Quawell

Q5000

UV-Vis SPECTROPHOTOMETER

USER'S MANUAL

V1.0.1



Rev 07-01-2010

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1. OVERVIEW

What Is The Q5000?

Quawell Q5000 is a micro-volume UV-Vis spectrophotometer. Its full spectrum (200-850nm) allows Q5000 to measure nucleic acids, microarray probes, purified proteins, and fluorescent and fluorescently-dyed proteins; monitor the density of suspended cellular, bacterial and yeast cultures; and perform general laboratory UV-Vis absorbance measurements. A unique technology used in the Q5000 holds 0.5-2.5 ul samples between upper and lower measurement surfaces without the use of a cuvette, and measures the sample in about 5 seconds with a high degree of accuracy and reproducibility.

How to Operate

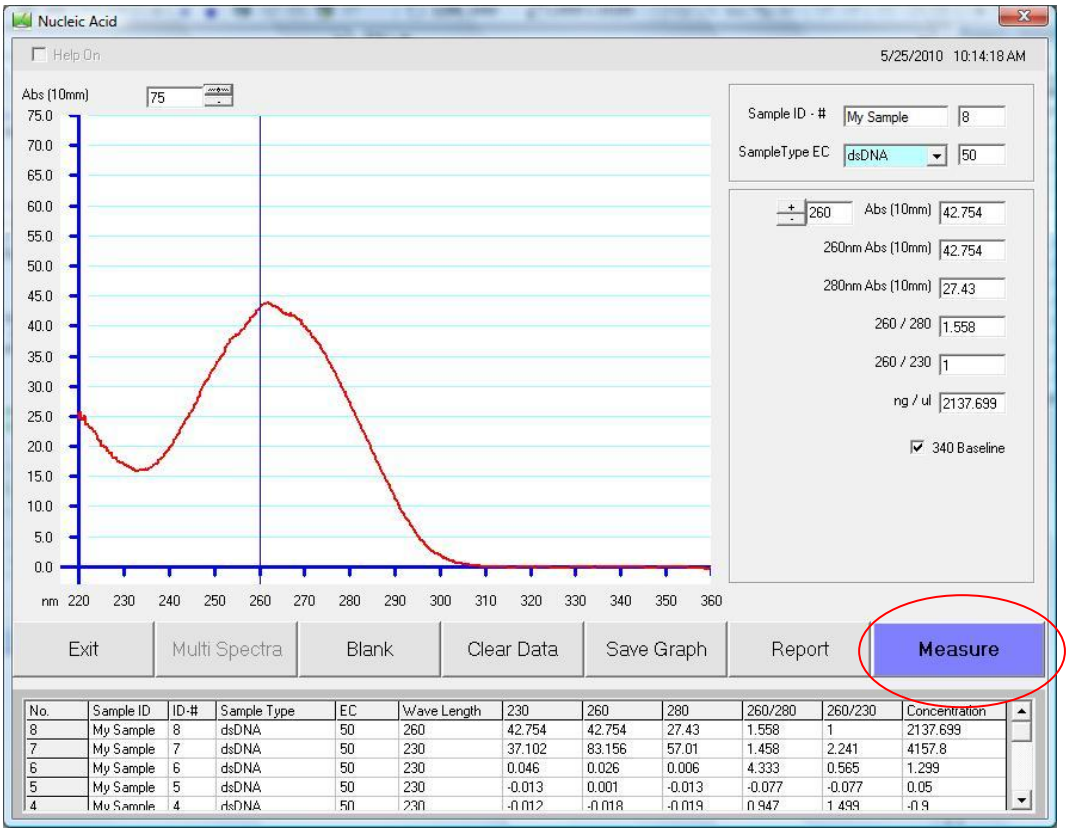
Operation of the Q5000 is simple and efficient. The Q5000 automatically performs all necessary measurements and calculations. The data are accumulated in the module data window and are easily exported to an Excel spreadsheet.

1. Lift the upper arm, add 2-2.5 ul sample to the lower measurement surface and close the upper arm.





2. Click the "Measure" button to complete all measurements and calculations.

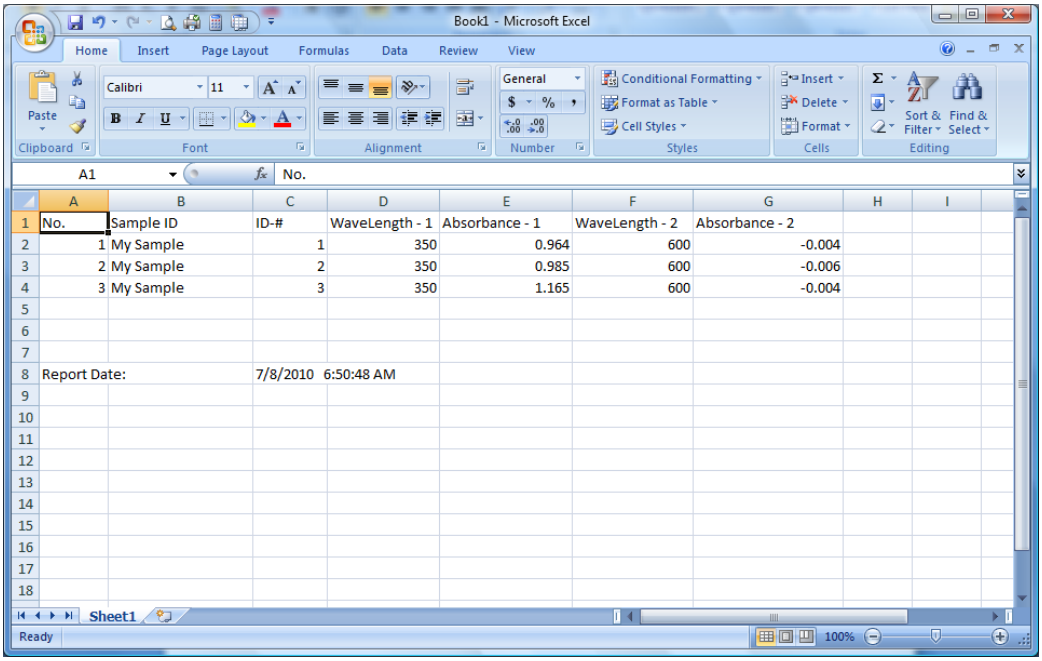


3. Lift the upper arm and remove the sample from both upper and lower surfaces with soft dry paper.





4. Export the data to an Excel worksheet by clicking the "Report" button.



No.	Sample ID	ID-#	WaveLength - 1	Absorbance - 1	WaveLength - 2	Absorbance - 2
1	My Sample	1	350	0.964	600	-0.004
2	My Sample	2	350	0.985	600	-0.006
3	My Sample	3	350	1.165	600	-0.004
Report Date:		7/8/2010	6:50:48 AM			

