Evolution 300 and Evolution 600

User Guide

Thermo
ELECTRON CORPORATION

Contents

Chapter 1 Welcome

Congratulations on your purchase of an Evolution™ 300 or Evolution 600 spectrophotometer from Thermo Electron! Our spectrophotometers integrate advanced hardware features with the power and flexibility of a wide range of Smart Accessories™.

Conventions used in this manual

This manual includes safety precautions and other important information presented in the following format:

Note Notes contain helpful supplementary information. ▲

Important Follow instructions labeled "Important" to avoid damaging the system hardware or losing data. ▲

⚠ Caution Indicates a potentially hazardous situation which, if not avoided, may result in minor or moderate injury. It may also be used to alert against unsafe practices. ▲

⚠ Warning Indicates a potentially hazardous situation which, if not avoided, could result in death or serious injury. ▲

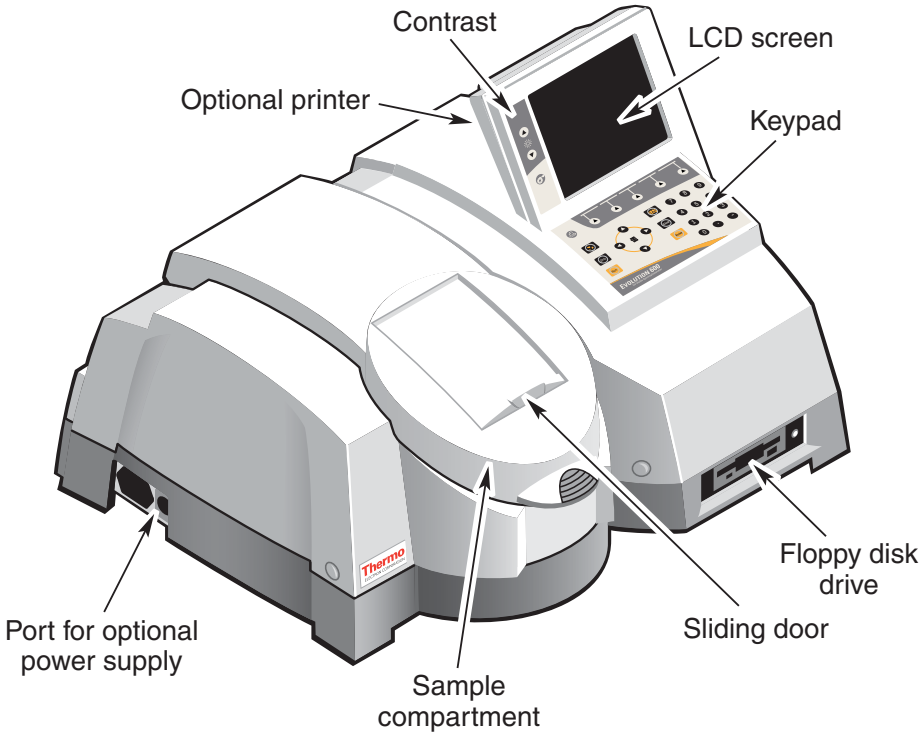
⚠ Danger Indicates an imminently hazardous situation which, if not avoided, will result in death or serious injury. ▲

Chapter 2 Spectrophotometer Basics

This chapter describes the major components of your spectrophotometer.

Spectrophotometer components

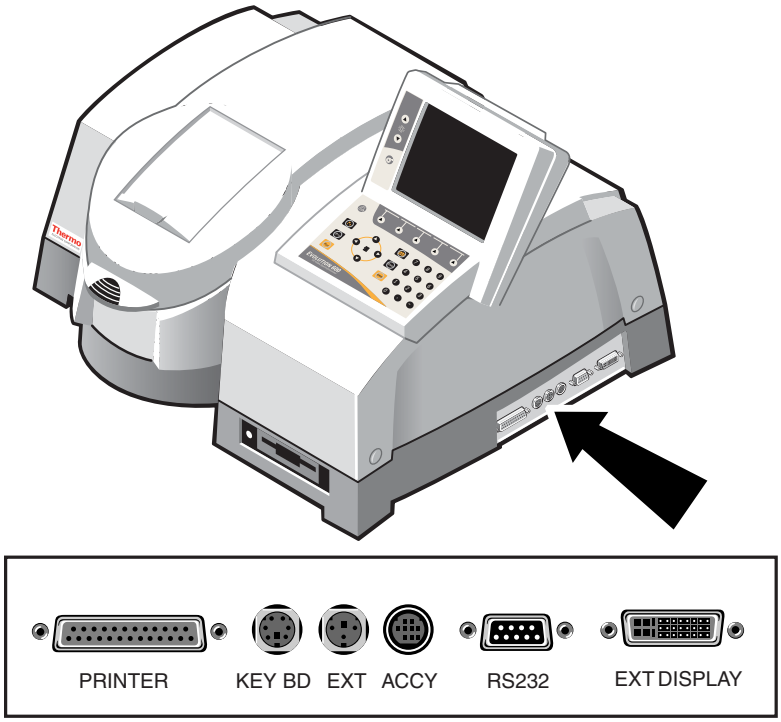
The following illustration identifies some major components visible on the outside of a typical Evolution spectrophotometer. (Some components may not be present on your spectrophotometer.)



Connectors

The following illustrations show the locations of the connectors on each side of the spectrophotometer.

Right side



Printer – Use to connect a printer with a parallel interface.

Key Bd – Use to connect an English keyboard (this is supplied by Thermo Electron to meet electrical emission requirements).

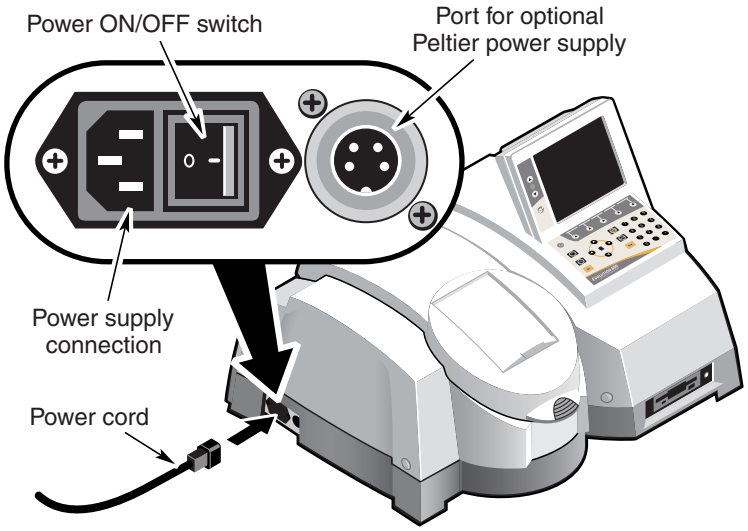
Ext – Use to connect an external TTL (or contact closure) trigger.

Accy –Uses to connect an external accessory.

RS232 – Use to connect a computer with an RS232 interface.

Ext Display – Use to connect to a computer monitor.

Left side

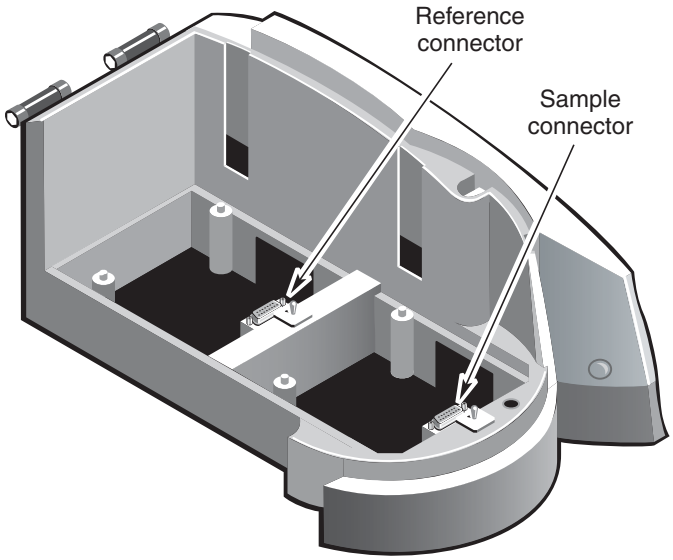


⚠ Danger Avoid shock hazard. Always power off the spectrophotometer and disconnect the power supply from the wall outlet or power strip before disconnecting the power supply from the spectrophotometer. ▲

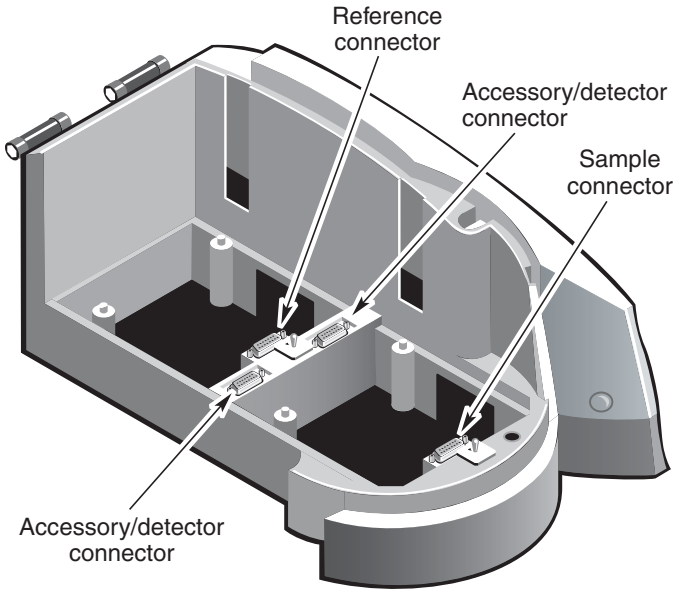
Connectors inside sample compartment

The following illustrations show the locations of the connectors inside the sample compartment.

Evolution 300



Evolution 600



Chapter 3 Accessories

This chapter briefly describes the types of sampling and system accessories that are available for your spectrophotometer. Complete descriptions and operating instructions are included with the accessories.

Manual accessories

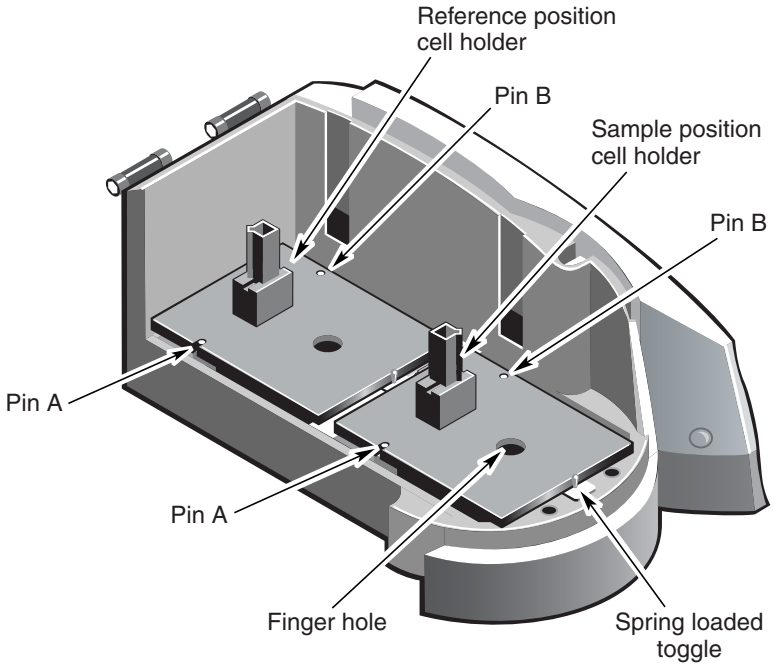
The Evolution 300 and the Evolution 600 are supplied with two standard rectangular cell holders. All manual accessories are installed and removed as follows:

Manual accessories are mounted on a common baseplate. This system ensures correct alignment of the accessory within the instrument automatically.

These accessories may be installed or removed without the need to switch off the instrument and may be installed in the sample (front) position and/or the reference (rear) position.

The accessory is positioned by two pins, A and B, at either side of the sample compartment, and a spring-loaded toggle towards the front of the instrument.

Installation



1. **Open the main lid of the sample compartment and remove any accessory that may be present in the sample (front) position.**

2. **Position the accessory so that the holes towards the rear are aligned over pins A and B in the instrument.**

   When properly aligned, gently push the front end of the accessory downward until plate snaps into place under the toggle.

   This positive location system ensures that the accessory is correctly aligned in the corresponding optical beam.

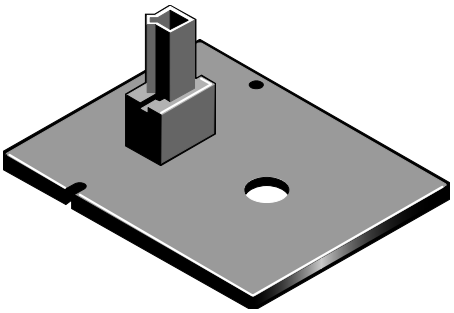
   For removal of the accessory:

   - Grip the accessory through the finger hole in the accessory and pull it upwards.

   - For accessories with no finger hole, grip the accessory using the handles.

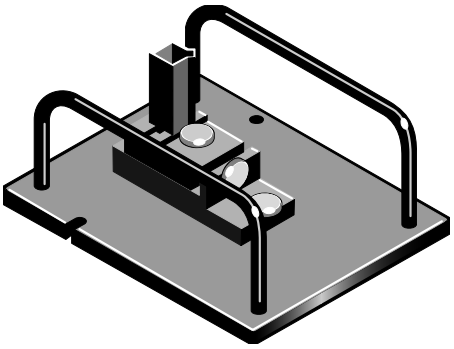
Standard rectangular cell holder

The instrument is supplied with two single cell holders (z = 8.5mm), which may be installed in the sample and reference positions.



Adjustable microcell holder

This accessory is recommended for use with small aperture cells. The adjustment permits precise positioning of the cell in the beam.



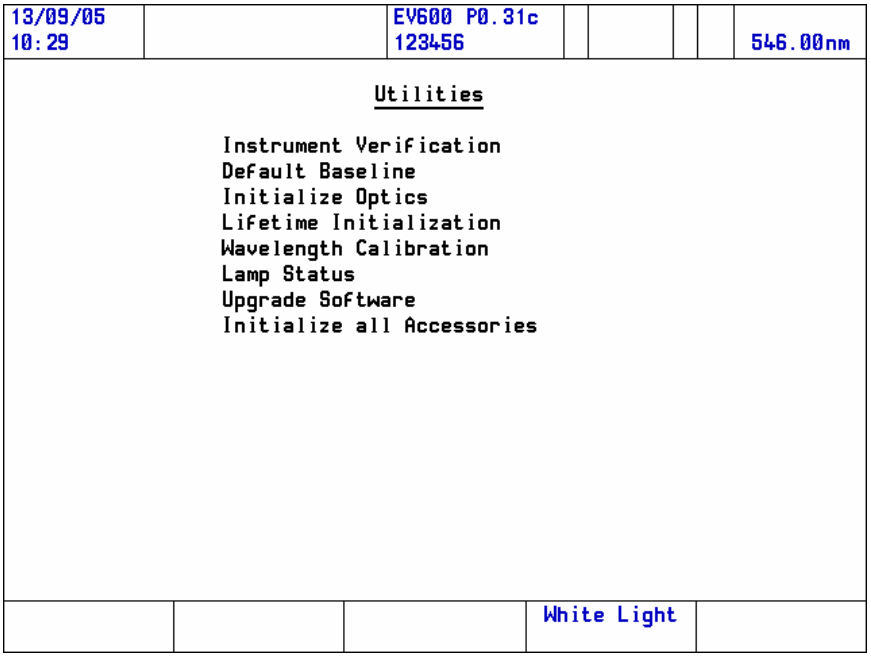
1. **Install the accessory into the instrument as described above.**

2. **Access the white light function in the operating software.**

   Local Control– From the Main Menu page, select Utilities, then press the White Light function key on the Utilities page.

   VISION – Select Command > White Light.

   The instrument will align the monochromator so that the zero order diffraction of the lamp(s) (white light) passes through the sample compartment. This provides a beam of white light, which can be seen when a business card or similar target is placed in the light path.



3. **Place the cuvette in the cell holder and loosen the securing screw A by turning it counterclockwise.**

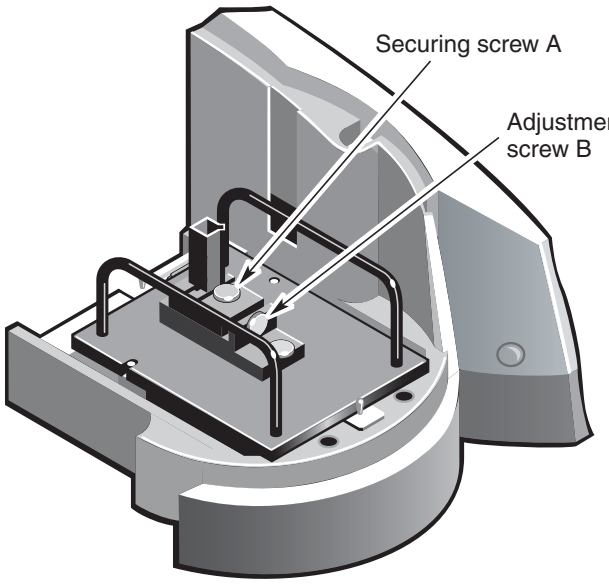
4. **Use adjustment screw B to align the cuvette so that the light beam falls on the cell aperture.**

Note Turning adjustment screw B clockwise moves the cell holder towards the front. Turning it counterclockwise moves the cell holder towards the rear. ▲

When the cell holder is aligned properly, the white light will not be seen on the edges of the cell holder.

5. **Tighten securing screw A to secure the cell holder position.**

When you have finished aligning the accessory in the beam, exit White Light mode by pressing White Light Off. This returns the monochromator to its normal position ready to continue taking measurements.

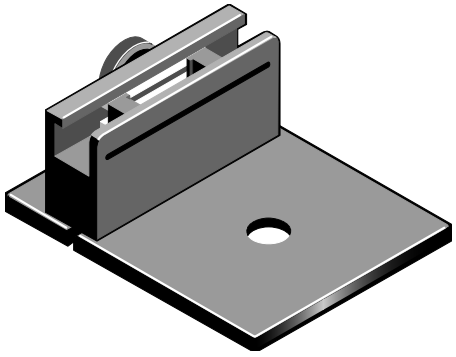


Long pathlength rectangular cell holder

This accessory is designed to accommodate rectangular cells with pathlengths ranging from 1 to 100mm.

The capacity of the cell holder is adjusted by turning the knob on the front, which alters the space between the gripping blocks.

Turn the knob until the space is longer than the pathlength required, insert the cell and reduce the space until the cell is held securely.

