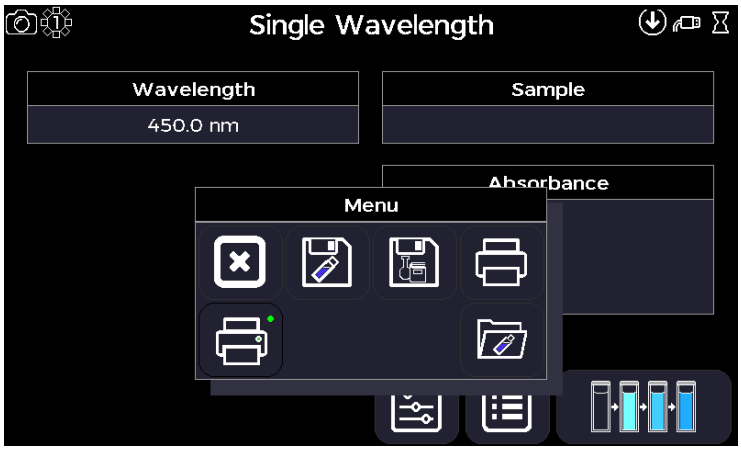
Print Via Computer (PVC)

Print *via* Computer (PVC) is an application running under Windows™ to enable the Ultrospec 7500 to transfer data into a PC environment. From there the data can be printed or saved in a variety of formats, including graphics and text formats or as an Excel™ file. 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 support several instruments simultaneously, limited only by hardware and the speed of the host system and is able to operate via USB simultaneously.

Installation and operating instructions for PVC can be found on the PVC USB Drive for the respective U7500 spectrophotometer or you can visit https://support.biochrom.co.uk for further information.

Manual Printing



If a method does not require sample data to be printed each time a measurement is taken it is possible to manually print sample data. This procedure is described below:

Set the desired print location in *Print to…* After collecting all required sample measurements select the Print icon from the options menu on the sample measurement screen.

Installing the External Printer

This part of the User manual explains how to install the external Seiko DPU S245 thermal printer, for any safety and operation precautions we refer to the Seiko DPU S245 User guide from the supplier's website: https://www.sii.co.jp/sps/eg/download/index.html

Open printer kit box and verify content:

DPU S245 Printer

Paper Roll

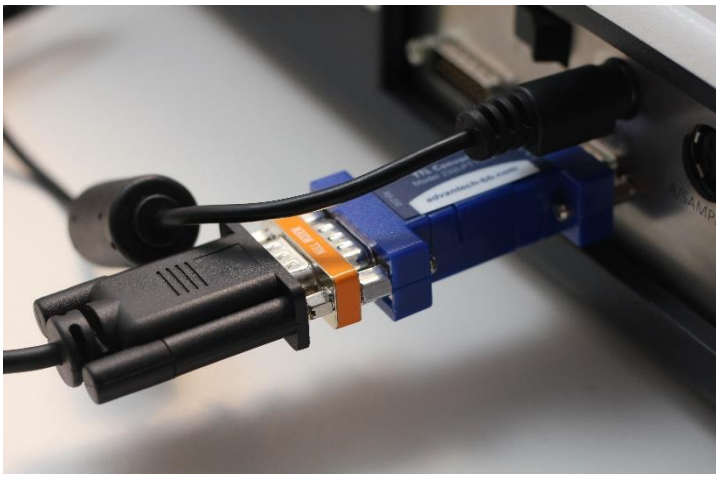
AC Adapter

Serial Cable

Power Cable

Insert paper roll into the printer and connect the printer with the AC Power Adapter.

Plug the serial cable with the blue TTL converter adaptor into the printing port of the back of the unit as shown below:



Serial Cable with TTL Converter

Connect the other end to the serial port, see picture below

Serial port

After setting the thermal paper in the printer, perform test printing. In test printing, the printer's function setting and character strings for testing are printed.

1. Make sure that the thermal paper is in the printer and the printer is turned off.
2. Press the POWER and FEED switches at the same time. When the POWER lamp lights, release the POWER switch, then the FEED switch.
3. Several dozens of lines of text are printed.
4. After test printing, the printer goes into print-ready mode.

The printer is now ready to use.