2. THE PATENT-PENDING TECHNOLOGIES

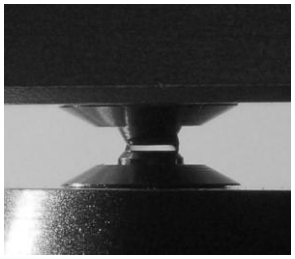


Figure 1: Initially the sample is loaded and the upper arm is closed.

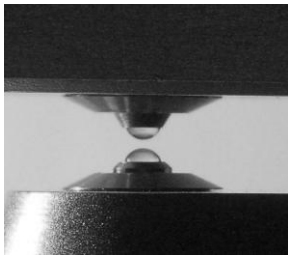


Figure 2: The upper arm is lifted by the actuator, due to molecular effects, the sample is broken into two separate parts, one part is attached to lower surface and the other part is attached to upper surface.

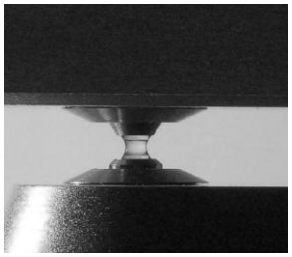


Figure 3: After reaching its home position, the upper arm is lowered. A liquid column is formed when the upper arm is lowered to the position where the two droplets touch. The measurement is then performed at this position without moving the upper arm upwardly again.

3. INSTALLATION

Computer Requirements

- Microsoft Windows XP, Vista or later
- Microsoft Office 2003 or later
- CPU: 400MHZ or higher
- 64MB RAM
- USB port
- Monitor: 1024x768

Software Setup

Note: Do not connect Q5000 to PC before installing the software.

- Insert the CD into CD driver and double click the file "setup" in the folder CD-RAM\Q5000 V1.0.0. Follow the prompts to finish installation of the software.
- Connect Q5000 to PC with the USB cable and the driver will be installed automatically.

"Version Conflict"

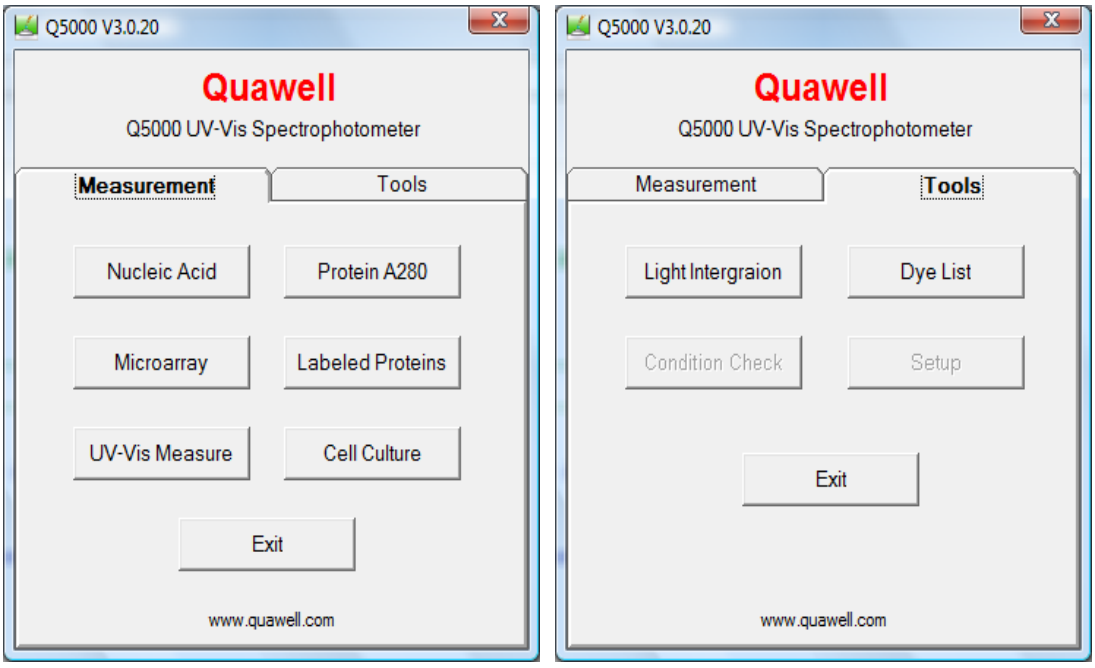
If you install Q5000 software in a computer with Windows Vista, please click option "Yes" when "Version Conflict" message appear as below: "A file being copied is older than the file currently on your system. It is recommended that you keep your existing file. Do you want to keep this file?"

"Abort", "Retry" and "Ignore" option

When you see a message that requires you select from options "Abort", "Retry" or "Ignore", please click option "**Ignore**" and continue to install Q5000 software.

4. MAIN MENU

Q5000 main menu contains two tabs, the Measurement tab and the Tools tab. The default is Measurement tab. The "Exit" buttons in both tabs will exit the Q5000 program.