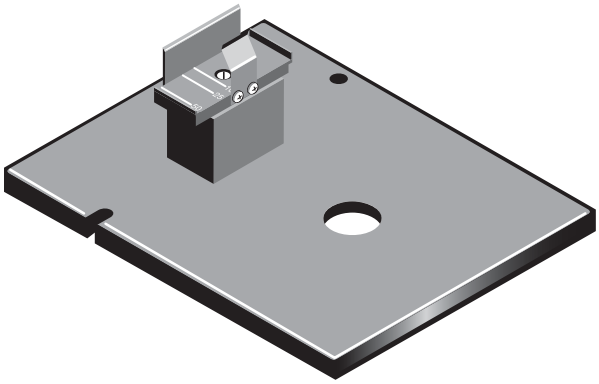


Important Do not overtighten. ▲

Cylindrical cell holder

This accessory is designed to accommodate cylindrical cells with 22mm external diameter and a pathlength of up to 50mm.

Push the cylindrical cell downwards into the cell holder so that it is secured in the required position by the spring clip and is in contact with the retaining ledge.

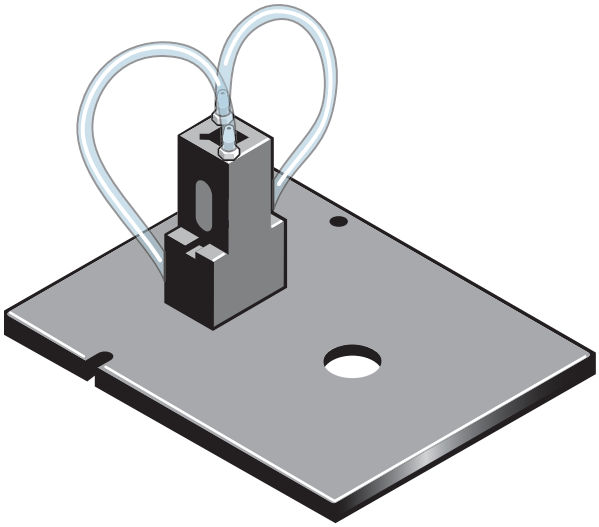


Thermostatted single cell holder

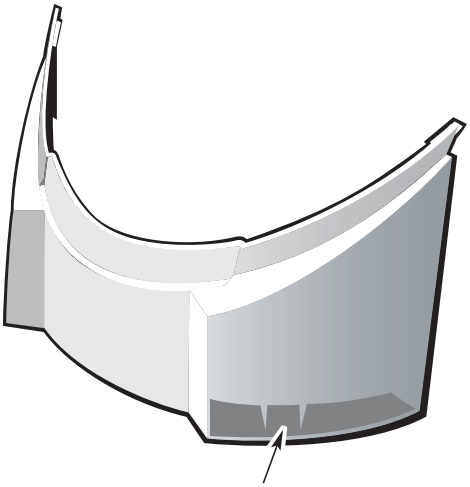
This accessory is designed to accommodate square cells with 8.8mm z-dimension and 10mm pathlength, and to operate at temperatures of 4 to 80°C (39 to 176°F).

Temperature control is achieved by means of an external temperature controlled circulator bath. Refer to the Parts List supplied on CD with your instrument for details of circulators available from Thermo Electron.

A tubing kit for Evolution thermostatted accessories is available for use with the recirculator, which includes the necessary connection kit for the circulator.



The sample compartment front panel has two slits through which the tubing can be routed. The slits are located to the left and right of the center, and open downwards. Foam baffles fit around the tubing to exclude light from the sample compartment.

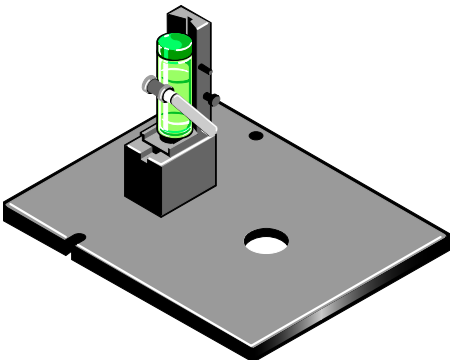


Position the in and out tubes either so that they will both pass through one port, or so that they pass on either side. Replace the sample compartment front panel, ensuring that the tubes are correctly located in the slit(s) and are not being pinched between the front panel and the body of the instrument. Ensure that the tubes do not obstruct the light beam. If necessary, secure the tubes using the self-adhesive tubing clips supplied.

Test tube/vial holder

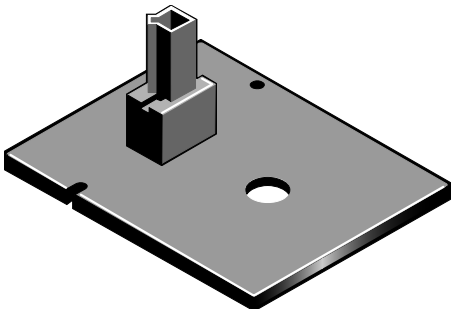
This accessory is designed for use with test tubes or vials from 10 to 21mm diameter and 40 to 100mm high.

Note Exceeding the recommended diameter may cause the retaining bar to clip the light beam. ▲



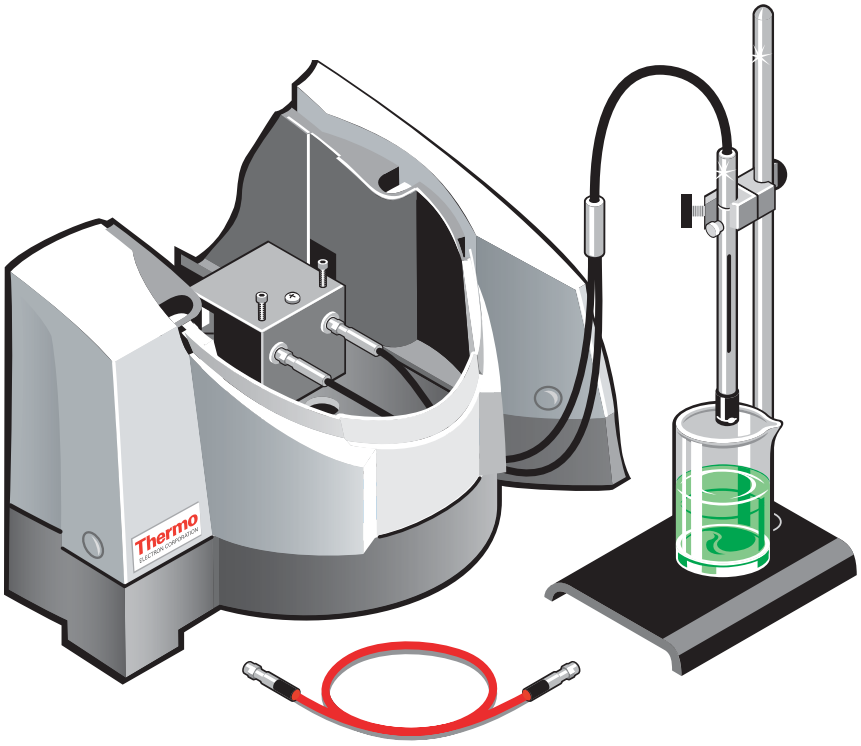
Rectangular cell holder

This cell holder allows the use of 15mm beam height filters and flowcells in the instrument.



Fiber optic sampling accessory

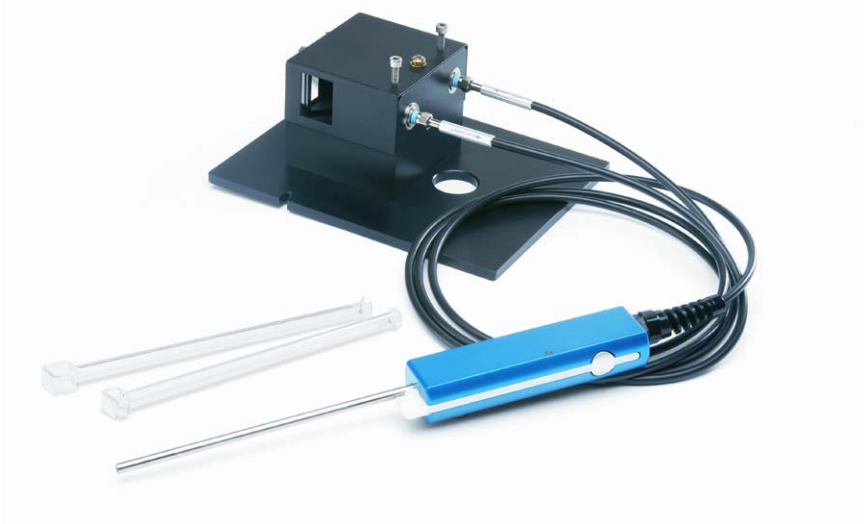
This accessory enables samples to be measured outside of the sample compartment.



Versa

The Versa fiber optic probe features a removable/disposable quartz tip. This tip covers the stainless steel portion of the probe and allows the probe to be used with strong acids.

If contamination is an issue in the laboratory, replacement tips are available. The tip of the probe is manufactured to the same tolerances of a standard cuvette and are available in 5, 10 and 20mm pathlengths.

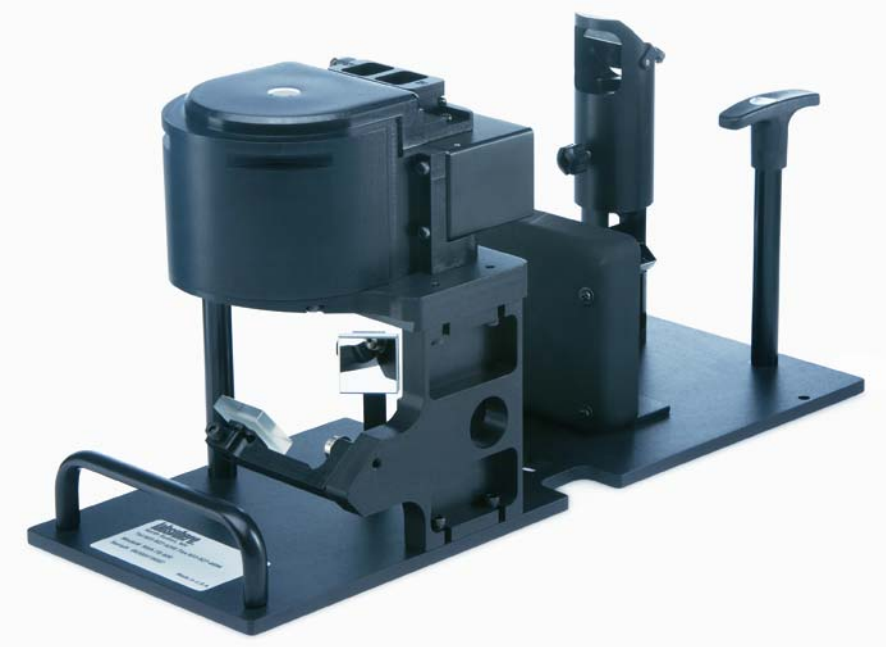


VN Absolute Specular Reflectance accessory

The VN Absolute Specular Reflectance accessory allows the absolute reflectance of samples as small as 12.5mm to be examined. The VN accessory has the advantage of having light only strike the sample surface once and requires no calibrated specular standards.

DRA-EV-600 Diffuse Reflectance accessory

The Diffuse Reflectance accessory allows solid samples to be measured horizontally outside the plane of the instrument. A double beam Spectralon® integrating sphere ensures high energy throughput and accuracy. A transmittance port is also included for making diffuse transmittance measurements. This DRA is fully CIE compliant for making color.



Solid Sample Holder

The Solid Sample Holder provides the capacity to measure a variety of solid samples in % Transmittance mode. A variety of sample holders are available allowing round, square, and odd-shaped samples to be measured. The industry standard 2" x 3" sample slides allow other accessories to be used with the Solid Sample Holder.

Smart Accessories

Smart Accessories are cell changers and sample holders that feature auto-recognition, smart alignment and serial number reporting, thereby saving you time and improving productivity. Smart Accessories are easily interchanged and ensure proper configuration of software methods.

Choose from a comprehensive selection for your analysis needs:

Smart Thermostatted Linear 8-Cell Changer — This is a fast, precise accessory for rapid measurement of up to 8 samples in manual, semiautomatic or fully automatic modes. It supports standard cuvettes, microcells, dissolution flowcells and solid samples of pathlengths from 10mm to 100mm. There is also a socket for connecting a temperature probe accessory, when purchased.

Smart Linear 16-Cell Changer — This is a fast, precise accessory for rapid measurement of up to 16 samples in manual, semiautomatic or fully automatic modes. It supports 4-channel cells and 4-channel flowcells.

Smart Thermostatted Rotary 7-Cell Changer — Consists of a carousel (also referred to as a cell holder) installed on a base unit. The base unit incorporates the drive motor, control circuitry and memory. There is also a socket for connecting a temperature probe accessory, when purchased.

Smart Peltier Thermostatted Single Cell Holder— For precise temperature control (e.g., DNA denaturation), this accessory provides temperature control from 5° to 110°C with ramping at 0.1° to 6°C per minute for a single 8.5mm cell and low head-space micro cells There is also a socket for connecting a temperature probe accessory, when purchased.

Smart Sipper — A multi-roller peristaltic pump that is mounted in the sample position of the sample compartment with Sip and Sip-and-run operating modes supported. Samples may be pumped to waste or returned. The sipper allows for calibration of tubing volume with minimum sample volume of 200µl with a single, standard cell holder. A flowcell is required.

Smart Calibration Validation Carousel (CVC) — Provides automatic testing of UV and visible wavelength accuracy, UV and visible absorbance accuracy, UV and visible stray light, resolution, noise and stability. Absorbance and wavelength filters are traceable to NPL or NIST via an ISO standard 17025 accredited calibration process. The CVC is supplied with a calibration certificate and shipping box for recalibration.

Mercury Lamp — Provides a means of wavelength calibration and verification in the UV and visible spectral ranges, using six fundamental mercury emission lines.

System accessories

Some system accessories are installed by Thermo Electron.

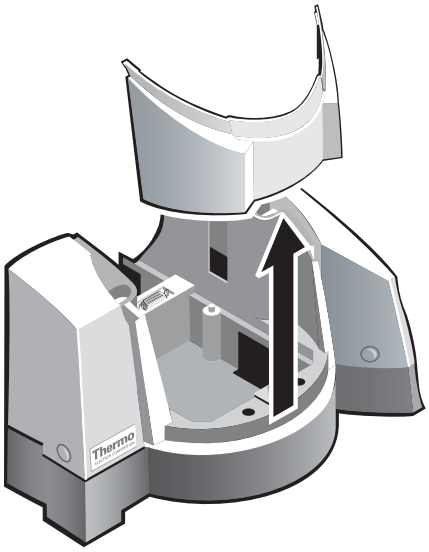
Internal printer — Located directly behind the LCD screen and is either factory or installed in the field by a qualified Thermo Electron service engineer.

Installing Smart Accessories

Positive location and automatic alignment of the accessories is achieved using pins A and B and the spring-loaded toggle as described for manual accessories. In addition, data communication is achieved via the serial D-connector underneath the front of the accessory, which plugs into the corresponding socket on the accessory. A tapered guide rod is provided beside the connector to assist in aligning the connector and socket during installation.

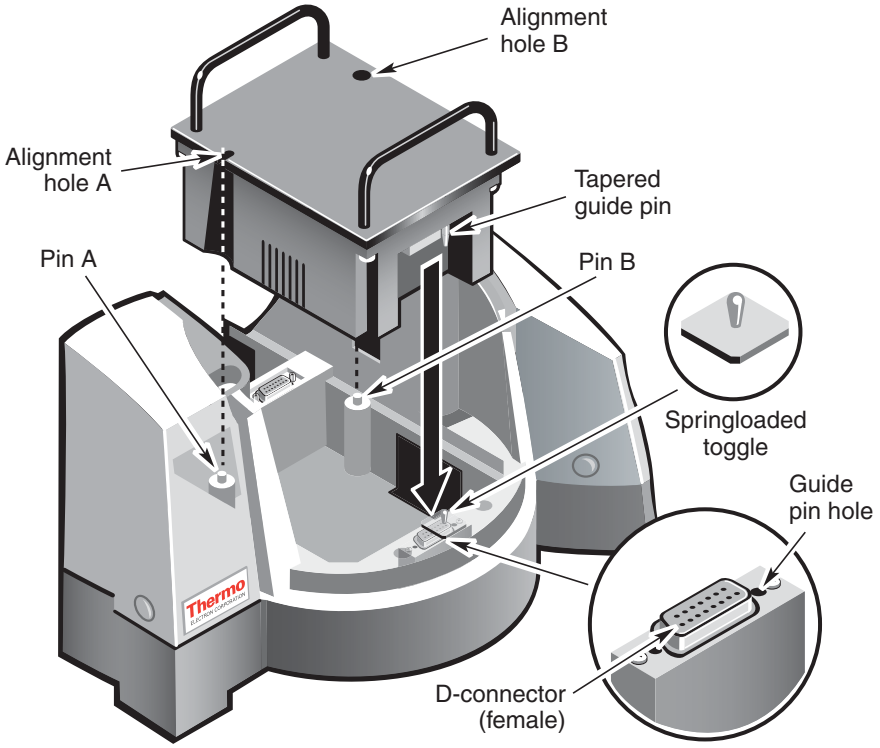
1. **Open the main lid of the sample compartment and remove any accessory that may be present in the sample (front) position.**

2. **Remove the front wall of the sample compartment by sliding it upward in its grooves.**



3. **Insert the tapered guide pin on the bottom front of the accessory into the corresponding guide pin hole located next to the D-connector (female). Do not press down to completely seat the accessory.**

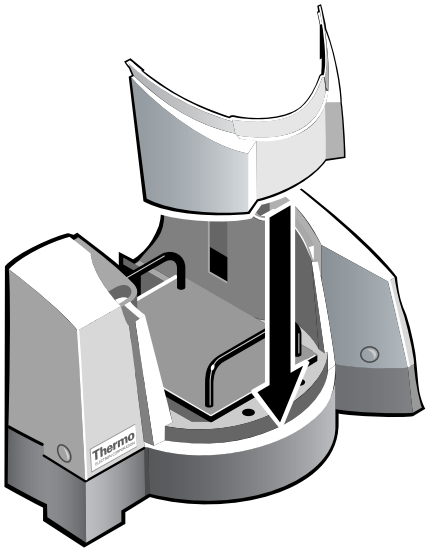
4. **Position the accessory so that alignment holes A and B are aligned with pins A and B on the instrument and press down gently.**



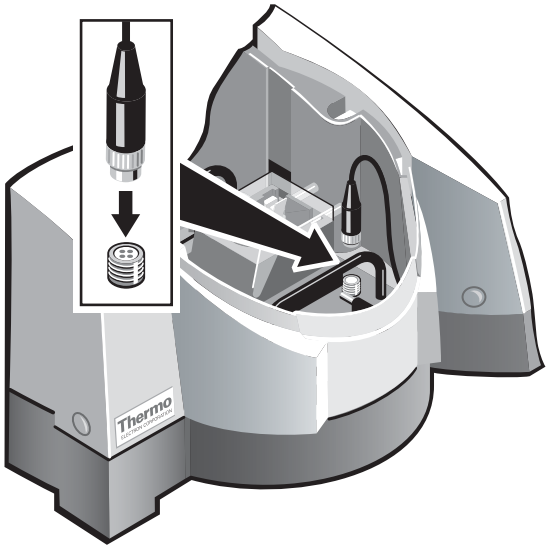
Note On the Smart Peltier Thermostatted Single Cell Holder, the alignment holes and pins are not visible due to the plastic cover over the unit. ▲

5. **Press down on the front of the unit to complete the connection between the accessory and the D-connector.**

6. **Replace the front wall of the sample compartment.**



7. **Connect the data cable (only for installation of the sipper or temperature probe).**



For removal of the accessory:

- Grasp the handles of the accessory and pull it upwards.