ACCESSORIES, SOFTWARE & DOCUMENTATION

Accessories List

80-2106-01	4-Position Cell Holder
80-2106-07	50 mm Single Cell Holder
80-2106-08	10 - 40 mm Water Heated Single Cell Holder (requires an external water thermocirculator)
80-2107-14	100 mm Single Cell Holder
80-2108-01	8-Position Cell Changer
80-2109-70	8-Position Water Heated Cell Changer (requires an external water thermocirculator)
80-2112-15	Sipper Unit
80-7100-50	Water Thermocirculator (range from 20°C to 60°C, temperature accuracy of ± 0.1°C)
80-7100-71	QuantiMate Micro-Volume Cuvette 200
80-7100-72	QuantiMate Micro-Volume Cuvette 500
80-2140-62	Printer Accessory Ultrospec 7500

PC Software

80-7100-31	Resolution Standard Software
80-7100-32	Resolution Life Science Software
80-7100-33	Resolution CFR Software

IQ/OQ and PQ Documentation

80-2119-89	IQ/OQ Documentation for Ultrospec 7500
80-2119-90	PQ Documentation for Ultrospec 7500

NOTE: IQ/OQ and PQ installations must be performed by a Field Service Engineer certified by Biochrom. For more information, please contact Biochrom or your local dealer.

Accessory Installation Guide

Instruction 1 – Multiple Position Cell Changers

Instruction 2 – Single Cell Holders

Instruction 3 – Water Heated Accessories

Instruction 4 – Sipper

Instruction 1 – Multiple Position Cell Changers

1. Remove the 8 Position Cell Changer, supplied as standard, by grasping the outer carousel with one hand. Then loosen the central finger screw with the other hand, until the cell changer comes free.
2. To install the new cell changer accessory, align the cell changer to the accessory motor by rotating the carousel until it falls into place. Secure the cell changer by grasping the outer carousel with one hand, and then tighten the central finger screw with the other hand until the cell changer is held tight.

Instruction 2 – Single Cell Holders

1. Remove the 8 Position Cell Changer, supplied as standard, by grasping the outer carousel with one hand. Then loosen the central finger screw with the other hand, until the cell changer comes free.
2. Insert the blanking plug, supplied with the single cell holder accessory, over accessory motor position.
3. To install the single cell holder accessory, align the keyhole clips over the sample compartments accessory studs. Then slide the keyhole clips to lock the cell holder in place.

Instruction 3 – Water Heated Accessories

1. Install the accessory base as per Instruction 1 or 2.
2. For water heated multiple position cell changers only, insert the anti-tangle plug into the central finger screw. Then fix the tube guide, supplied already threaded onto the tubes, using the screw attached to the holes at the front of the sample compartment base.
3. For all water heated accessories, unscrew and remove the blanking plate from the front of the sample compartment lid and replace it with the tube blanking plate provided.
4. The tubes can then be connected to a thermostatic water circulator.

Instruction 4 – Sipper

1. Remove the 8 Position Cell Changer, supplied as standard, by grasping the outer carousel with one hand. Then loosen the central finger screw with the other hand, until the cell changer comes free.
2. Unscrew and remove the blanking plate from the front of the sample compartment lid.
3. To install the Sipper accessory, align the keyhole clips over the cell changer drive mechanism studs.
4. Rotate the pump rotor by hand until the metal drive peg drops in place, taking care to position the beak mechanism and beak plug in the space left by the blanking plug. Then slide the keyhole clips to lock the Sipper in place.
5. Plug the Sipper lead into the appropriate cell compartment socket.
6. To install the single cell holder accessory, align the keyhole clips over the sample compartments accessory studs. Then slide the keyhole clips to lock the cell holder in place and plug any accessory lead into the appropriate cell compartment socket.
7. Insert the flowcell into the cell holder with the inlet and outlet PTFE transport tubes attached: The inlet tube should be at the front of the flowcell (identified by an arrow on the glass), and the flowcell should be facing towards the left.

CONTACT INFORMATION

Biochrom Ltd.

Unit 7, Enterprise Zone, 3970 Cambridge Research Park

Beach Drive, Waterbeach, Cambridge, UK, CB25 9PE

Phone: +44 1223 423 723

Biochrom U.S.

84 October Hill Road

Holliston, Massachusetts 01746

Phone: +1 508 893 8999| toll-free +1 800 272 2775

Harvard Bioscience (China) Co., Ltd.

Room 8C, Zhongxi Tower, 121 Jiangsu Road,

Changning District, Shanghai, China, 200050

Phone: +86 21 6226 0239

Support Centre:

https://support.biochrom.co.uk

E-mail: support@biochrom.co.uk

Websites:

www.biochrom.co.uk

www.biochromspectros.com

Harvard Bioscience • 84 October Hill Road • Holliston, MA 01742 • USA
Phone 800-272-2775 • support@hbiosci.com • www.harvardbioscience.com