The Measurement tab:

1. Nucleic Acids
2. Microarray
3. UV-Vis Measure
4. Protein A280
5. Labeled Protein
6. Cell Culture

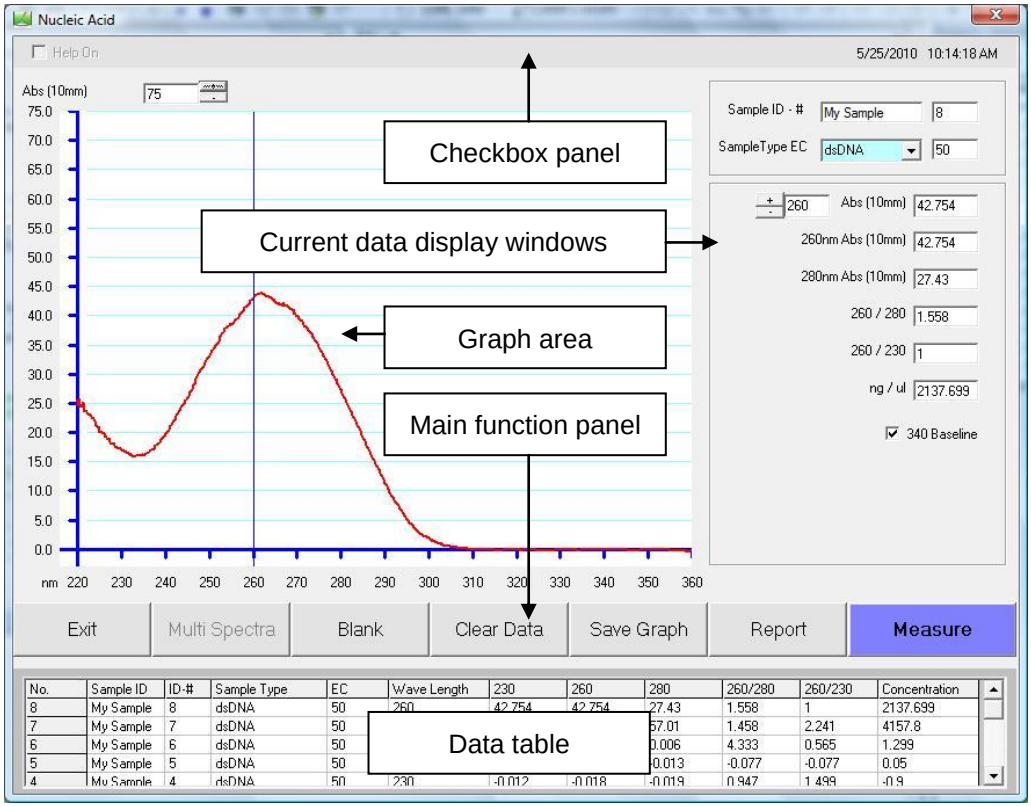
The Tools tab:

1. Light Integration: Allows the user to adjust the light integration.
2. Dye List: edit the dye data

Software Version Number: it is on the top right side of the menu. Please check our website www.quawell.com on the bottom of the main menu for software upgrades and product news.

5. APPLICATION MODULE INTERFACE

The following is the Q5000 sample measurement user interface. The window is divided into five different areas:

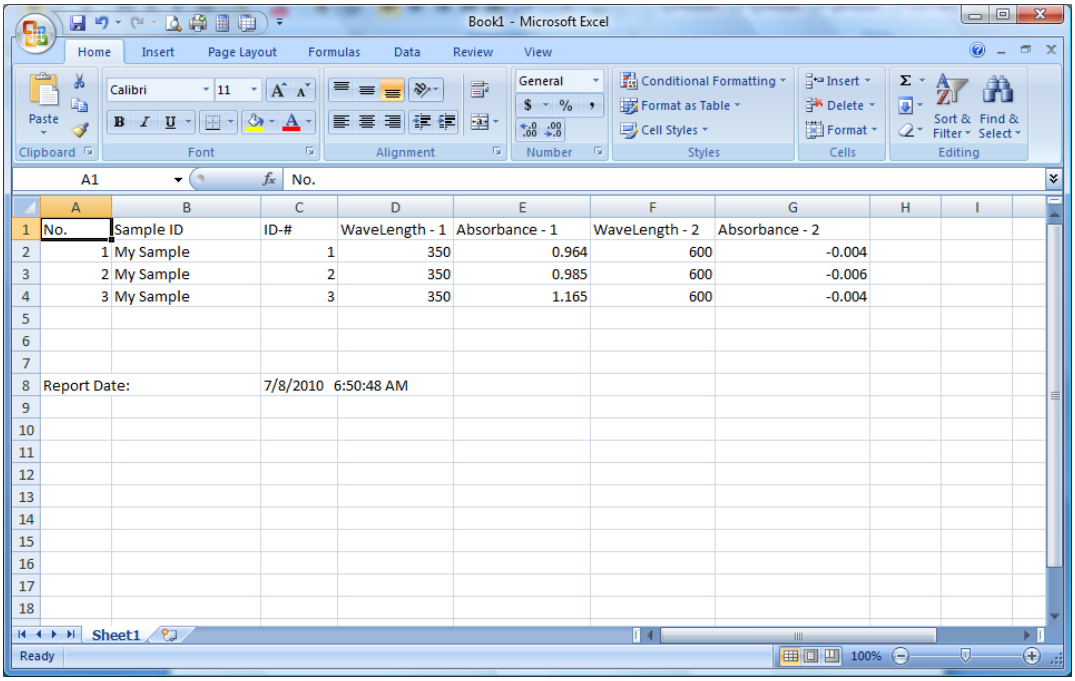


1. **Checkbox Panel:** contains online-help and other options. The user can select the option based on the particular need.
2. **Current data display windows:** display the data and information of the sample being measured.
3. **Main function Panel:** contains all main functions for sample measuring and data handling.
4. **Graph area:** displays the absorbance (Y-axis) and spectrum (X-axis).
5. **Data Table:** used for holding and displaying the accumulated data. The maximum number of rows is 1000.

6. SAVING THE DATA AND INFORMATION

Note: Q5000 program does not save the data automatically. All data and information for the sample being measured will be lost if it is not saved.

User can save the data using the "Report" function to export the data from the data table to an Excel spreadsheet.