The software will display a message confirming that the accessory has been removed. Press Clear to remove this message.

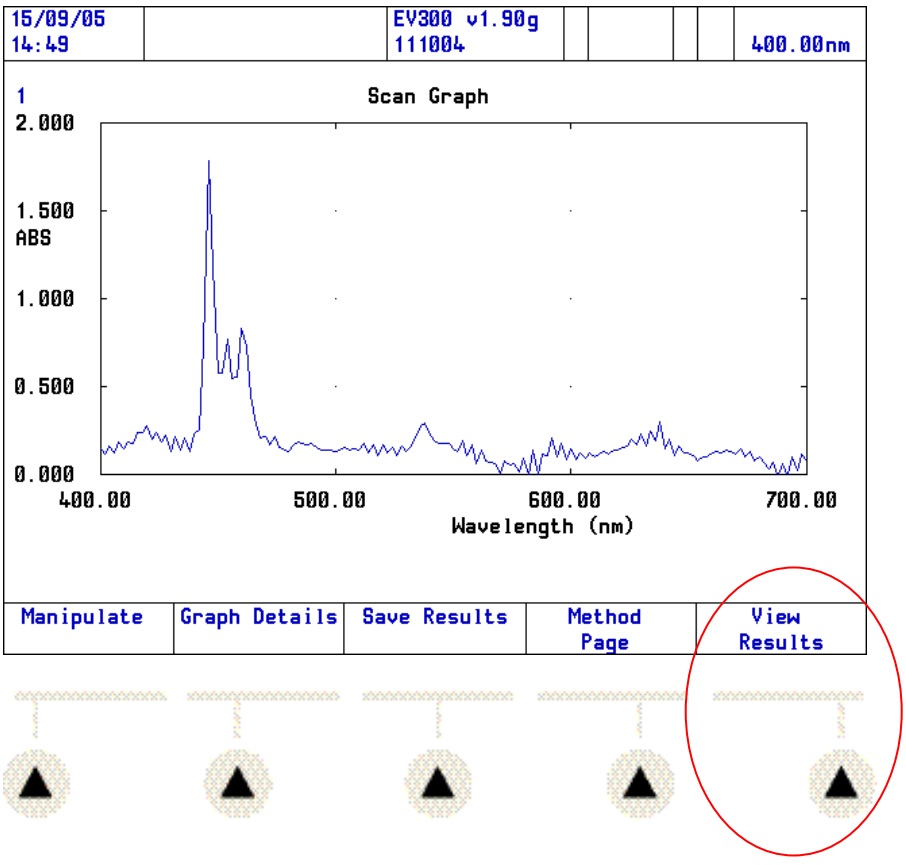
Chapter 4 – Using the Instrument

This chapter provides important information about using your instrument to analyze samples. The topics that are discussed include:

- Local Control software.
- VISION*security* and VISION*pro*.

Local Control

Function keys are operational in the context for each screen and are shown at the bottom of the display.



Buttons on the instrument panel:

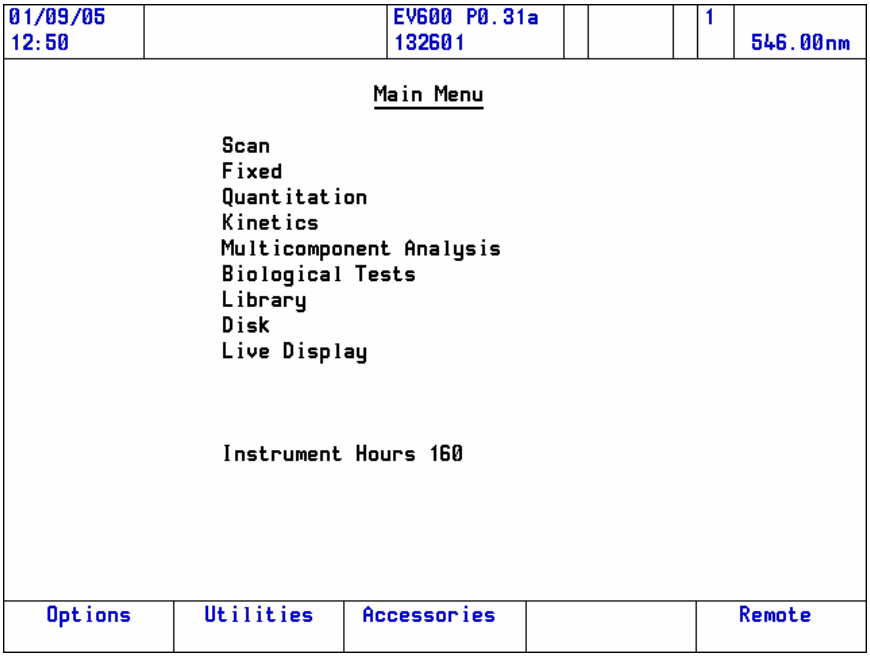
To do this….	Use ….	
Print the current screen contents to the selected printer or disk.		**Print**
Return to the Main Menu or the SmartStart™ menu, if SmartStart is enabled.		**Home**
Perform a zero measurement or baseline scan without additional prompts.		**Zero**
Perform the measurements specified in the current method.		**Run**
Display the contents of the instrument library		**Library**
Clear error message or cancel pop-up menus.		**Clear**
Confirm highlighted choices in menus and text entry boxes.		**Enter**
Select menu items and characters in text entry screens.		**Arrow keys**
Enter numbers, decimal point and minus sign.		**Numeric keypad**

Buttons to the left of the display:

To do this…	Use…	
Increase or decrease brightness of display.		**Bright**
Feed paper for internal printer (if installed).		**Feed**

Main Menu page

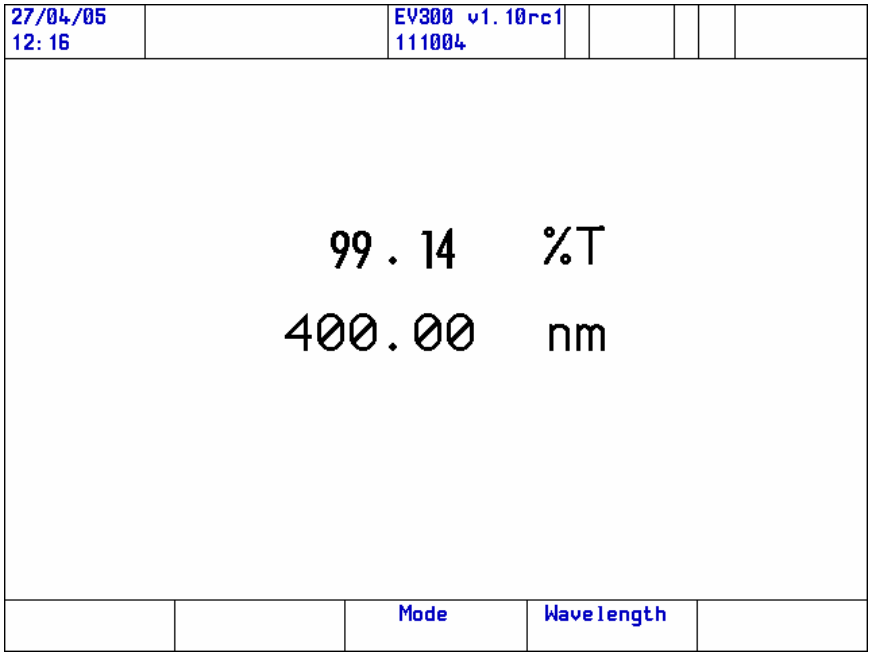
The major functions of the software are accessed from the Main Menu page.



Use the up and down arrow keys to highlight the required menu item and press Enter to confirm your selection.

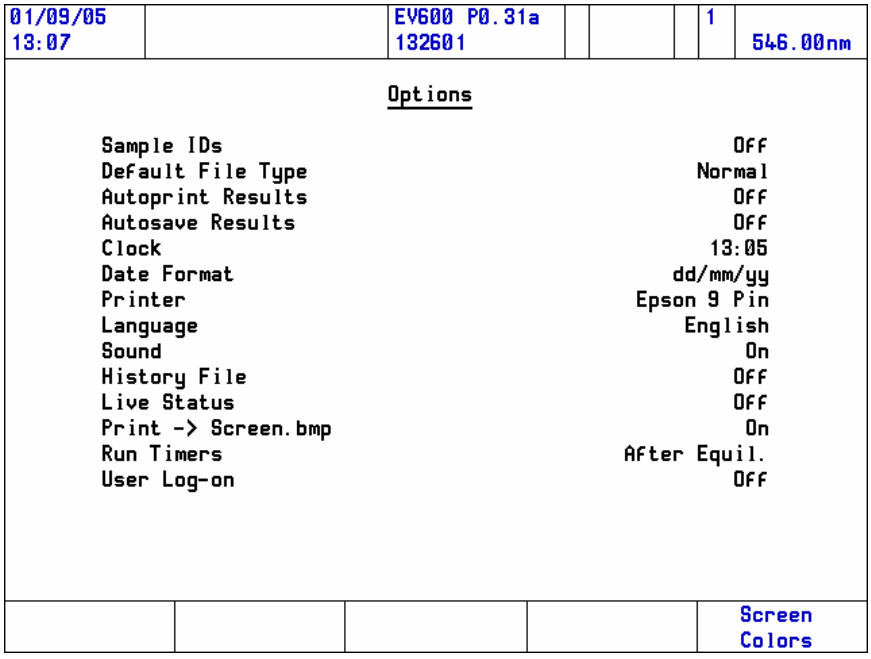
The menu enables you to move between the six major applications and to view the contents of the Library or the contents of the disk currently in the drive.

Here is an example of a live status display:



Options window

To access the Options window from the Main Menu, press the Options function key.



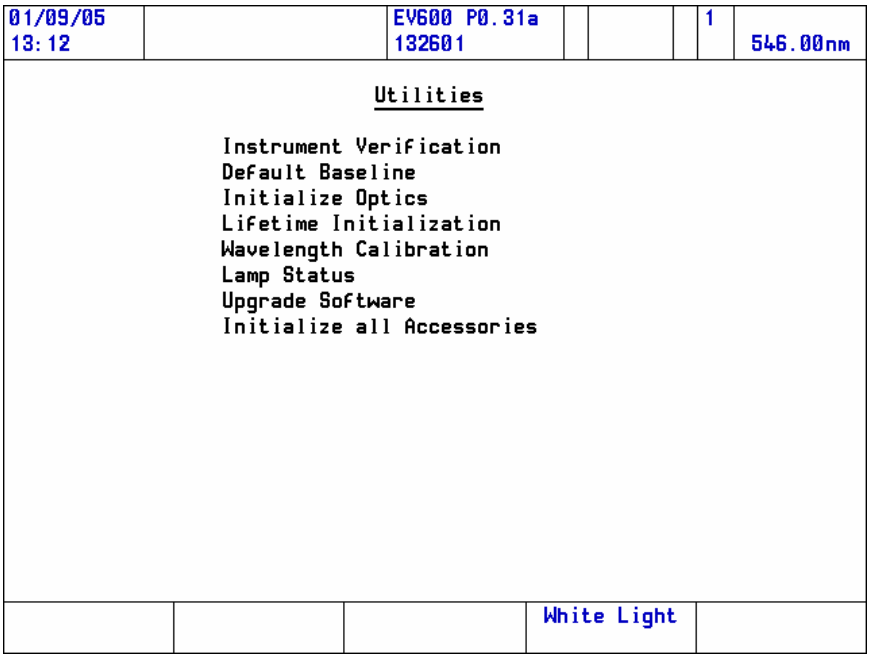
The primary use of each parameter is summarized in the following table:

Parameter	Function
Sample IDs	When toggled on, the user enters a name seed and start number for the results files. The number is automatically incremented for each sample.
Default File Type	Allows the user to select the file type of the saved data. Normal is the native file type of the software and can be imported into the VISION software package. This file type can be used only by the Local Control or the VISION***pro***/VISION***security*** software. The comma separated values (CSV) format enables data to be exported to computer third-party applications (e.g., spreadsheet).
Autoprint Results	Toggled On – test setup, data and results are automatically printed at the end of each run. This option uses the currently selected printer. Toggled Off – no automatic printout. User must Print to obtain a printout if desired.
AutoSave Results	Toggled On – test results are automatically saved after each experiment in a user-named file stored in either the internal instrument memory or on a disk. When On, the user must enter a name deed and start number. Toggled Off – data is not automatically saved. User must select Save Data for each set of data if they want to save the data.
Clock	Allows user to set the time and date.
Date Format	Selects the desired date format.
Printer	Selects the active printer.
Language	Selects the appropriate display language.
Sound	Toggled On – any error is indicated by an on-screen message and audible beep. Toggled Off – on-screen messages are the only indication of an error.

Parameter	Function
History File	Toggles On – significant events (e.g., recalibration) are automatically recorded in the History File. Toggled Off – no events can be recorded in the History File.
Live Status	Toggled On – the display of the live reading at the top right-hand corner of the screen includes current wavelength and data. Toggled Off – the display of the live reading at the top right-hand corner of the screen includes only the current wavelength.
Print -> Screen.bmp	Toggled On – allows the user to create a bitmap of the current screen contents to be saved to a disk when the Print button is pressed. Toggled Off – printing function is directed to the installed printer.
Run Timers	Enable up to four countdown timers to be incorporated into the method sequence.
User Log-on	Toggled On – a password is required to access the instrument. Toggled Off – no password is required to access the instrument.
Screen Colors	Allows the user to select the colors used in the display.

Utilities window

To access the Utilities window, press the Utilities function key. Highlight and press Enter to select a menu item.



The primary use of each parameter is summarized in the following table:

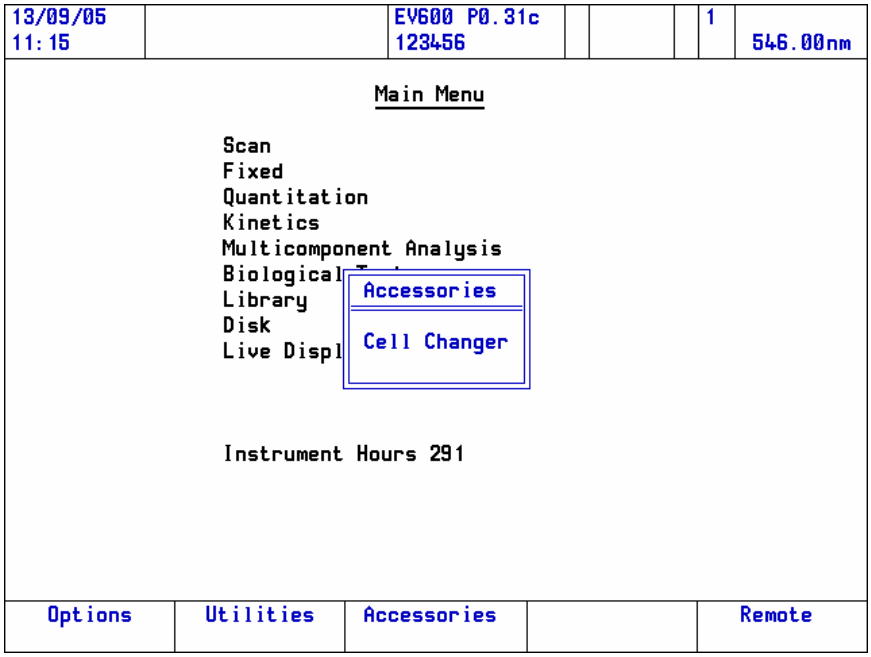
Parameter	Function
Instrument Verification	Accesses the menu for instrument verification tests.
Default Baseline	Re-scans the stored default baseline. Ensure that a new default baseline is really needed before selecting this option.
Initialize Optics	Re-initializes optics without switching the instrument off, then on.
Lifetime Initialization	Performs a lifetime initialization. (Ensure that a lifetime initialization is really needed before selecting this option.)
Wavelength Calibration	Performs a wavelength calibration using the optional mercury lamp accessory. Ensure that a wavelength calibration is really needed before selecting this option.
Lamp Status	Shows current status of the installed lamps.

Parameter	Function
Upgrade Software	Upgrades instrument software from disk.
Initialize all Accessories	Initializes installed accessories.
White Light	Causes a beam of white light to pass through the sample compartment. (Used to align accessory.)

Smart Accessories

To access the accessory controls, press the Accessories function key on the Main Menu page.

A pop-up menu shows the accessories currently installed. Here is an example:

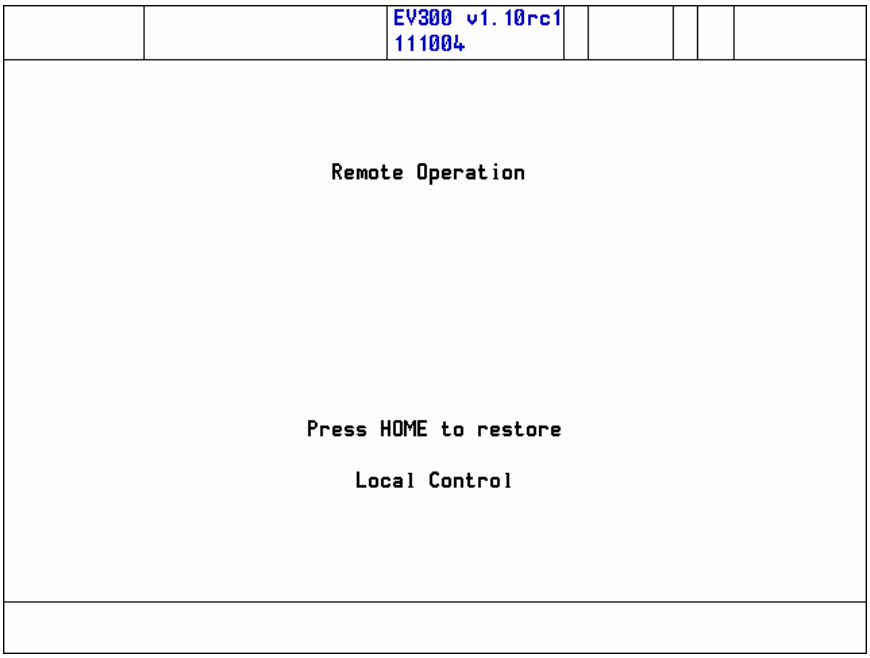


Highlight the required accessory and press Enter to open the setup page.

When accessories are installed, additional items related to the control of the accessories are added to the method parameter pages.

Remote

When the Remote function key is selected, Local Control software passes control of the instrument to VISION software, communicating with the spectrophotometer via the RS232 interface.



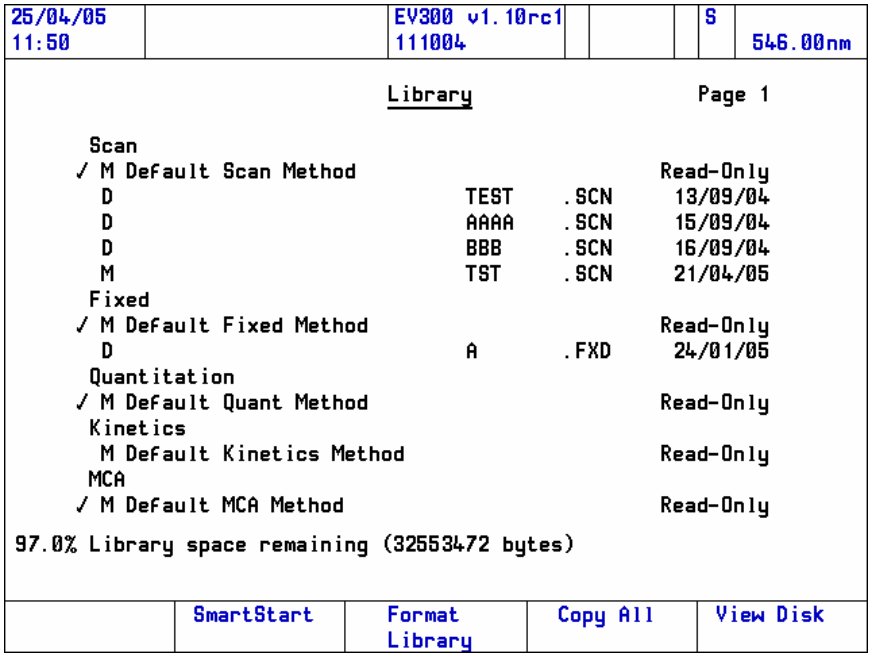
To return to Local Control in VISION, select Command > Local Control and then press Home on the spectrophotometer.

SmartStart

The instrument can be configured so that frequently used methods are displayed on the SmartStart menu. This then replaces the Main Menu as the Home page, enabling common methods to be accessed conveniently from the start-up screen.

How to Select files to be included in the SmartStart menu:

1. **Press Library.**
2. **Highlight a file to be included in the SmartStart menu.**
3. **Press the SmartStart function key.**

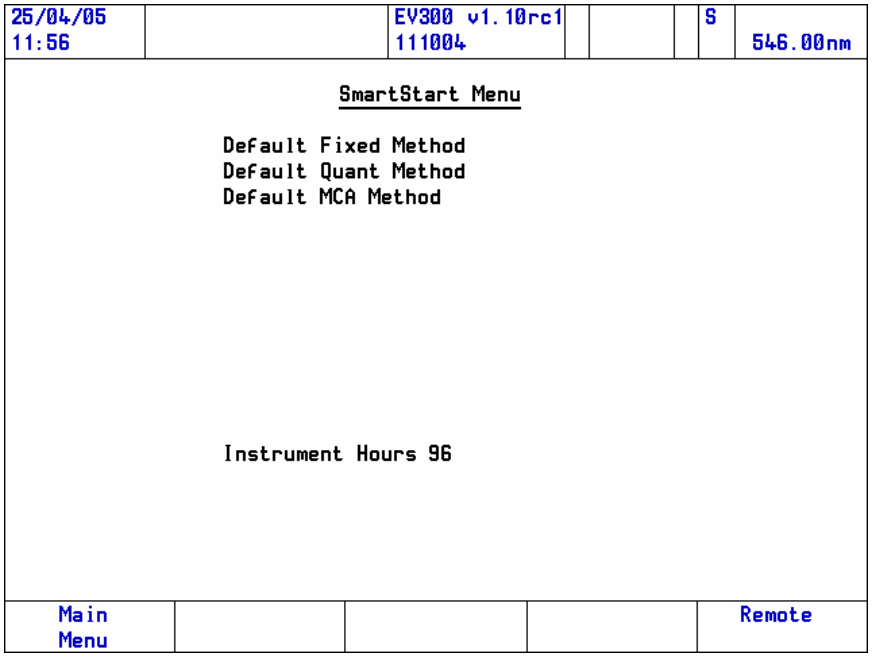


A checkmark appears beside the file after the SmartStart function key has been pressed.

Continue in this way until all required files have been chosen for the SmartStart menu.

4. **Press Home.**

All of the selected items appear on the SmartStart Menu page.



Note When SmartStart is enabled, the Main Menu is accessed by selecting the Main Menu function key at the lower left-hand corner of the SmartStart page. ▲

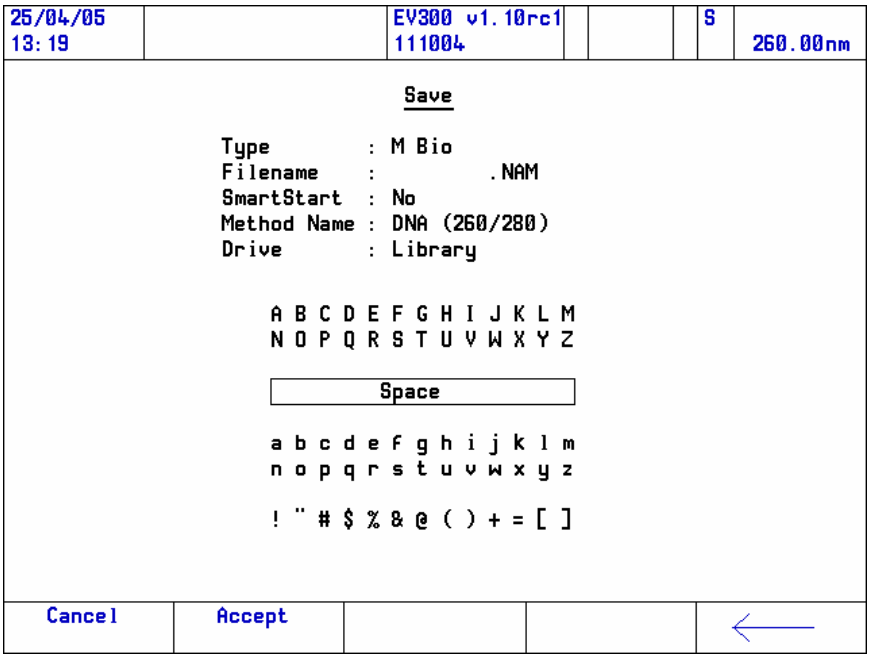
To remove a file from the SmartStart menu:

- Highlight the checked file(s) in the Library.
- Press SmartStart.

Text entry

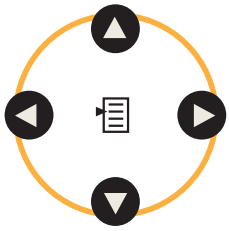
Text items are entered using the text entry screen.

Here is an example of a text entry screen:



How to

Navigate in the text entry screen:



Use the arrow keys to move left/right and up/down to highlight the required character.

To facilitate navigation, the key action will 'wrap' so that, for example, pressing the left arrow from A will move the highlight to M.

1. **Press Enter to move the selected character into the edit field.**

   Continue until all the required characters are in place.

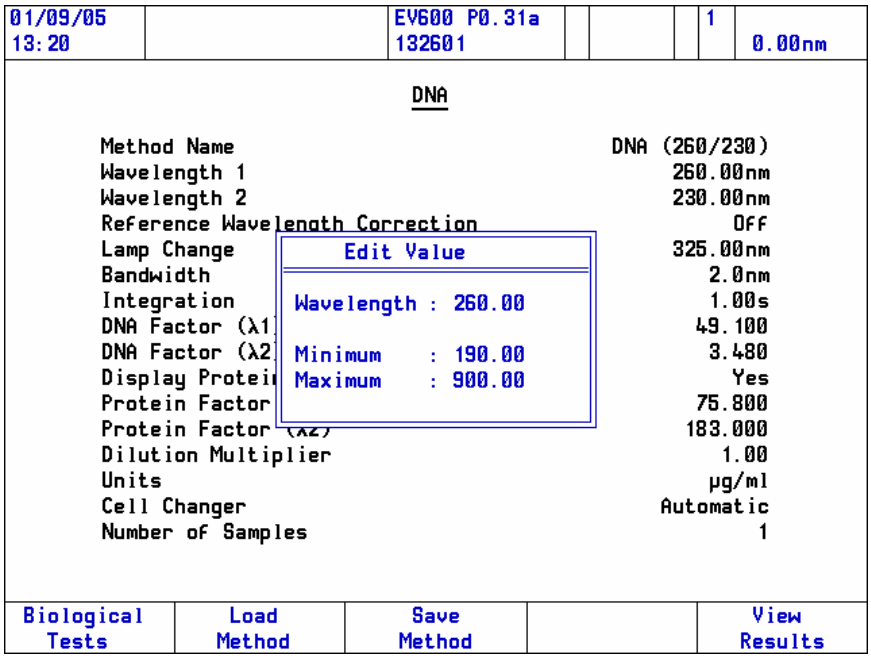
2. **Press the Accept function key.**

Note Pressing the Cancel function key will close the text entry screen without making any changes. ▲

Note The backspace function key may be used to erase mistakes. ▲

Editing numeric fields

To edit the value of a numeric field, highlight the item.



An Edit Value box appears. Key in numbers with the numeric keypad, taking note of any information such as limits displayed in the Edit Value box.

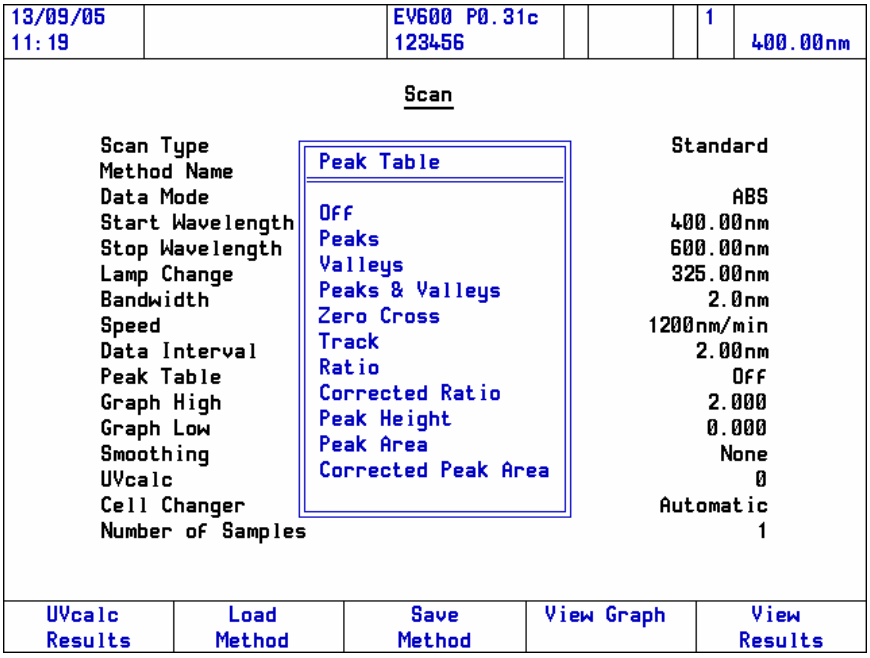
Press Enter when the numeric entry is complete, or Cancel to terminate numeric entry to close the Edit Value box without making any changes.

When setting up a page requiring multiple numeric entries (e.g., a Standards page), press the Accept function key when the page is complete or C to close the page without making any changes.

Pop-up menus

Use the up/down arrow keys to move the highlight to your menu choice and then press Enter to confirm.

Here is an example of a pop-up box:



File operations

You can store files in the instrument Library or on a floppy disk. The contents of the Library can be viewed, accessed and maintained at any time by pressing the Library button on the instrument keypad.

How to

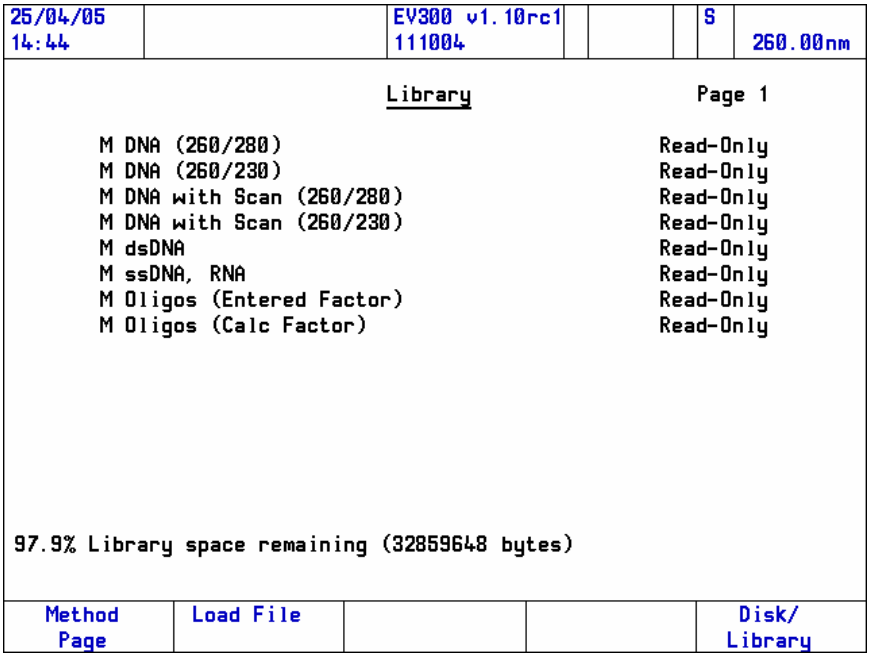
Access the Library or disk from the Main Menu:

1. **Highlight the required field using the up/down arrow keys and press Enter.**
2. **Choose the required operation from the menu.**
3. **Press Enter.**

   Load Method and Save Method function keys are available on application parameter pages. Save Results function keys are available on all Results pages.

Note When a Load Function key is selected, the Library page is disabled showing the files that are appropriate to the current context. ▲

4. **Press the Disk/Library function key to display the contents of the disk, if required.**



5. **Highlight the required file by using the up/down arrow keys and then press Load File function key.**

How to

Save a method:

When a Save Method function key is selected, the Save dialog opens with the Filename highlighted ready for entry.

Parameter	Function
Filename	Allows the user to enter a file name. When filename has been entered, press the Accept function key.*
SmartStart	Allows the user to place this method in the SmartStart menu when the SmartStart menu is set as the home page.
Method Name	Allows the user to enter method name (optional).
Drive	Allows the user to select whether the method is saved to disk or instrument Library.

* Press the Save function key (available after a filename has been entered) to save, or Clear to exit without saving.

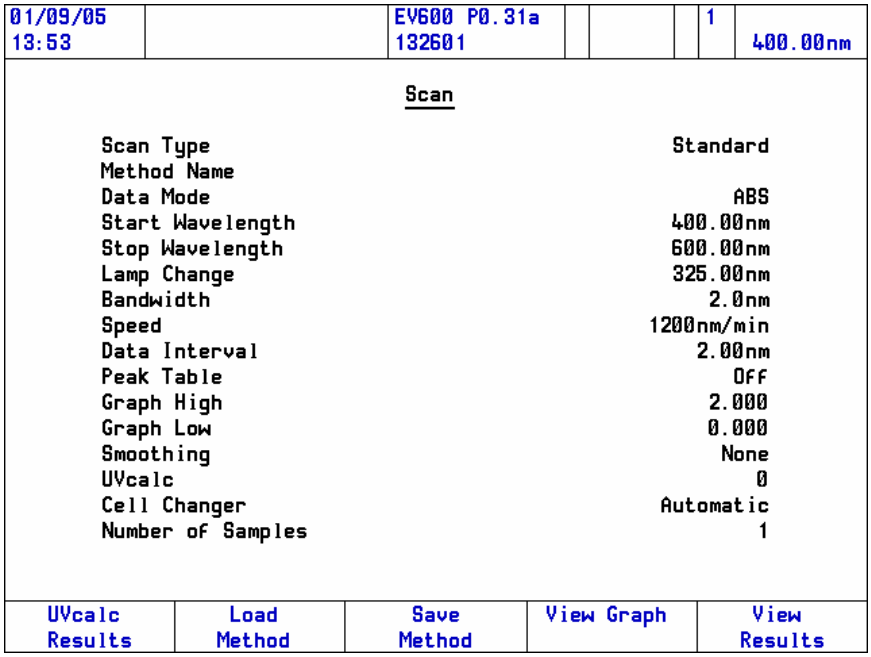
Scan application

Follow these steps to run a Scan application:

1. **Start the Scan application.**

   To do this, highlight Scan on the Main Menu page and press Enter.

   The Scan method page will open. The menu may contain more items than are shown in the example, depending on the way the software is configured and which accessories are installed.



2. **Set up Method.**

Parameter	Function
Scan Type	Toggles between Standard and Intelliscan. • When Standard is selected, the user is allowed to enter a scan speed. • When Intelliscan is selected, the Speed item is used to select the Intelliscan mode, which also determines the data interval.
Peak Table	Sets up a Peak Table for peak operations that will be performed at run time.*
Graph High	Enter maximum values for the graph y-axis.*
Graph Low	Enter minimum values for the graph y-axis.*

Parameter	Function
Smoothing	Selects the degree of smoothing required at run time.*
UV*calc*	Refer to "Using UV*calc*".

*Alternatively, these operations are all available as post-run manipulations.

3. **Set up any Accessory Methods required (only when a Smart Accessory is installed).**

4. **Save method.**

   To save the method to the instrument Library or disk, press the Save Method function key and follow the on-screen instructions.

5. **Load a method from the instrument Library or from disk.**

   To do this, press the Load Method function key or press Library.