	A	B	C	D	E	F	G	H	I
1	No.	Sample ID	ID-#	WaveLength - 1	Absorbance - 1	WaveLength - 2	Absorbance - 2		
2	1	My Sample	1	350	0.964	600	-0.004		
3	2	My Sample	2	350	0.985	600	-0.006		
4	3	My Sample	3	350	1.165	600	-0.004		
5									
6									
7									
8	Report Date:		7/8/2010	6:50:48 AM					
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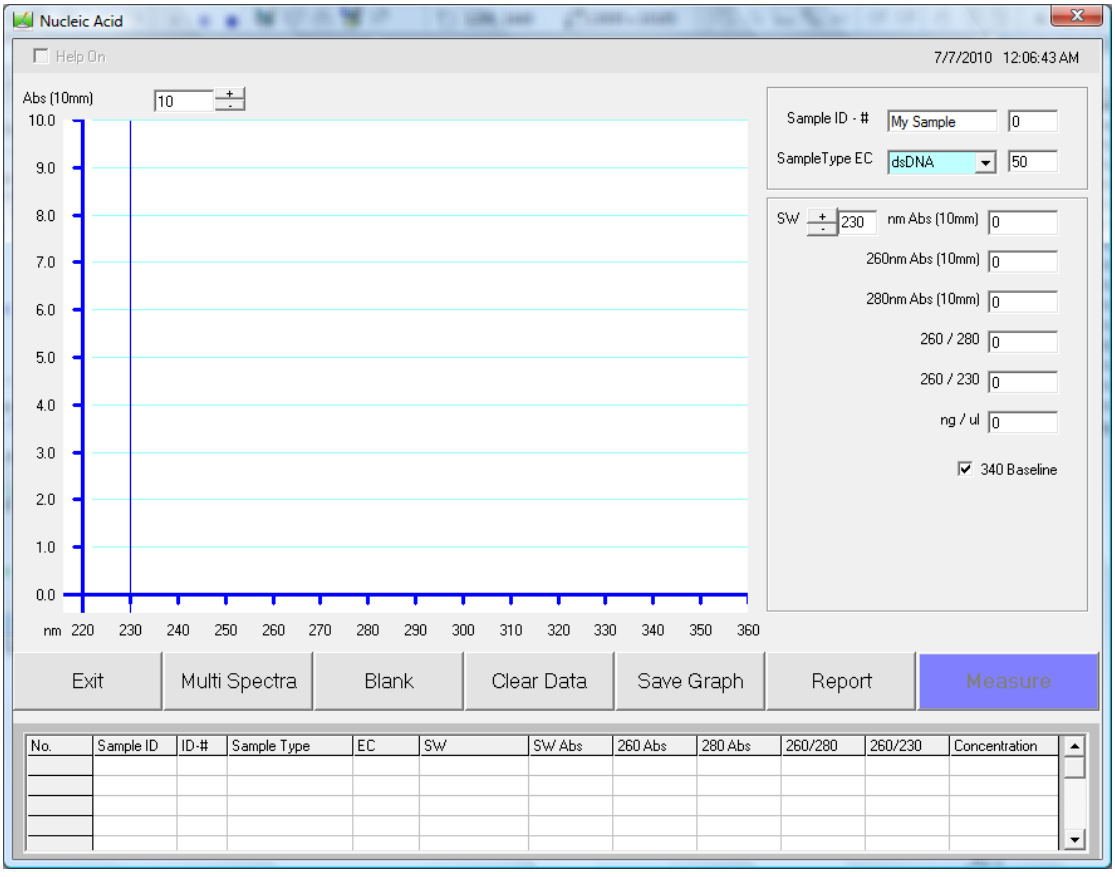
7. NUCLEIC ACIDS

Overview

The Q5000 measures nucleic acid absorbance, automatically calculates concentration, and displays the data on the screen.

Nucleic Acid Screen Features

Select Nucleic Acid from the Q5000 main menu and the user interface shown below will appear on the screen.



- **340 nm Baseline:** the absorbance of nucleic acid at 340nm should be very

nearly zero. If the Baseline option is checked, the absorbance value of the baseline is subtracted from the absorbance at 260 nm and all spectra are referenced to this zero. The default value is check.

- **Graph Display**: The Y-axis indicates the absorbance value and the X-axis is for the wavelength. The maximum absorbance value is automatically adjusted to the highest absorbance value. The user is also allowed to manually enter the absorbance value.
- **Sample ID - #:** there are two windows that display the sample ID information. The left window displays the sample ID input by the user. The default is "My Sample". The right window displays the ID number for that sample ID. The ID number begins at 1 and increases by 1 for each measurement. Changing the Sample ID will automatically reset the ID number to 1.
- **Sample Type – EC:** chose the sample type for the sample to be measured. There are 3 different sample types for nucleic acids: dsDNA, ssDNA and RNA. The default is dsDNA. The sample type is disabled after blanking. The extinction coefficient of the sample selected will appear in the EC window.



- **SW and Abs (10mm):** user selected wavelength and corresponding absorbance. The selected wavelength and absorbance are not utilized in any calculations.
- **260 nm Abs (10mm):** absorbance of the sample at 260 nm.
- **280 nm Abs (10mm):** absorbance of the sample at 280 nm.

- **260/280:** ratio of absorbance at 260 nm and 280 nm.
- **260/230:** ratio of absorbance at 260 nm and 230 nm.
- **ng/ul:** sample concentration in ng/ul.

Measurement Range (dsDNA)

Concentration	Reproducibility	Abs accuracy (1 mm)
1 to 5000 ng/ul	1-100 ng/ul ±2 ul >100 ng/ul ±2%	2%

Sample Volume

There is no specific requirement for the sample volume; however for the best accuracy and reproducibility we recommend 2-2.5ul.