6. **Remove any unwanted samples from sample compartment.**

7. **Insert a suitable reference, if required, and close the lid of the sample compartment.**

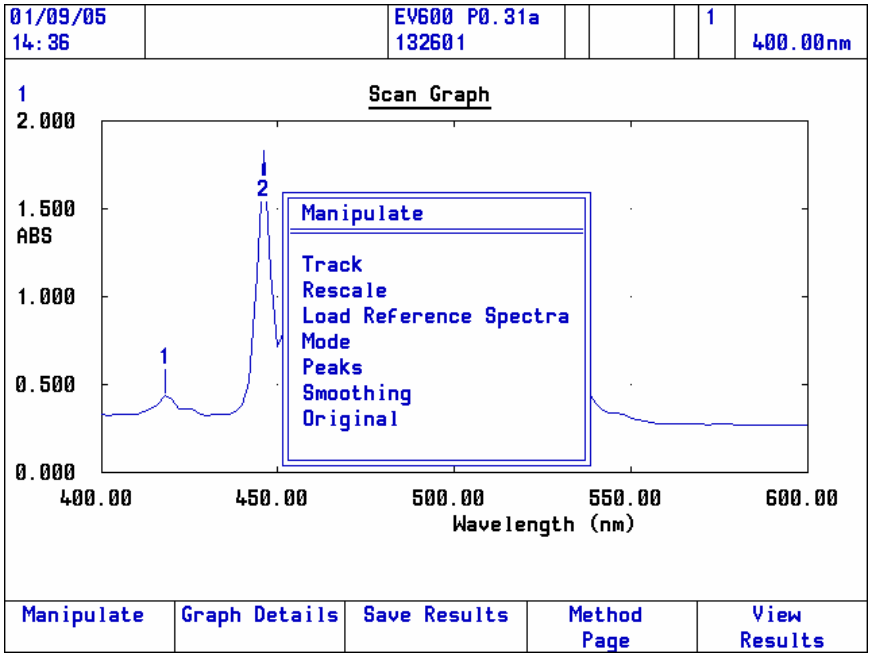
8. **Press Zero to measure a baseline.**

9. **Open lid of the sample compartment and insert the sample and then close the lid.**

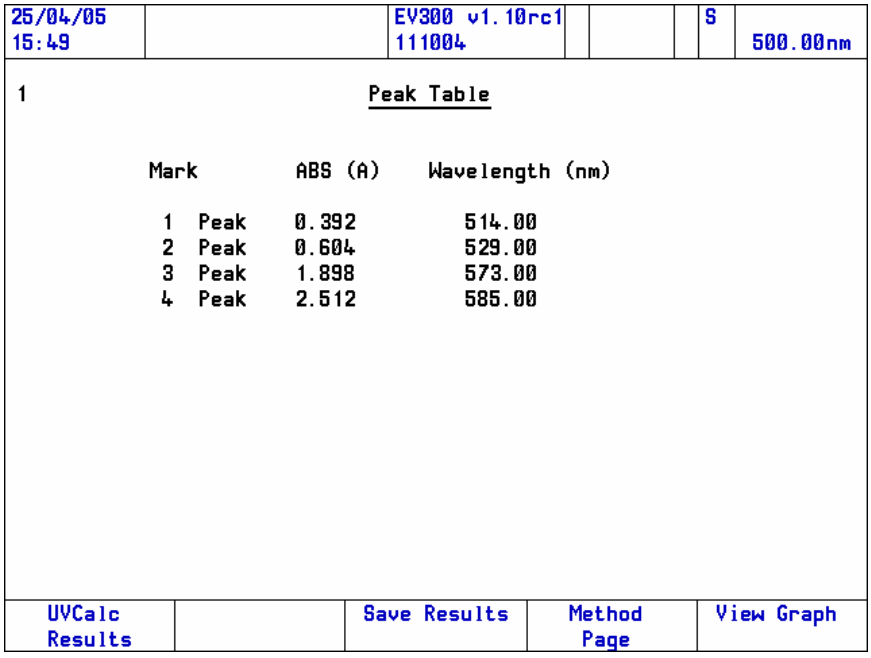
10. **Press Run.**

    The Scan Graph page will appear.

11. Press the Manipulate function key to open the Manipulate menu and choose the post-run manipulation required.



For example, press the View Results function key to see the results of Peak Table operations or post-run manipulations.



12. Save results to the instrument Library or disk.

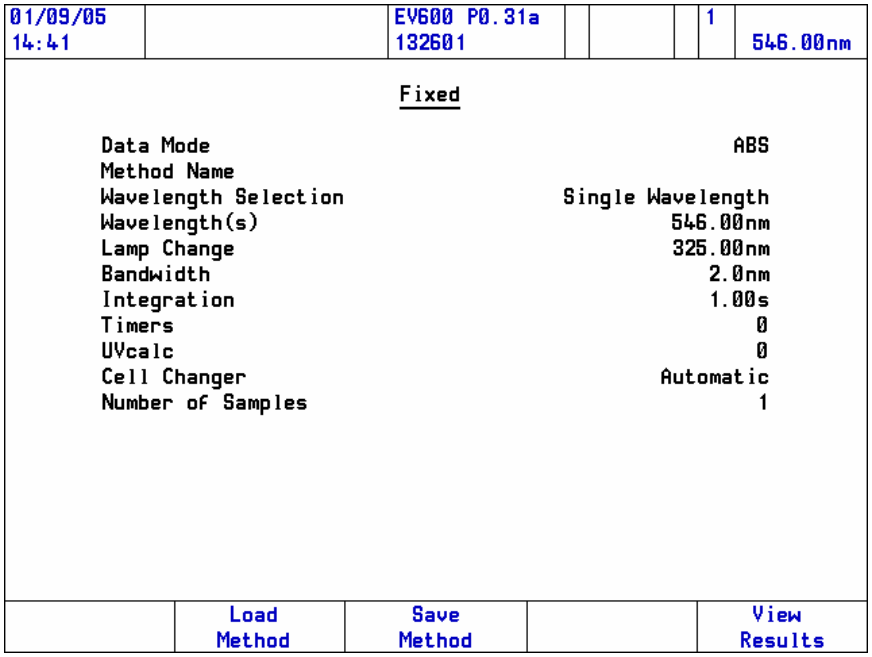
To do this, press the Save Results function key on the Scan Graph page.

Fixed application

Follow these steps to run a Fixed application:

1. Highlight Fixed on the Main Menu page and press Enter.

The Fixed Method page will open. The menu may contain more items than are shown in the example, depending on the way the software is configured and which accessories are installed.



2. Highlight each parameter in turn and press Enter.

Parameter	Function.
Data Mode	Toggles between %T and ABS.
Method Name	Allows user to enter a method name.
Wavelength Selection*	Allows the user to select either Single, Multiple, or Serial Wavelength measurements.
Wavelength(s)	Sets the wavelength(s) where the measurement(s) is/are made.

Parameter	Function.
Lamp Change	Allows the user to select between deuterium, automated (instrument uses either the deuterium or tungsten lamp to run the selected wavelength) and tungsten lamps.
Bandwidth	Sets spectral bandwidth.
Integration	Allows the user to determine the integration (measurement) time.
Timers	Enables up to four countdown timers to be incorporated into the method sequence. A timer title allows the user to specify the action (e.g., shake, boil) associated with the timer. At the end of the timer period, the program may continue to the next step or pause to await action. Timers may be multiple use or single use.
UV*calc*	Refer to "Using UV*calc*".

***Wavelength Selection** — Use the table below to help you choose between the three different wavelength options:

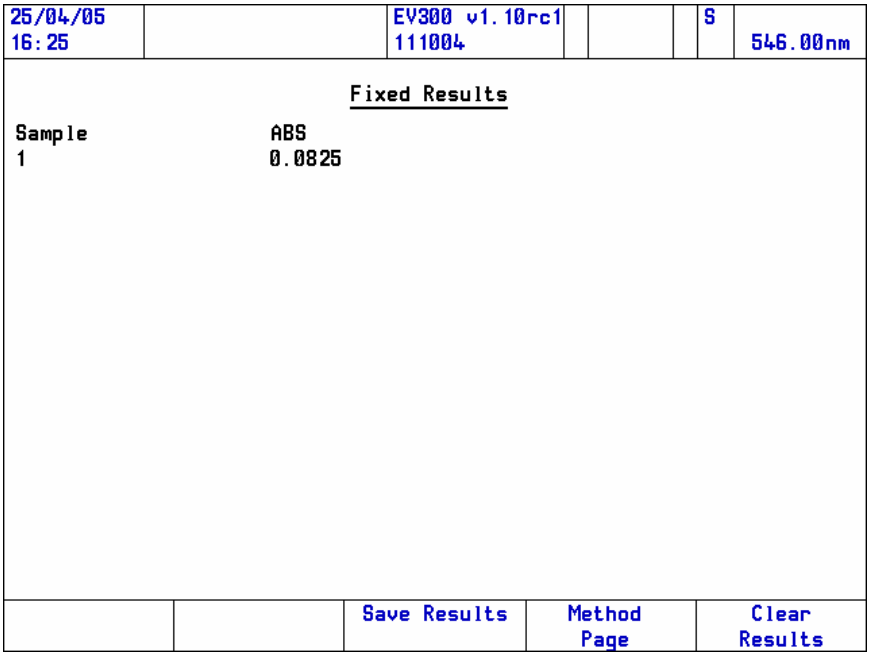
Select this wavelength…	To do this…..
Single Wavelength	Measure each sample at one wavelength (the same for each sample).
Multi Wavelength	Measure each sample at up to 20 wavelengths (the same for each sample).
Serial Wavelength	Measure up to 9 samples at one wavelength (different for each sample).

3. Load a method from the instrument Library or from disk.

Press the Load Method function key and follow the on-screen instructions. To save the method to the Library or disk, press the Save Method function key and follow the on-screen instructions.

4. Clear the beams or insert suitable references and close the lid of sample compartment.

5. **Press Zero to zero the instrument.**
6. **Open sample compartment lid and insert the samples.**
7. **Close the lid of the sample compartment and press Run.**
8. **Press the View Results function key to see the Fixed Results Table.**

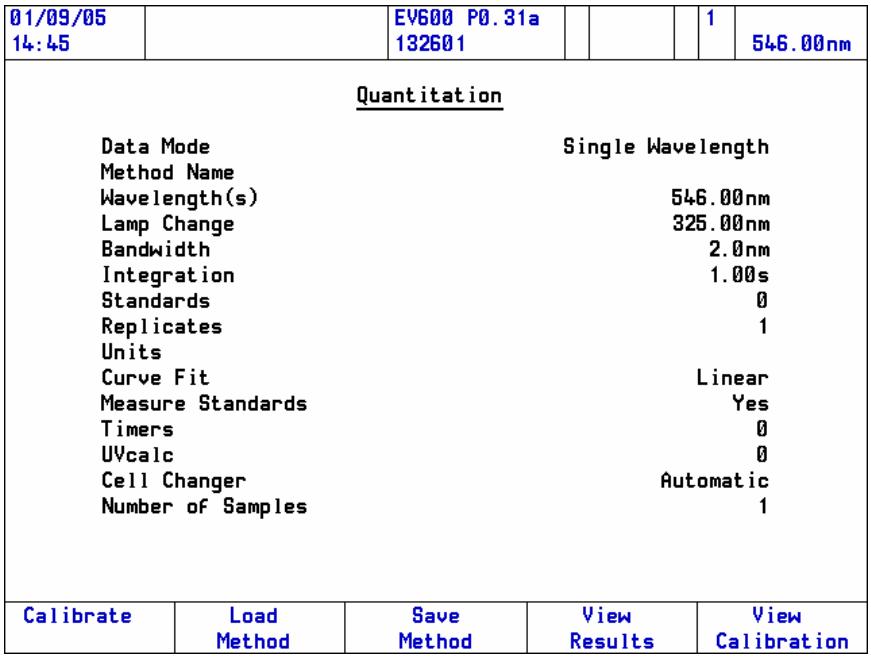


From this page, it is possible to save the results to the instrument Library or to disk.

Quantitation application

Follow these steps to run a Quant application:

1. **Start the Quantitation application and press Enter.**



   The Quantitation method page will open.

   The menu may contain more items than are shown in the example, depending on the way the software is configured and which accessories are installed.

2. **Set up the method and press Enter.**

   Highlight each parameter in turn. This will cause the item to toggle between two values or open a menu, or open a numeric entry box.

   - Seven different data modes are available.
   - The number of wavelengths required will depend on the Data Mode.
   - Highlight Standards and press Enter to open the Standards Entry page. Enter the concentration of each standard. Press the Accept function key when all standards have been entered.
   - There may be 1, 2 or 3 replicates per standard.

- Normally, Measure Standards is set to Yes. When set to No, absorbance values may be entered manually.

- The minimum number of standards required will depend on the curve fit.

Linear:	2
Linear through 0:	1
Quadratic:	3
Quadratic through 0:	2

  Normally, a calibration contains more than the minimum number of standards.

- Timers and UV*calc* equations can be used, if required.

3. **Press the Load Method function key to load a previously saved method from the instrument Library or from disk.**

4. **Clear the beams or insert suitable blank(s) and close the lid of the sample compartment.**

5. **Press Zero to zero the instrument.**

6. **Press the Calibrate function key.**

   Follow the on-screen instructions, which will depend on whether a cell programmer is in use and on the number of standards and replicates.

7. **View the graph and statistics.**

   This is done when the calibration is complete. Change the curve fit if necessary.

8. **Press the Save Method function key to save the calibrated method in the instrument Library or to disk.**

9. **Press Run to analyze samples using the calibrated Quantitation method.**

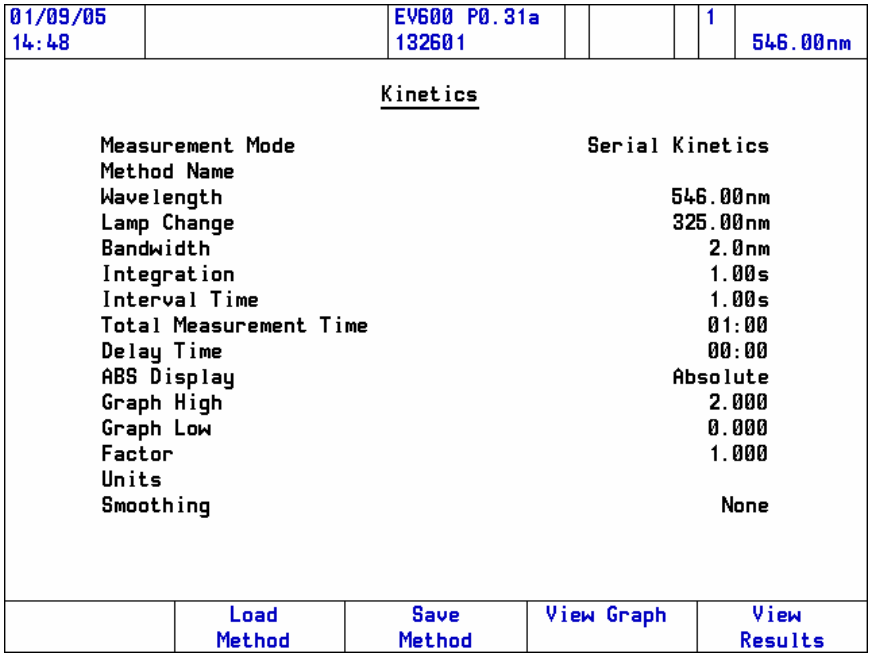
10. Press the View Results function key to see the results table.

Function keys on the Quant Results page enable you to save the results to the instrument Library or to disk, or to clear the results table ready for a new batch of samples.

Kinetics application

Follow these steps to run a Kinetics application:

1. Start the Kinetics application and press Enter.



The Kinetics Method page will open.

The menu may contain more items than are shown in the example, depending on the way the software is configured and which accessories are installed.

Note The layout and composition of the Kinetics Method page will depend on the Measurement Mode selected. ▲

Parameter	Description
Serial Kinetics	Each sample is measured at the specified wavelength for the specified Total Measurement Time.
Multi-wavelength	The absorbance of each sample is measured at two wavelengths for the specified Total Measurement Time. The Cycle Time specifies the interval between measurements at the same wavelength.
Difference	The absorbance of each sample is measured at two wavelengths for the specified Total Measurement Time and the difference calculated. The Cycle Time is the interval between measurements at the same wavelength.
Parallel Kinetics	Only available with cell changer. The absorbance of each sample is measured in turn for the specified number of cycles. The Cycle Time is the interval between measurements on the same cell. Total Measurement Time is calculated from the number of cycles and the cycle time, and is not editable in Parallel Kinetics mode.

2. Set up the Method and press Enter.

Highlight each parameter in turn. This will cause the item to toggle between two values, or open a menu, or open a numeric entry box.

	Interval time (s)	Integration time (s)
Evolution 300	**0.02 (min)** **999.99 (max)**	**0.01 (min)** **999.99 (max)**
Evolution 600	**0.13 (min)** **999.99 (max)**	**0.03 (min)** **999.99 (max)**

- Enter one wavelength in Serial or Parallel mode. Multi-wavelength or Difference modes require two wavelengths. Highlight the wavelength line and press Enter. Key in the required wavelength and press Enter.

- Enter the required Total Measurement Time in Serial, Multi-wavelength and Difference modes. In Parallel mode, the Total Measurement Time is displayed but cannot be edited.

- In Multi-wavelength, Difference and Parallel modes, enter the cycle time.

- If a delay is required between the start of the run and the first measurement, enter a delay time.

- Choose whether to display the absolute values of the absorbance, or the change in the absorbance relative to the initial measurement. If Relative absorbance is chosen, an additional menu item appears to set positive or negative slope.

- Set up Graph High and Graph Low to accommodate the expected absorbance range. The graph can be reconfigured post-run, if required.

- Enter a factor and units, if required.

- Smoothing may be specified at run time or applied post-run.

- Save the new method into the instrument Library or to disk. Press the Save Method function key and complete the save dialog as required.

3. **Load a previously saved method from the instrument Library or disk.**

   Press the Load Method function key, select the required file and load it.

4. **Clear the beams or insert appropriate blank(s) and close the lid of the sample compartment.**

5. **Press Zero.**

6. **Load the cell holder, initiate the reaction(s) as necessary and close the lid of the sample compartment.**

7. **Press Run.**

The method will be executed and the absorbance(s) of the sample(s) will be plotted on the Rate Graph.

When the Pause function key is selected, data collection will cease but the clock will continue to run. When the Pause function key is selected again, data collection will resume. There will be a gap in the Kinetics Graph for the duration of each pause. In Parallel Rate, the pause will not take effect until the current cycle of measurements has been completed.

8. **Press the Manipulate function key to access the Manipulate menu.**

9. **Rescale, smooth or change the absorbance display, if required.**

   Select Original to return to the unmanipulated data.

10. **Set up the rate calculations.**

    Section allows up to 5 markers to be placed on the graph defining up to 4 contiguous time intervals over which the rate statistics will be calculated.

    When the markers have been placed, press the function key to exit from the Section Mode.

11. **Press the View Results function key to see the results of the rate calculations performed using Section.**

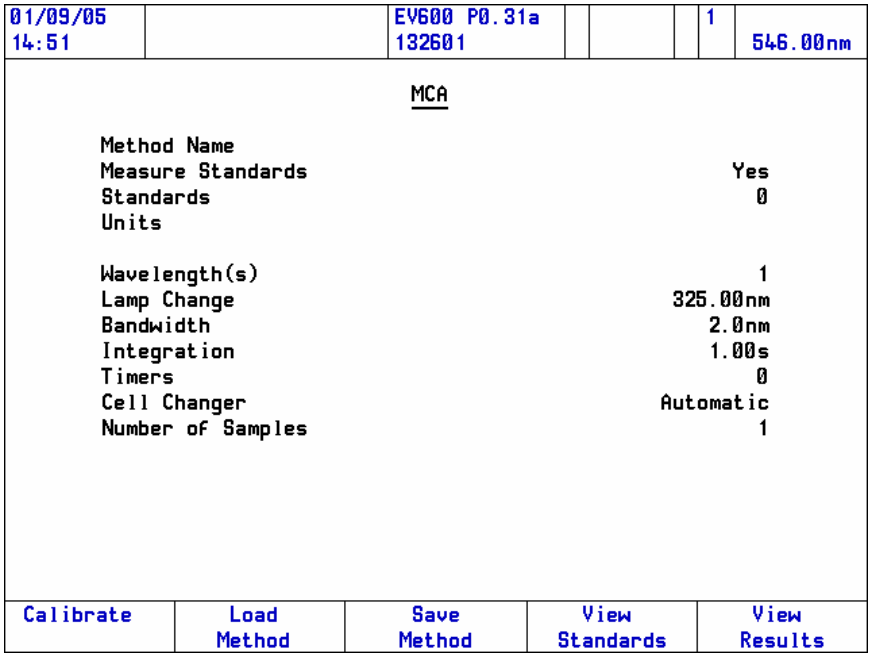
    Function keys on the Results page enable you to save the results to the instrument Library or to disk.

12. **Press the Save Results function key to save the results to the instrument Library or to disk.**

Multicomponent application

Follow these steps to run a Multicomponent (MCA) application:

1. **Start Multicomponent Analysis and press Enter.**



The MCA method page will open.

The menu may contain more items than are shown in the example, depending on the way the software is configured and which accessories are installed.

2. **Set up method parameters.**

Parameter	Function
Measure Standards	Use Enter to toggle between Yes and No. • **Yes:** Enter the Ids of the standards and concentrations into the Standards Table. After pressing the Calibrate function key, the user will be prompted to measure each standard. • **No:** Highlight the first standard ID in the Standard Table and press Enter. The Library page will open showing available .FXD results files. Highlight the required Fixed multiwavelength result file and press the Load File function key. The method parameters will be set to those used to measure the first standard and cannot be changed. All subsequent standards must have been measured using exactly the same method. Each file must contain data for a single sample. Use Clear Results between Fixed measurements.
Standards	Transfers to the MCA standards page. • When Measure Standards = Yes, enter an ID and concentration for each standard. • When Measure Standards = No, load Standards as described above and enter a concentration for each standard.
Wavelengths	• When Measure Standards = Yes, enter the wavelengths at which standards and samples will be measured. Wavelengths may be entered from a scan. • When Measure Standards = No, the wavelengths used are defined by the standard method.

Note At least one wavelength must be entered for each standard. Choose wavelengths at which there are significant differences between standards. ▲

3. **Clear the beams or insert suitable blank(s) and close the lid of the sample compartment.**

4. **Press Zero.**

5. **Press the Calibrate function key.**

   When Measure Standards = Yes, you will be prompted to measure each standard in turn.

   When Measure Standards = No, the calibration will be calculated from the standards that have been entered.

6. **Save the calibrated method to the instrument Library or to disk.**

   A previously calibrated method can be loaded from the instrument Library or from disk.

   When a valid calibration is present, pressing Run causes the absorbance of the sample to be measured at each of the wavelengths specified in the method. The concentration of each component is then calculated from the calibration.

7. **Press the View Results function key to transfer to the MCA Results page.**

   The up/down arrow keys can be used to view previous/following results.

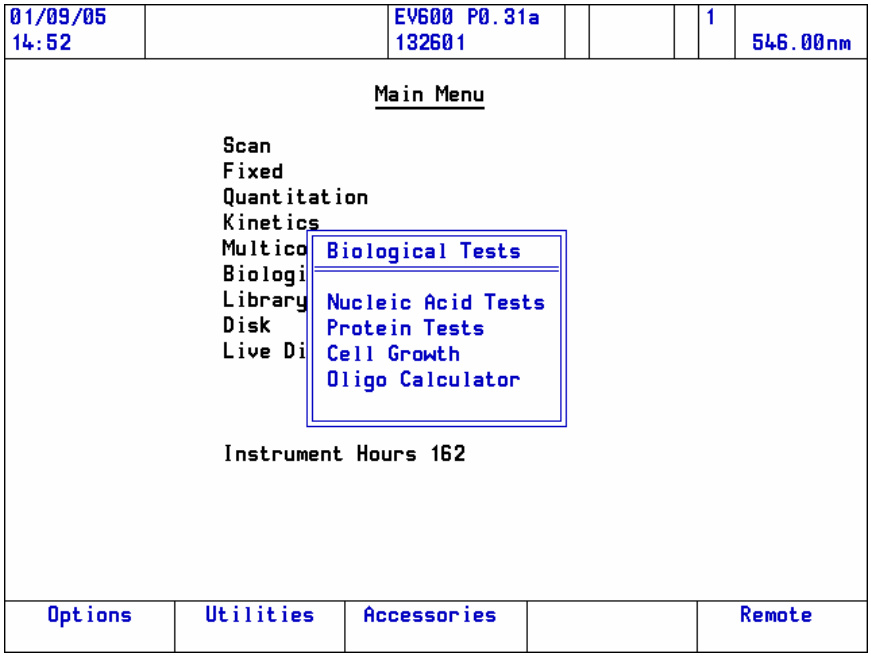
8. **Press the Save Results function key to save results to the instrument Library or to disk.**

Biological tests

Follow these steps to run Biological tests:

The menu enables you to move between the four major applications.

To run any of the biological tests available, highlight Biological Tests on the Main Menu and press Enter.

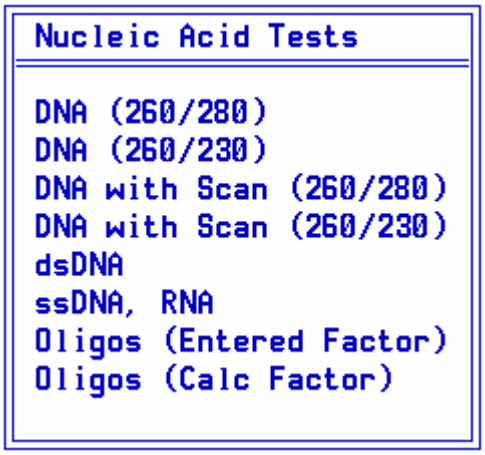


How to

Run a Nucleic Acid test:

1. **Choose Nucleic Acid Tests and press Enter.**

2. **Highlight the Nucleic Acid Tests and press Enter.**



   Choose the required test from the Nucleic Acid Tests sub-menu.

   The relevant test page opens. The content and layout of the page will depend on the test selected, the way the software is configured and which accessories are installed.

3. **Set up method parameters.**

The wavelength and factor values contained in the method are recognized values established in the literature. Values may be edited, if required.

4. **Highlight Reference Wavelength Correction and press Enter to toggle Off/On.**

   When On, a Reference Wavelength is added to the parameter list.

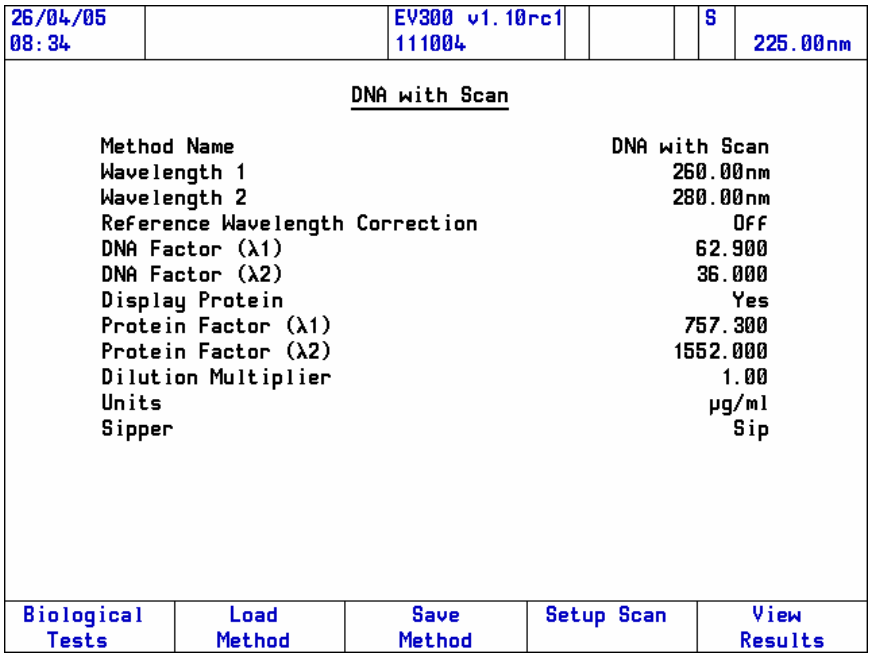
5. **Highlight Display Protein and press Enter to toggle Yes/No.**

   When Yes, the protein factors are displayed.

6. **Edit the Dilution Multiplier, if required.**

7. **Edit the Units, if required.**

8. **When using a DNA method with scan, press the Setup Scan function key.**



This will access the Setup Scan page from which the start and stop wavelengths can be edited.

If the method has been edited, it may be saved to the instrument Library or to disk. A different file name must be used.

9. **Clear the beams or insert suitable blank(s) and close the lid of the sample compartment.**

10. **Press Zero.**

    A message “Measuring Baseline” appears.

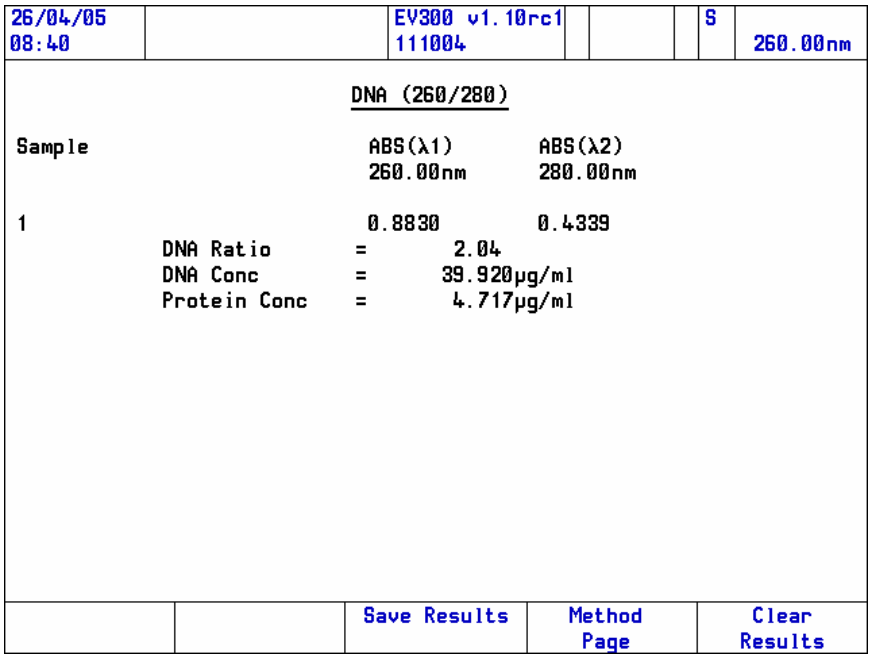
11. **Insert the sample in the cell holder and close the lid of the sample compartment.**

12. **Press Run.**

    A message “Measuring Sample” appears.

Note Quartz cells MUST be used for all DNA measurements. Glass or plastic cells are not suitable because they do not transmit at the wavelengths used. ▲

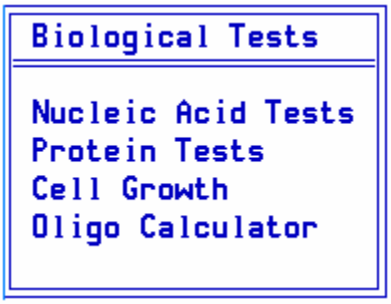
The results page contains the results of the calculations and the Scan when applicable.



13. **Press the Save Results function key to save the results to the instrument Library or to disk.**

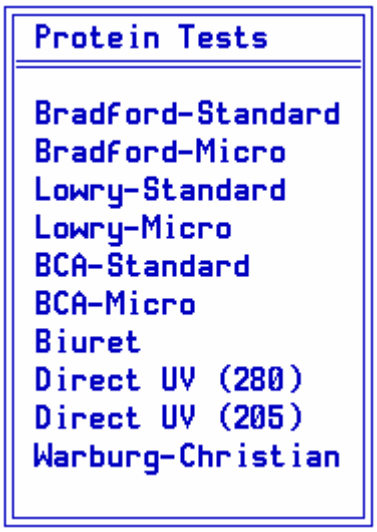
How to Run a Protein test:

1. **Highlight Biological Tests on the Main Menu page and press Enter.**



   The Biological Tests sub-menu will open.

2. **Highlight Protein Tests and press Enter.**



   Choose the required test from the Protein Tests sub-menu. The relevant test page opens. The content and layout of the page will depend on the test selected, the way the software is configured and which accessories are fitted.

   The method parameters are documented values published in the literature. Parameters may be edited, if required.

   The Standards Table contains recommended concentrations but can be edited as required.

3. **Edit the Units and enter High/Low limits, if required.**

4. **Highlight Statistics and press Enter to toggle Off/On.**

5. **Prepare the standards.**

   Ideally the protein used for preparation of the standards is structurally similar to the unknown. Alternatively, bovine serum albumin or immunoglobulins are both commonly used as protein standards.

6. **Clear the beams or insert suitable blank(s) and close the lid of the sample compartment.**

7. **Press Zero.**

   A message "Zeroing" appears.

8. **Press the Calibrate function key and follow the on-screen instructions to perform the calibration.**

9. **Save the calibrated method to the instrument Library or to disk, using a new file name.**

Note A previously calibrated method can be loaded from the instrument Library or from disk. Press the Load Method function key. ▲

10. **Once the calibrated method is present, place the sample in the cell holder and close the lid of the sample compartment.**

11. **Press Run.**

    The results will be seen on the results page.

Note Quartz cells MUST be used for all Direct UV 260, Direct UV 280 and Warburg-Christian methods. Glass or plastic cells are not suitable because they do not transmit fully at the wavelengths used. ▲

12. Save the results.

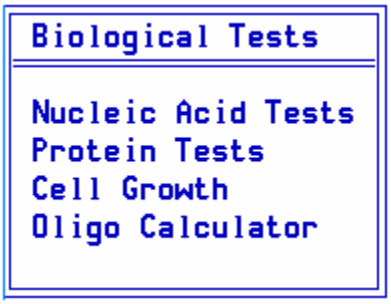
Press the Save Results function key on the Results page.

How to

Measure cell growth:

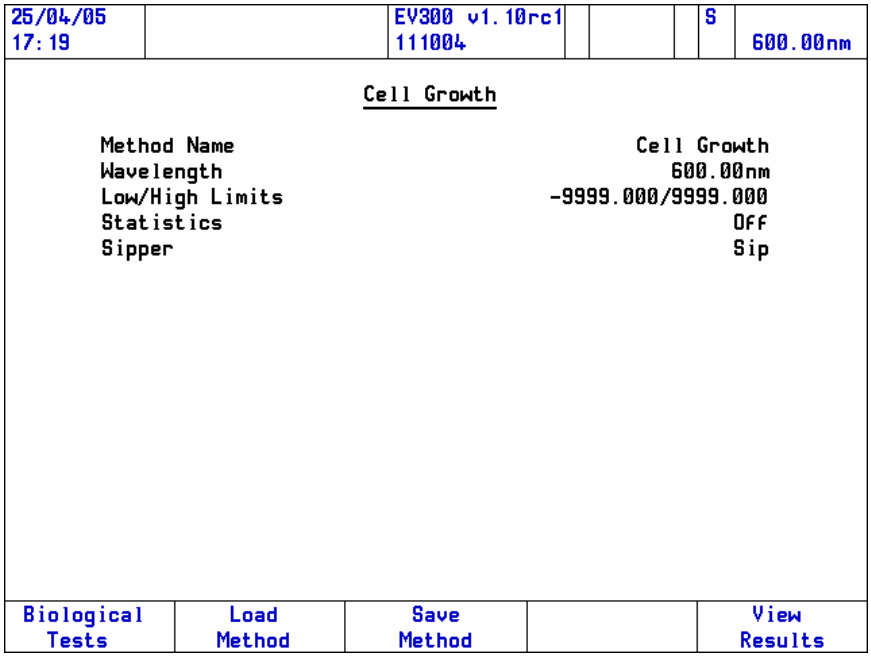
The cell growth measurement uses absorbance at 600nm to indicate the progress of cell growth in a sample. The instrument does not perform any calculations or graph the data.

1. Highlight Biological Tests on the Main Menu page.



The Biological Tests sub-menu will open.

2. Choose Cell Growth and press Enter.



The relevant test page opens. The content and layout of the page will depend on the way the software is configured and which accessories are installed.

3. **Clear the beams or insert suitable blank(s) and close the lid of the sample compartment.**

4. **Press Zero.**

   A message "Zeroing" appears.

5. **Place the unknown(s) in the correct cell position(s) and close the lid of the sample compartment.**

6. **Press Run.**

   The sample number and absorbance are displayed on the Results page.

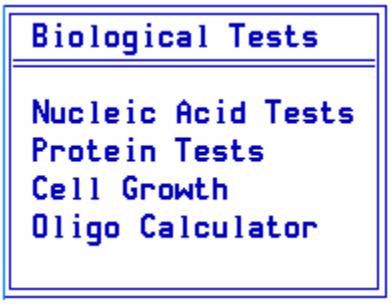
How to

Use the Oligo calculator:

The oligonucleotide calculator determines the following data for a base sequence that you enter:

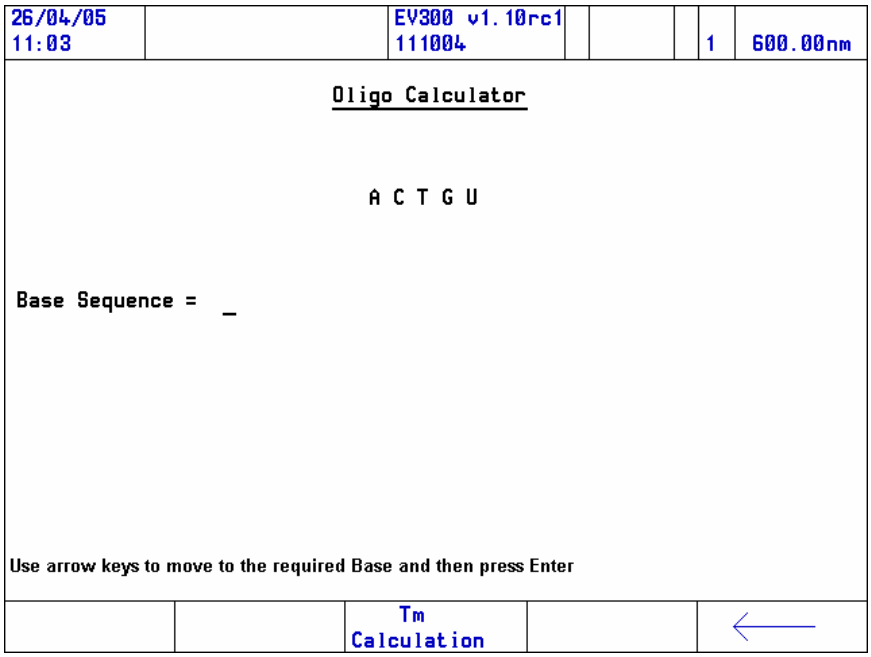
- Number of bases
- Percent GC content
- Molecular weight
- Absorptivity (extinction coefficient)
- Conversion factor to be used in oligonucleotide measurements
- T_m for oligos up to 20-mers, DNA, DNA hybrids and RNA-RNA hybrids

1. **Highlight Biological Tests on the Main Menu page.**



The Biological Tests sub-menu will open.