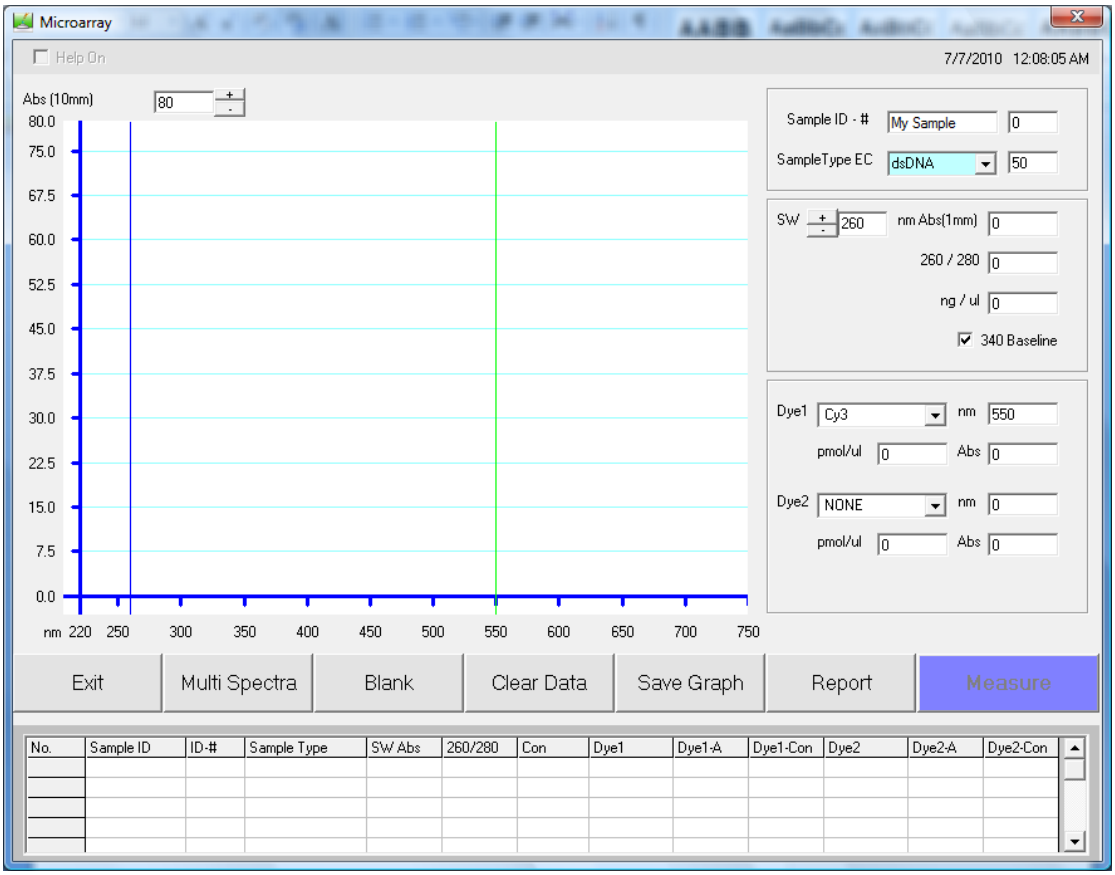
8. MICROARRAY

Overview

The Microarray module analyzes fluorescently-labeled nucleic acid probes. It simultaneously measures the concentration of the fluorescent tag and the nucleic acid at appropriate wavelengths.

Microarray Screen Features

Select Microarray from the Q5000 main menu and the application interface will appear on the screen.



- **340 nm Baseline:** the absorbance value of the 340 nm baseline is only used for the sample concentration calculation reported at 260 nm. The default of 340 nm baseline is checked.
- **750 nm Baseline:** it normalizes the visual spectrum display and calculates a baseline between 400 nm and 750 nm for dye concentration.
- **Graph Display**: The Y-axis indicates the absorbance value and the X-axis is for the wavelength. The maximum absorbance value is automatically adjusted to the highest absorbance value. The user is also allowed to manually enter the absorbance value.
- **Sample ID - #:** there are two windows that display the sample ID information. The left window displays the sample ID input by the user. The default is "My Sample". The right window displays the ID number for that sample ID. The ID number begins at 1 and increases by 1 for each measurement. Changing the Sample ID will automatically reset the ID number to 1.
- **Sample Type – EC:** chose the sample type for the sample to be measured. There are 3 different sample types for nucleic acids: dsDNA, ssDNA and RNA. The default is dsDNA. The sample type is disabled after blanking. The extinction coefficient of the sample selected will appear in the EC window.
- **SW and Abs (10mm):** user-selected wavelength and corresponding absorbance that are not utilized in any calculations.
- **260/280:** ratio of absorbance at 260 nm and 280 nm.
- **ng/ul:** sample concentration in ng/ul.
- **Dye1 and Dye 2 Selection Windows**
    1. **Dye1 or Dye2 drop-down list:** displays the dye that is pre-predefined

using the Dye List Editor. Please see Section 11 for details on how to predefine the list.

2. **Abs:** absorbance of Dye1 or Dye2.
3. **pmol/nl:** concentration of Dye1 or Dye2 in pmol/nl.
4. **Vertical Lines:** the green line indicates the peak position of the wavelength for Dye 1, and the blue vertical line indicates the peak position of the wavelength for Dye 2.