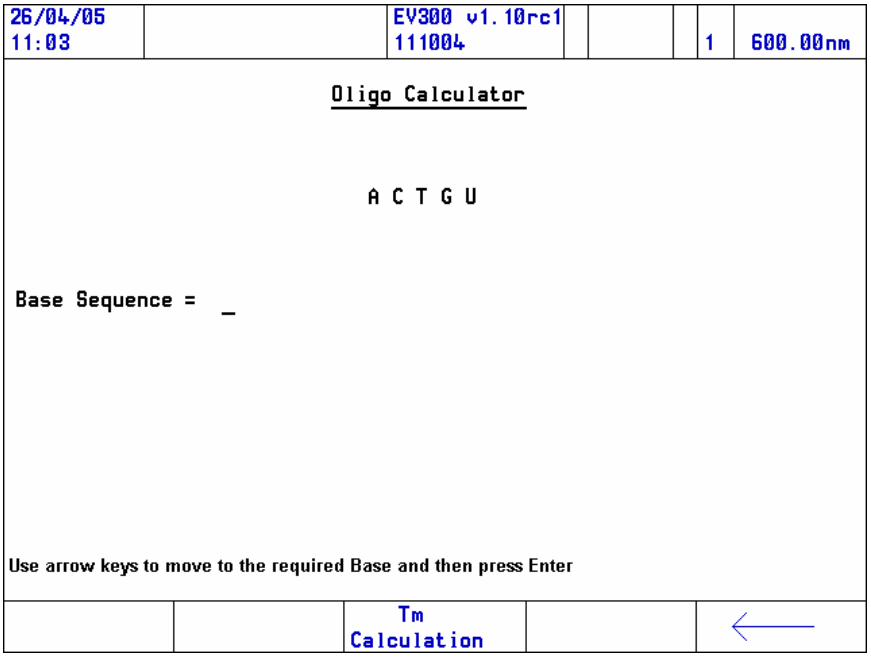
2. **Choose Oligo Calculator and press Enter.**



The Oligo Calculator page opens.

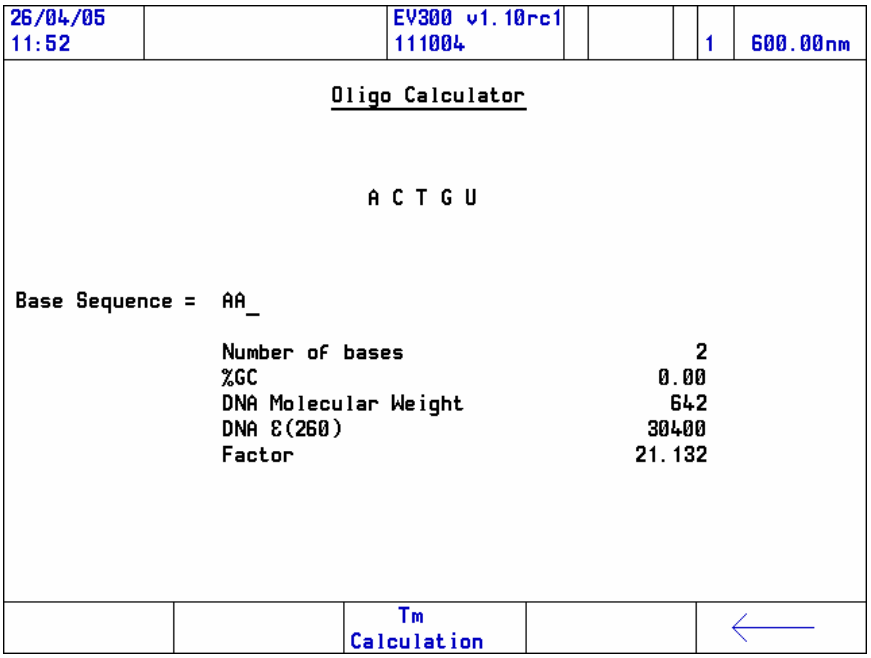
3. **Specify a base sequence.**

You will need to enter a base sequence before you can run the oligonucleotide calculations.



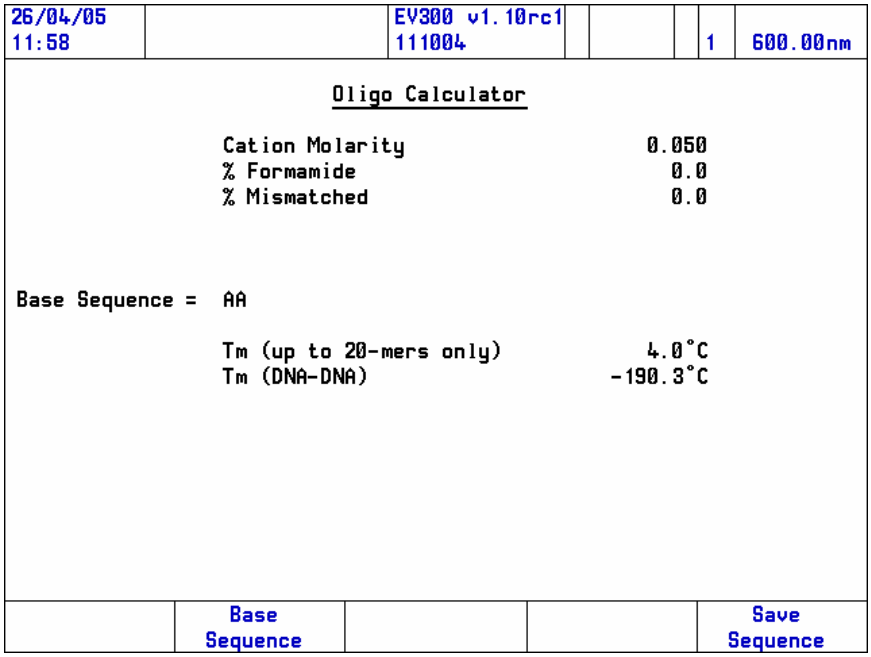
With the base sequence screen displayed, press the right/left arrow keys to select the required base.

4. **Repeat these steps until you have specified the entire base sequence.**



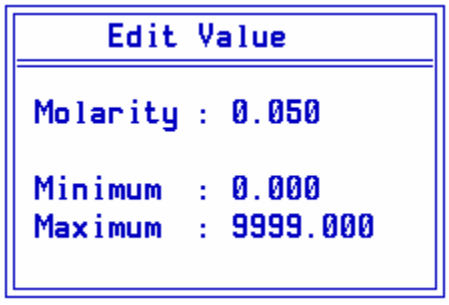
The displayed number of bases, %GC content, DNA Molecular Weight, DNA absorptivity (extinction coefficient), and factor will be updated as each new base is added to the sequence.

5. **Press the T_m Calculation function key to view the melting point calculator.**

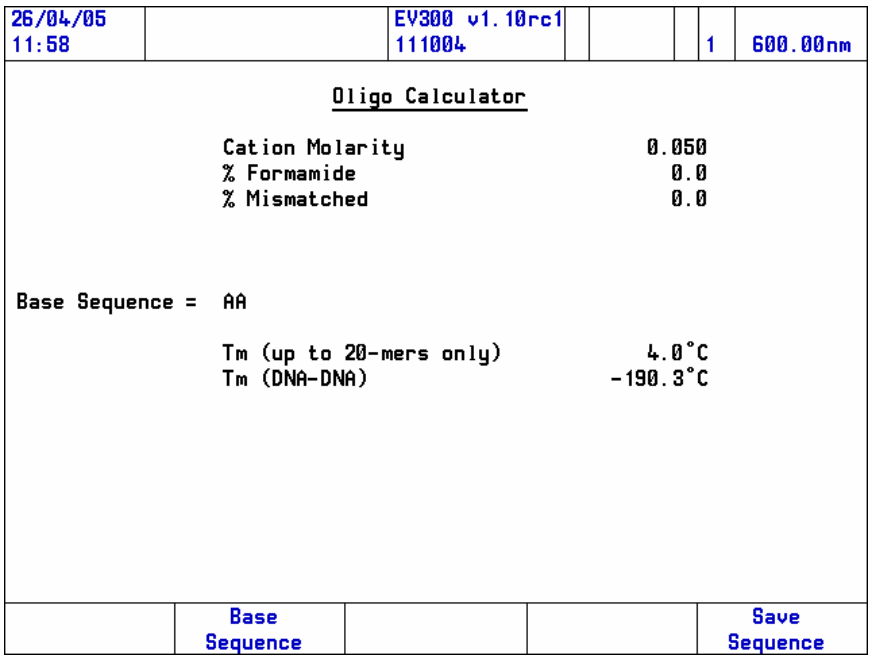


6. **Highlight parameter to be changed and press Enter.**

A pop-up menu will appear. Here is an example:



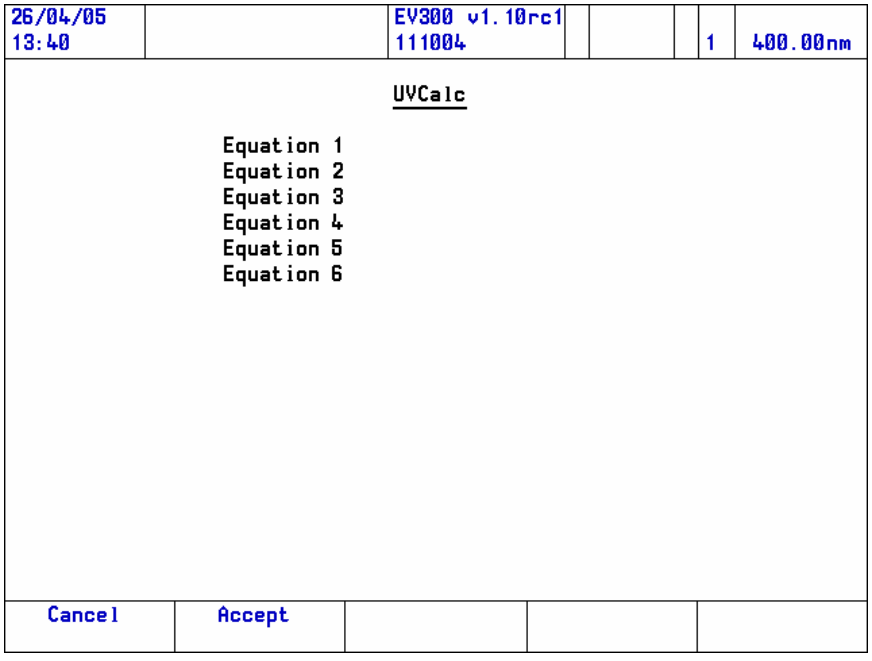
Once all parameters have been set appropriately, the relevant set of melting point predictions will be displayed.



Using UV*calc*

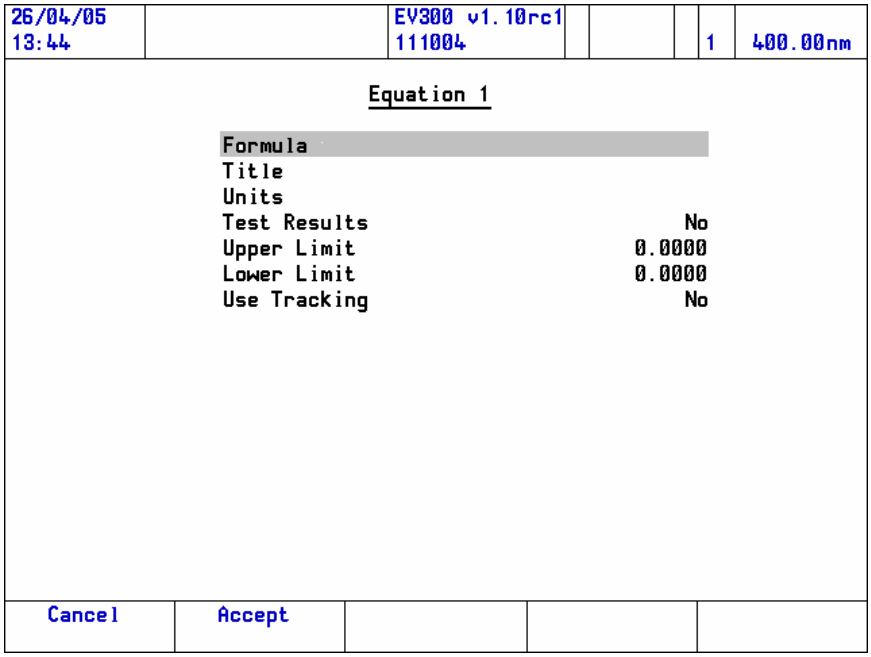
UV*calc* enables customized calculations to be incorporated into Scan, Fixed and Quant methods.

1. **Highlight UV*calc* on the selected application page and press Enter.**



The equation selection page opens.

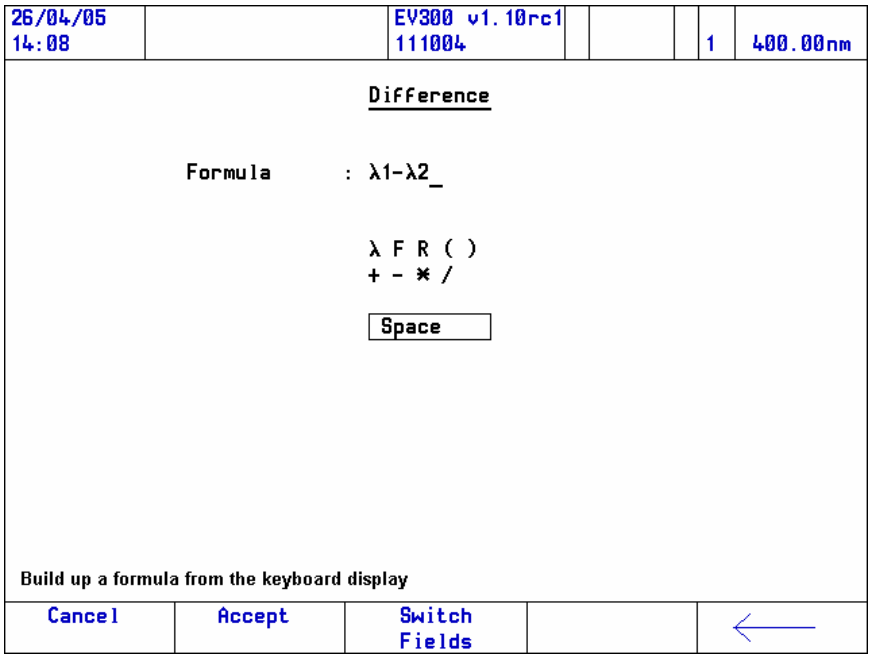
2. **Highlight the equation to be edited (Equation 1 if you are starting a UV*calc* calculation) and press Enter.**



The equation entry page for the selected equation opens, with the Formula line highlighted.

3. **Press Enter to edit the formula.**

The formula page opens.



A formula may contain any combination of the following elements:

Measurement — Choose between once only and repeated.

Factor — Choose between fixed or entered by the user at run time.

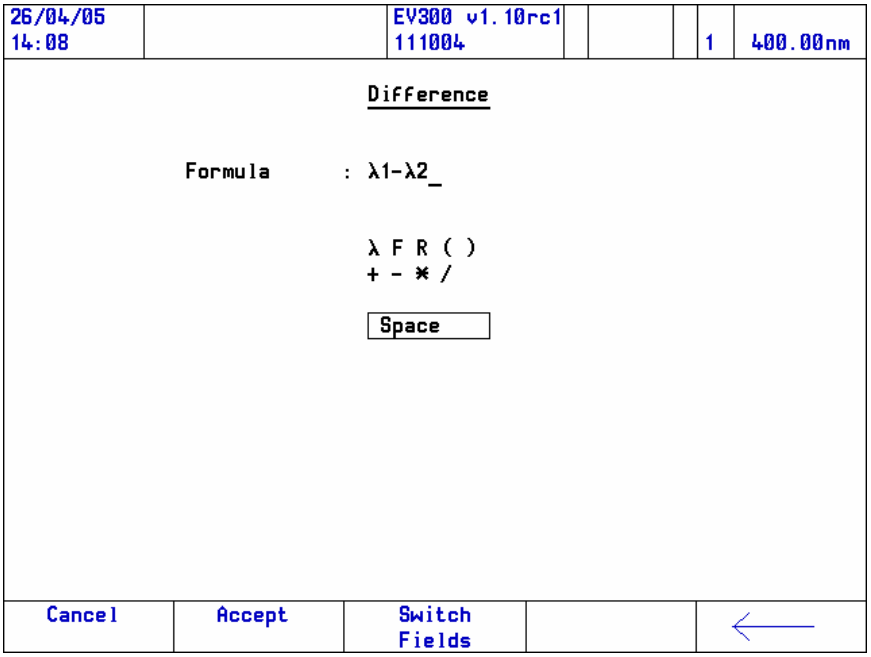
Result — A result carried over from a previous UV*calc* equation brackets (parentheses). Nested brackets are supported.

Operators — Add, subtract, multiply, divide spaces may be used, if required.

4. **Use the arrow keys to move the highlight to the required element and press Enter.**

   Further options will be made available as required. The back arrow and function keys may be used to edit the equation.

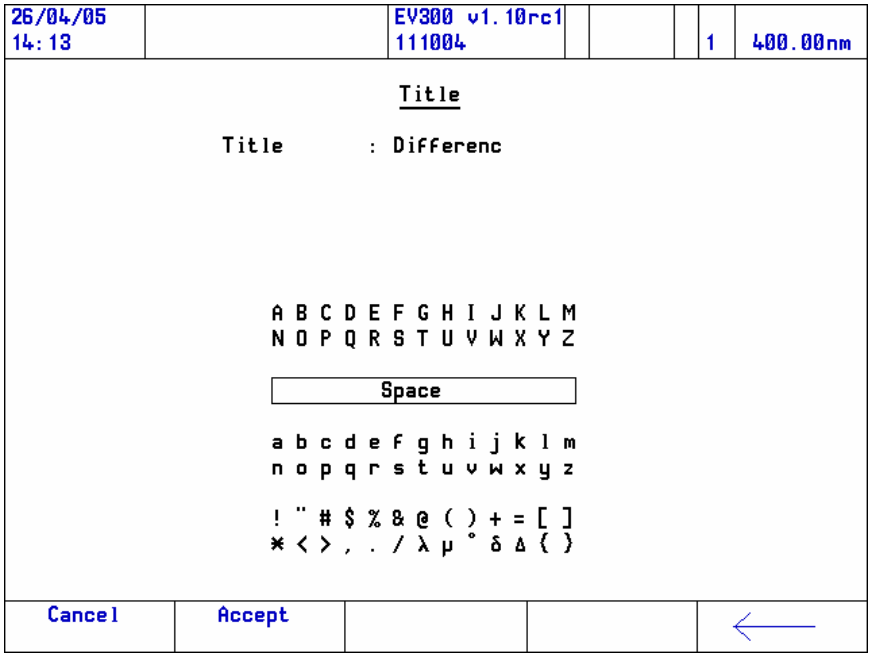
5. **When the equation is set up, press the Accept function key.**



The Cancel button enables you to exit without changing.