Measurement Range (Cy3)

Concentration (pmol/nl)	Reproducibility
0.2-100	**+/- 0.2%**

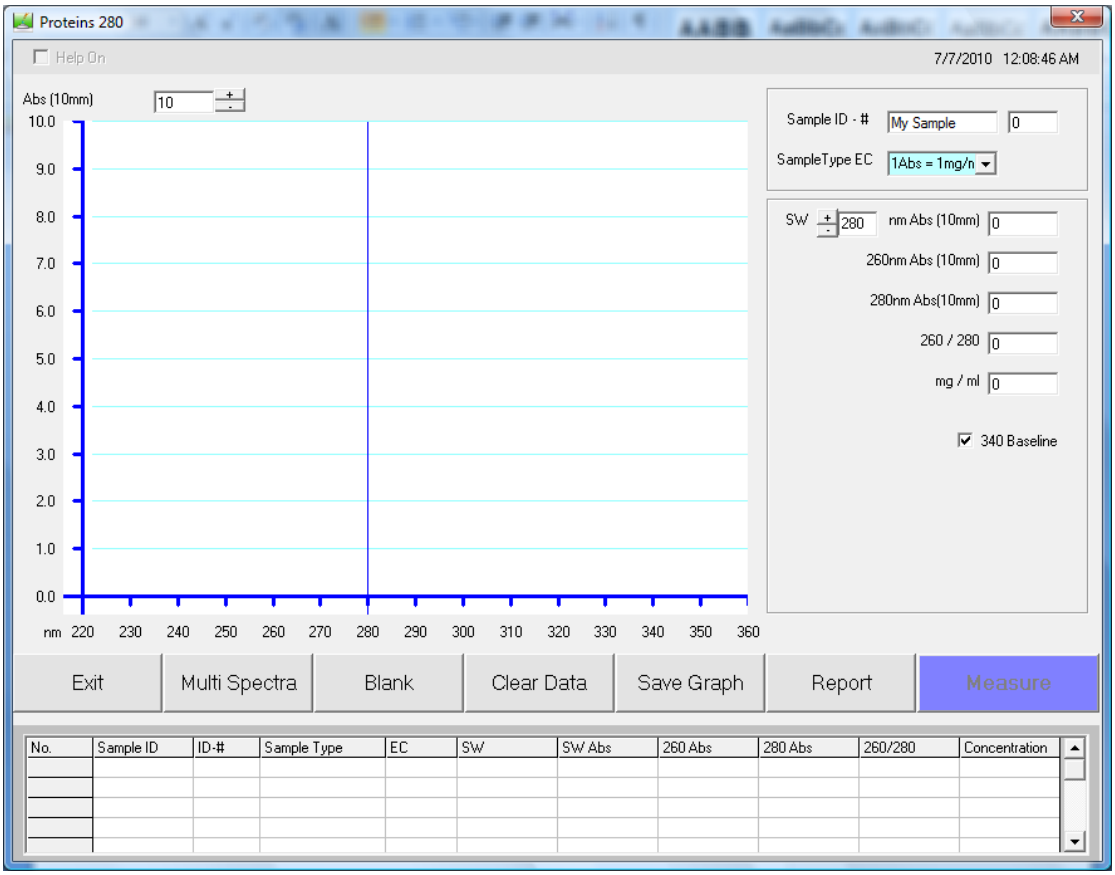
9. PROTEIN A280

Overview

Protein A280 measures the protein's absorbance at 280 nm and calculates the concentration. Since no protein standards are required, Protein A280 is fast and convenient.

Protein A280 Screen Features

Select Protein A280 from the Q5000 main menu and the module interface will appear on the screen.



- **340 nm Baseline:** the absorbance value of 340 nm baseline is only used for the sample concentration calculation reported at 280 nm. The default of 340 nm baseline is checked.
- **Graph Display**: The Y-axis indicates the absorbance value and the X-axis is for the wavelength. The maximum absorbance value is automatically adjusted to the highest absorbance value. The user is also allowed to manually enter the absorbance value.
- **Sample ID - #:** there are two windows that display the sample ID information. The left window displays the sample ID input by the user. The default is "My Sample". The right window displays the ID number for that sample ID. The ID number begins at 1 and increases by 1 for each measurement. Changing the Sample ID will automatically reset the ID number to 1.
- **Sample Type:** chose the sample type for the sample to be measured. There are 4 different sample types for protein measurement: 1Abs = 1mg/ml, BSA, IgG, and Lysozyme. The default is 1Abs = 1mg/ml. The sample type is disabled after blanking. The extinction coefficient of the sample selected will appear in the EC window.
- **SW and Abs (10mm):** the user selected wavelength and corresponding absorbance. The user-selected wavelength and absorbance are not utilized in any calculations.
- **Ads260 (10mm):** absorbance of the sample at 260 nm.
- **Abs280 (10mm):** absorbance of the sample at 280 nm.
- **260/280:** ratio of the absorbance at 260 nm and 280 nm.
- **mg/ml:** sample concentration in mg/ml.

Measurement Range (BSA)

Concentration	Reproducibility	Absorbance accuracy （1mm）
0.05 -100 mg/ml	±2%	Abs 0-0.2 ±0.002abs Abs 0.2-1 ±0.02abs

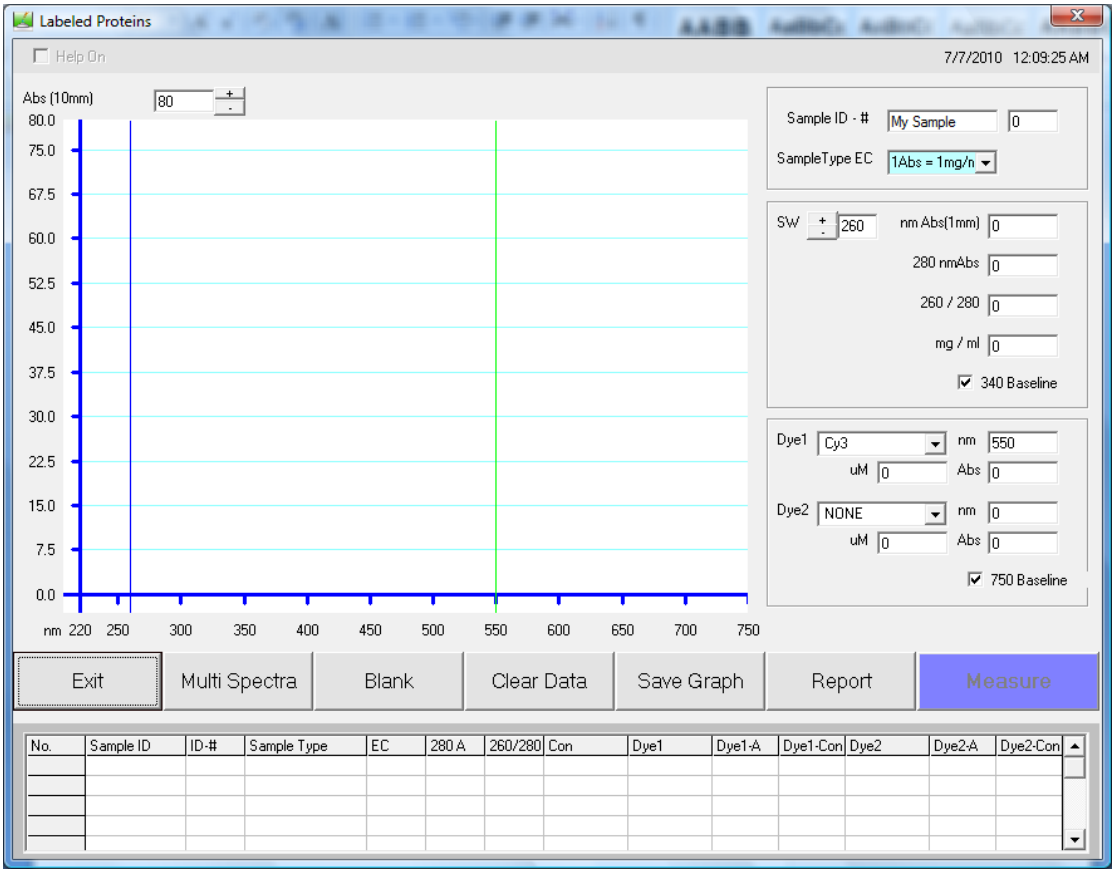
10. LABELED-PROTEINS

Overview

The Labeled-Proteins function will simultaneously measure both protein and fluorescent dye concentrations at appropriate wavelengths.

Proteins and Labels Screen Features

Select Proteins and Labels from the Q5000 main menu and the module interface will appear on the screen.



- **340 Baseline:** the absorbance value of 340 nm baseline is only used for the sample concentration calculation reported at 280 nm. The default of 340 nm baseline is checked.
- **750 Baseline**: it normalizes the visual spectrum display and calculates a baseline between 400 nm and 750 nm for dye concentration.
- **Graph Display**: The Y-axis indicates the absorbance value and the X-axis is for the wavelength. The maximum absorbance value is automatically adjusted to the highest absorbance value. The user is also allowed to manually enter the absorbance value.
- **Sample ID - #:** there are two windows that display the sample ID information. The left window displays the sample ID input by the user. The default is "My Sample". The right window displays the ID number for that sample ID. The ID number begins at 1 and increases by 1 for each measurement. Changing the Sample ID will automatically reset the ID number to 1.
- **Sample Type:** chose the sample type for the sample to be measured:
    1. 1Abs = 1mg/ml
    2. BSA
    3. IgG
    4. Lysozyme
- **SW and Abs (10mm):** the user selected wavelength and corresponding absorbance. The wavelength can be selected by using the up/down arrows or input the number in the wavelength box. The user-selected wavelength and absorbance are not utilized in any calculations.
- **Abs280 (10mm):** absorbance of the sample at 280 nm.
- **260/280:** ratio of the absorbance at 260 nm and 280 nm.

- **mg/ml:** sample concentration in mg/ml.
- **Dye1 and Dye 2 Selection Windows**
    1. **Dye1 or Dye2 drop-down list:** displays the dye that is pre-predefined using the Dye List Editor. See Section 11 for details on how to predefine the list.
    2. **Abs:** absorbance of Dye1 or Dye2.
    3. **uM:** concentration of Dye1 or Dye2 in uM.
    4. **Vertical Lines:** the green line on the absorbance-wavelength graph indicates the peak position of the wavelength for Dye 1, and the blue vertical line indicates the peak position of the wavelength for Dye 2.