6. Add title to equation, if required.

From the Equation Entry page, it is possible to enter a title for the equation, units for the results and to enable the result to be tested in order to establish whether it falls between defined limits. Here is an example:



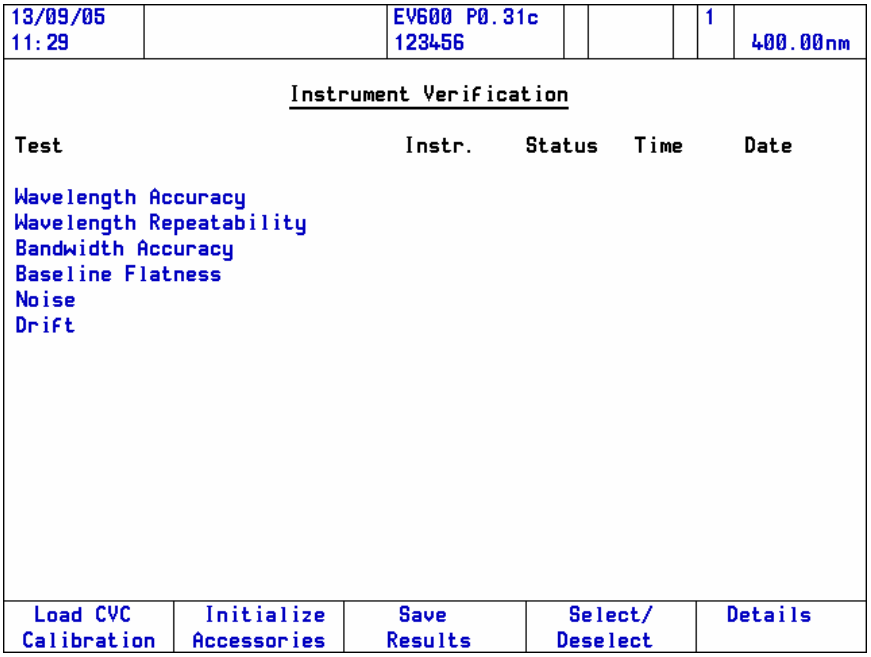
In each case, highlight the item and pres Enter and then enter the test and/or numbers in the usual way.

Instrument verification tests

The menu may contain fewer items than are shown, depending on the accessories installed.

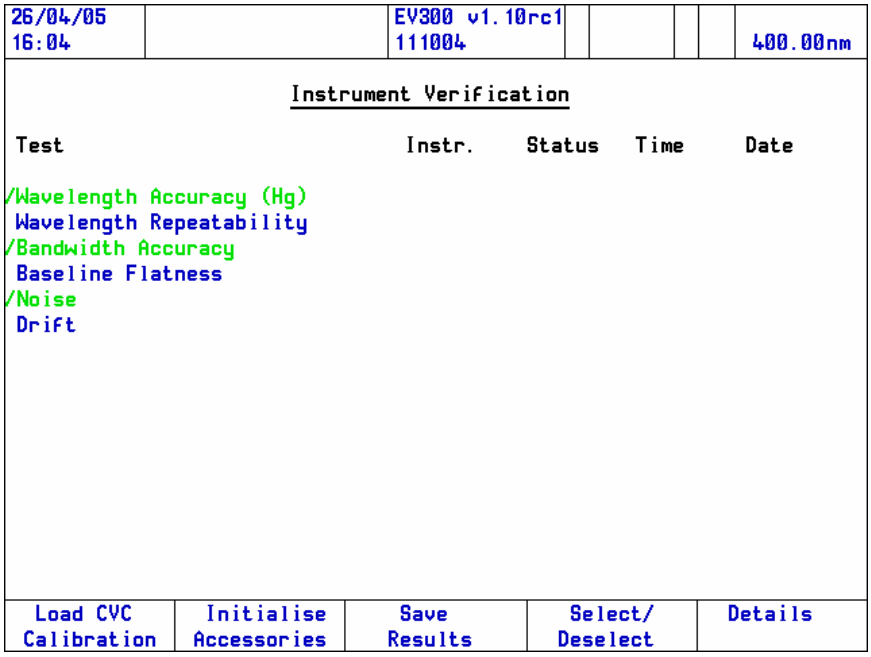
Note Turn on the instrument at least an hour before starting performance qualification tests. ▲

1. **From the Main Menu page, press the Utilities function key.**
2. **Highlight Instrument Verification and press Enter.**



3. **Highlight the test(s) required and press the Select/Deselect function key to select it.**

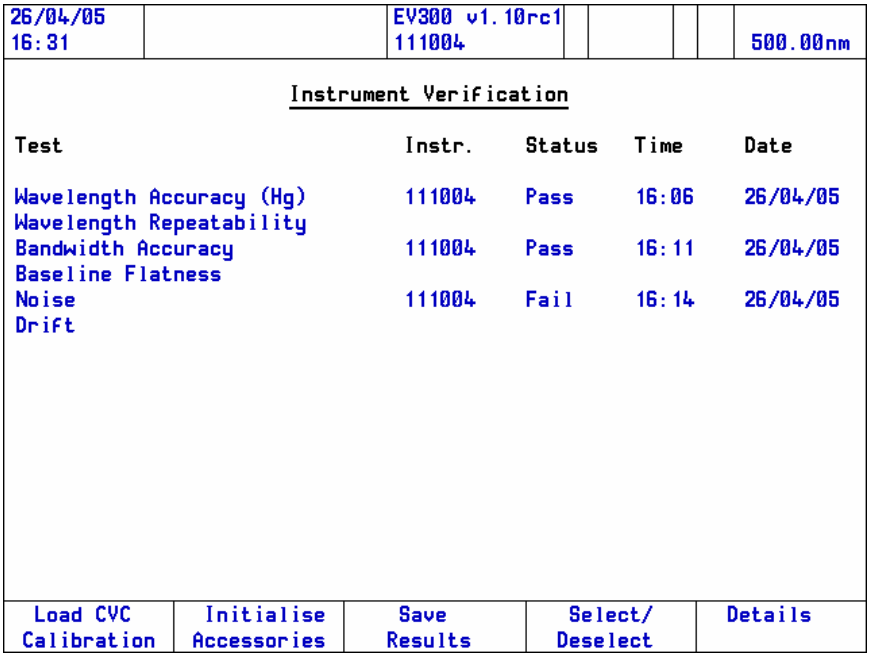
   Here is an example:



4. **Press Run after selection(s) has been made.**

Note The drift test will take at least an hour. ▲

5. **Press the Summary function key to return to the Instrument Verification page.**



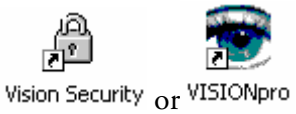
6. **Press the Save Results function key to save the current results to the Library or to disk.**

   Previous results can be loaded using the Load Results function key or by using the Library or disk menu items on the Main Menu page.

   Further tests will become available when an instrument verification accessory (CVC or Mercury lamp) is installed. Refer to the appropriate accessory manual.

VISION*security* and VISION*pro*

This section provides a brief introduction to the functionality of VISION. For more detailed information, refer to the comprehensive on-line Help system on the VISION CD.



To start VISION, you can either:

- Double-click on the VISION icon on the Windows desktop;
- Select Start > Programs > VISION*pro* > VISION*pro;* or
- Start > Programs > VISION*security* > VISION*security* as appropriate.

By default, VISION uses the Com 1 port on the computer.

Chapter 5 Maintenance

This chapter provides important information about maintaining your spectrophotometer. The subjects that are explained include:

- Routine maintenance.
- Cleaning instrument exterior.
- Internal printer.
- Light sources.

Routine maintenance

The information given in this section deals only with those parts of maintenance or service that can be safely carried out by the user. Work other than that detailed should be carried out by a Thermo Electron trained service engineer.

- The interior should be kept as dust-free as possible.
- The sample compartment cleaned regularly.
- Wipe off any spilled chemicals immediately.

Cleaning instrument exterior

The exterior and sample compartment of the instrument can be cleaned periodically as follows:

⚠ Caution Do not allow moisture to leak into the instrument. ▲

1. **Switch off the spectrophotometer and disconnect from AC power source.**
2. **Using a lint-free cloth dampened with a weak solution of detergent and water, wipe the exterior surface of the instrument as necessary.**
3. **Wipe over with a cloth dampened with plain water.**
4. **Dry the surface with another cloth.**

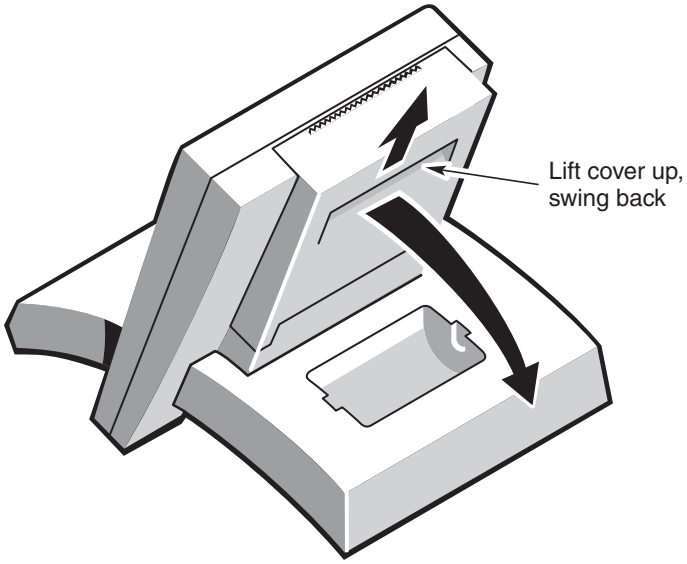
Internal printer

Some instruments are provided with a factory installed internal printer. Keep the paper stored at room temperature, away from light. Spare paper is available from Thermo Electron.

To change the internal printer paper:

1. **Open the printer housing.**

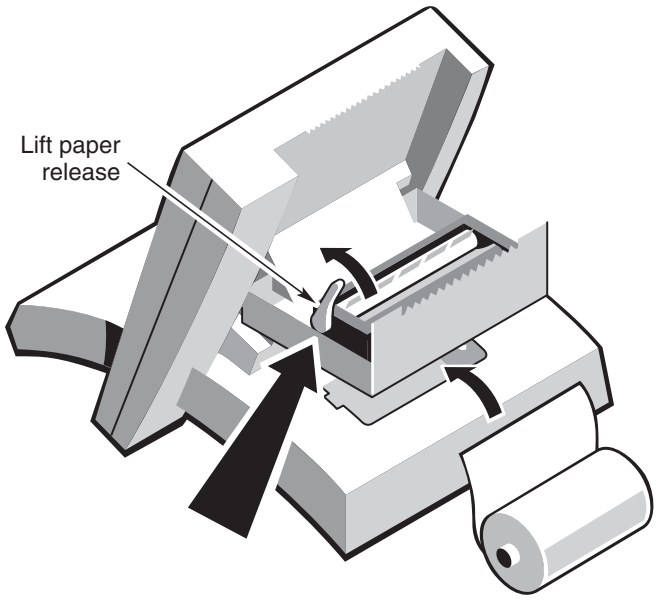
   To do this, lift the cover and swing it away from the display.



2. **Turn the paper release lever through 90°, towards the display.**

3. **Feed the paper, shiny side down, into the input slot.**

   Ensure that the paper emerges correctly through the printer and approximately 10cm (4 in) has been pulled clear.

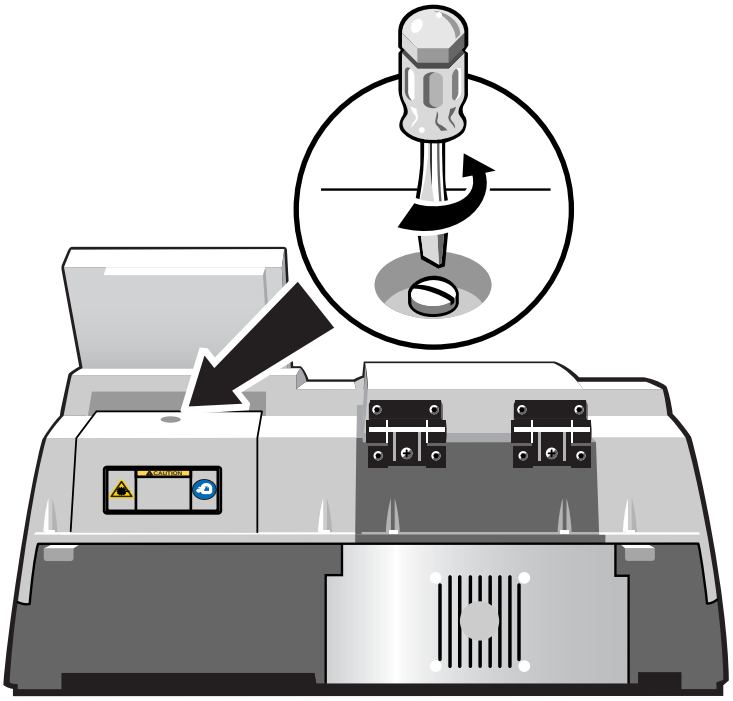


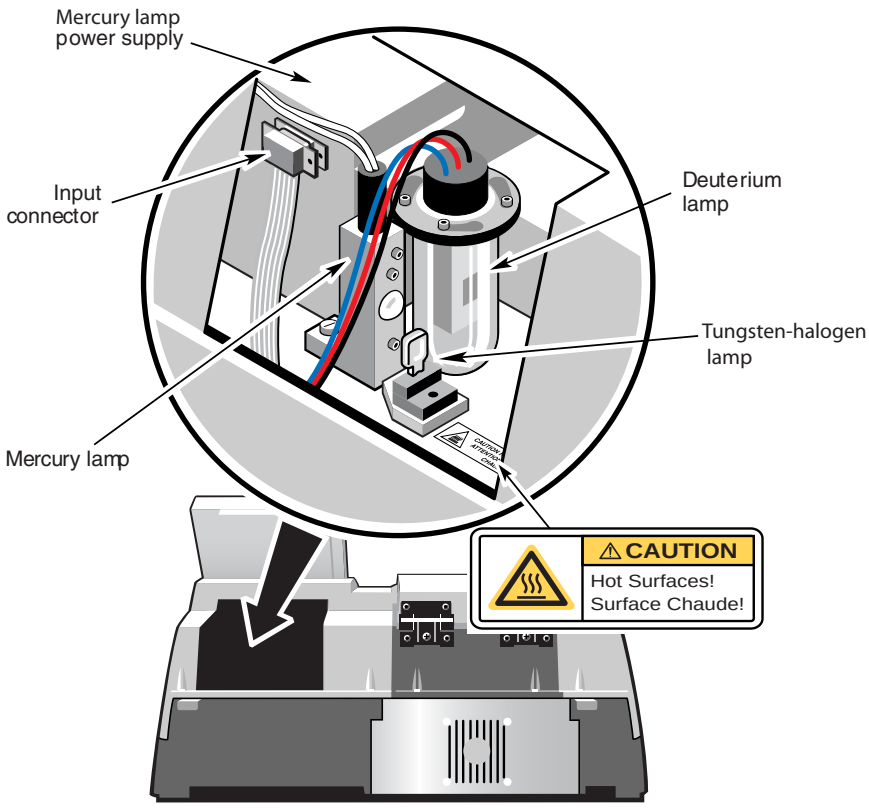
4. **Push the paper release lever back into place.**

5. **Replace the printer housing.**

Light sources (Evolution 600 only)

The tungsten-halogen and deuterium lamps get very hot while the instrument is operating. Before removing either lamp, turn off the instrument and allow it to cool for 15 minutes.





⚠ Danger Avoid shock hazard. Always power off the spectrophotometer and disconnect the power supply from the wall outlet or power strip. ▲

⚠ Warning The following lamp replacement procedures require removal of the lamp area cover. Operation of the instrument with the optics cover removed exposes the operator and other observers to ultraviolet radiation, which can damage the human eye and cause burning of the skin. All personnel in the area must wear protective UV-absorbing safety glasses. ▲

Deuterium lamp

UV radiation from a deuterium lamp can be harmful to the skin and eyes. Always view the lamp through protective glasses that will absorb UV radiation.

1. **Turn off the spectrophotometer and unplug from the AC outlet.**

⚠ Caution Allow at least 15 minutes for the instrument to cool before removing protective cover. ▲

2. **Remove back cover.**
3. **Remove the burned out bulb from its socket.**
   a. Using a 2.5mm hex driver, turn screws 2 turns counterclockwise.
   b. Rotate the lamp and pull upward until out of housing.
   c. Unplug inline connector.
4. **Insert new lamp.**

⚠ Caution Never touch the bulb with your fingers. Oil from your skin may cause the bulb to burn out quickly or explode. ▲

   a. Plug in inline connector.
   b. Place lamp in lamp housing aligning hex screws.
   c. Using 2.5mm hex driver tighten screws.

5. **Replace rear cover.**
6. **Plug in the spectrophotometer to the AC outlet and turn on the power switch.**

Note Lamp hours and energy (if applicable) must be reset from the controlling software. ▲

Tungsten-halogen lamp

Always view the lamp through protective glasses that will absorb UV radiation.

1. **Turn off the spectrophotometer and unplug from the AC outlet.**

⚠ Caution Allow at least 15 minutes for the instrument to cool before removing protective covers. ▲

2. **Remove back cover.**
3. **Remove the burned out bulb from its socket.**

   Before removing, note the orientation of the lamp socket.

   a. Using a 2.5mm hex wrench, loosen the two securing screws.

   b. Holding the lamp socket by the mounting tabs, remove the copper spring clip.

   c. Unplug the tungsten-halogen lamp from socket.

4. **Insert new bulb.**

⚠ Caution Never touch the bulb with your fingers. Oil from your skin will cause the bulb to burn out quickly or explode. ▲

   a. Holding the lamp socket by the mounting tabs, insert the bulb pins into the socket.

   b. Replace the copper spring clip.

   c. Replace the lamp socket ensuring the orientation is correct.

   d. Align the securing screws and tighten with hex driver.

5. **Replace rear cover.**
6. **Plug in the spectrophotometer to the AC outlet and turn on the power switch.**

Note Lamp hours and energy (if applicable) must be reset from the controlling software. ▲

Mercury lamp (Evolution 300 and Evolution 600)

Refer to the accessory user's guide for directions on replacing the mercury lamp.