Measurement Range

Concentration	Reproducibility
0.1 -20 mg/ml (BSA)	± 2%
0.2-100 uM (Cy3)	± 2%

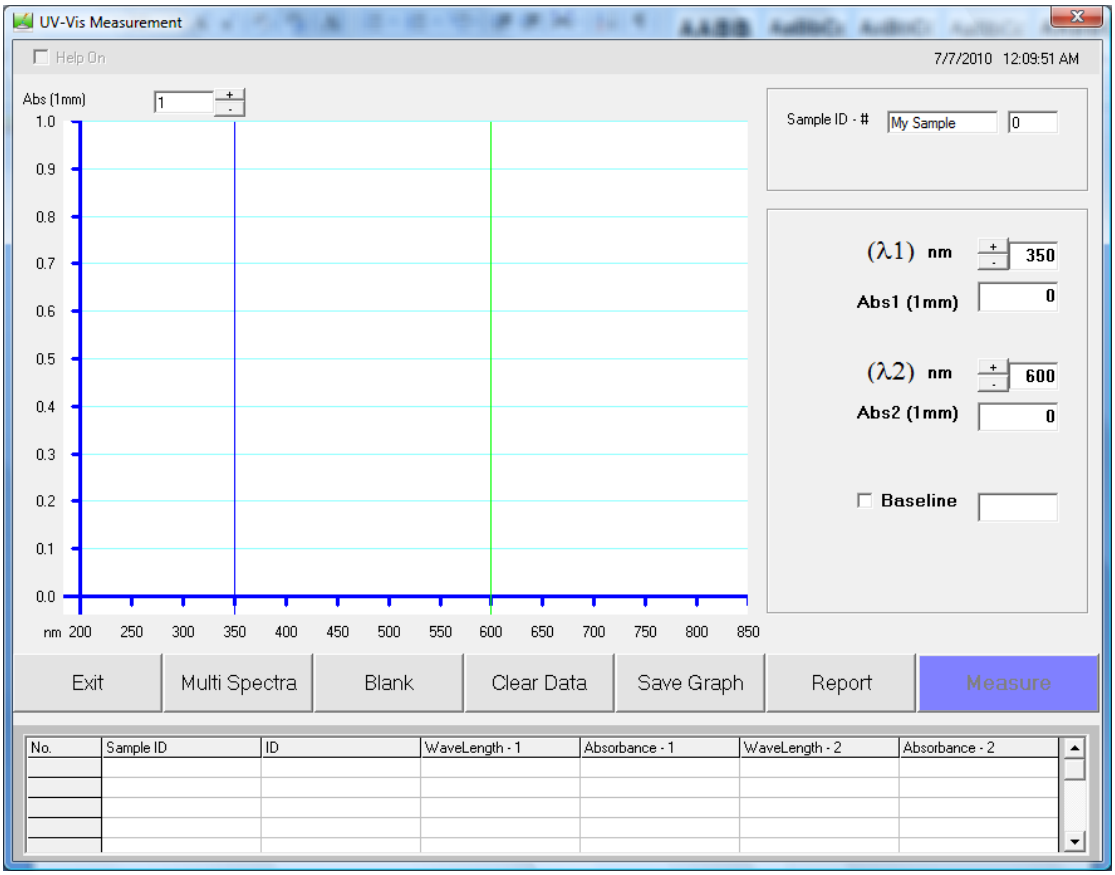
11. UV-VIS MEASUREMENT

Overview

The Q5000 can function as a general-use laboratory spectrophotometer. The UV-Vis measurement module provides the operator with a sample absorbance range from 220 to 850 nm.

UV-Vis Measurement Screen Features

Select UV-Vis from Q5000 main menu and the module interface will appear on the screen.



- **Graph Display**: The Y-axis indicates the absorbance value and the X-axis is for the wavelength. The maximum absorbance value is automatically adjusted to the highest absorbance value. The user is also allowed to manually enter the absorbance value.
- **Sample ID - #:** there are two windows that display the sample ID information. The left window displays the sample ID input by the user. The default is "My Sample". The right window displays the ID number for that sample ID. The ID number begins at 1 and increases by 1 for each measurement. Changing the Sample ID will automatically reset the ID number to 1.
- **(λ1) nm:** select the wavelength by using the up/down arrows.
- **Abs 1 (1mm):** corresponding absorbance at (λ1) nm.
- **(λ2) nm:** select the wavelength by using the up/down arrows.
- **Abs 2 (1mm):** corresponding absorbance at (λ2) nm.
- **Baseline:** if selected, the absorbance value of the baseline is subtracted from the absorbance of Abs1 and Abs2.
- **Vertical Lines:** the green line on the absorbance-wavelength graph indicates the peak position of the wavelength for (λ1)nm, and the blue vertical line indicates the peak position of the wavelength for (λ2)nm.

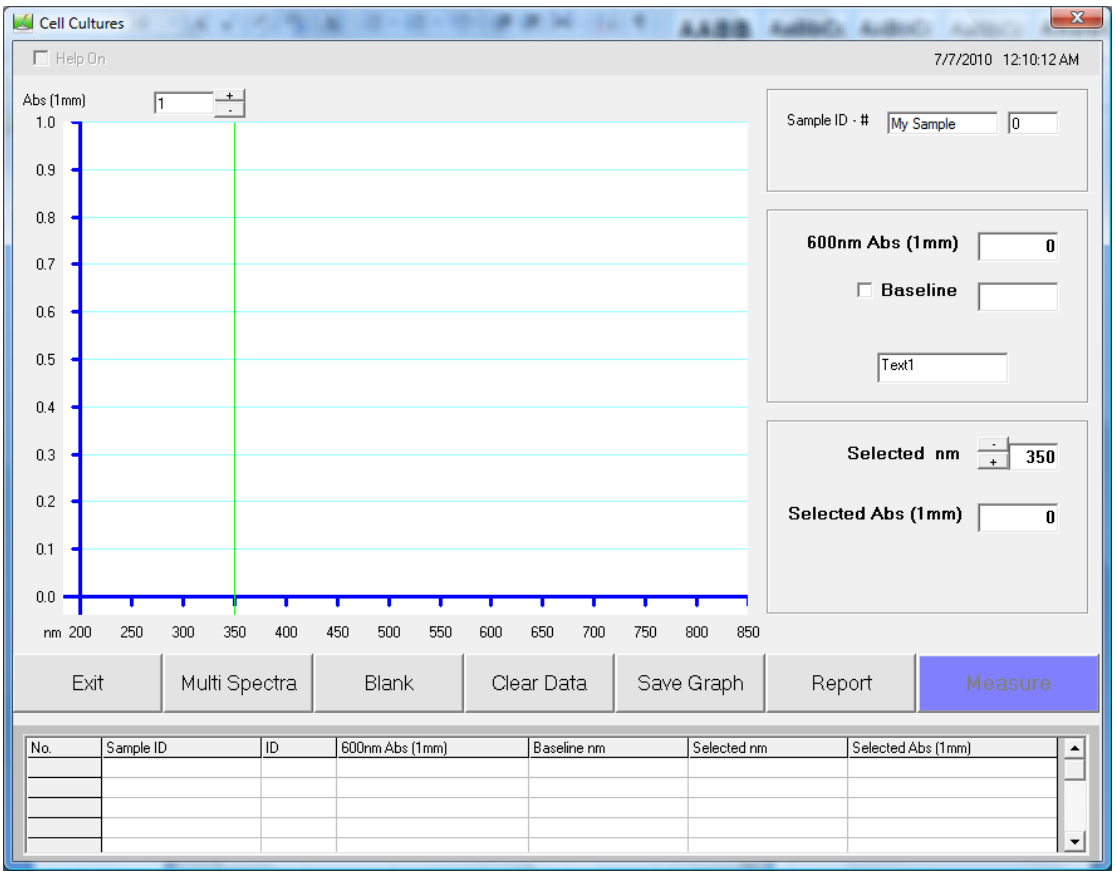
12. CELL CULTURES

Overview

The Q5000 allows laboratories to monitor the density of suspended cell and microbial cultures by measuring their light scatter at 600 nm.

Cell Cultures Screen Features

Select Cell Cultures from Q5000 main menu and the module interface will appear on the screen.



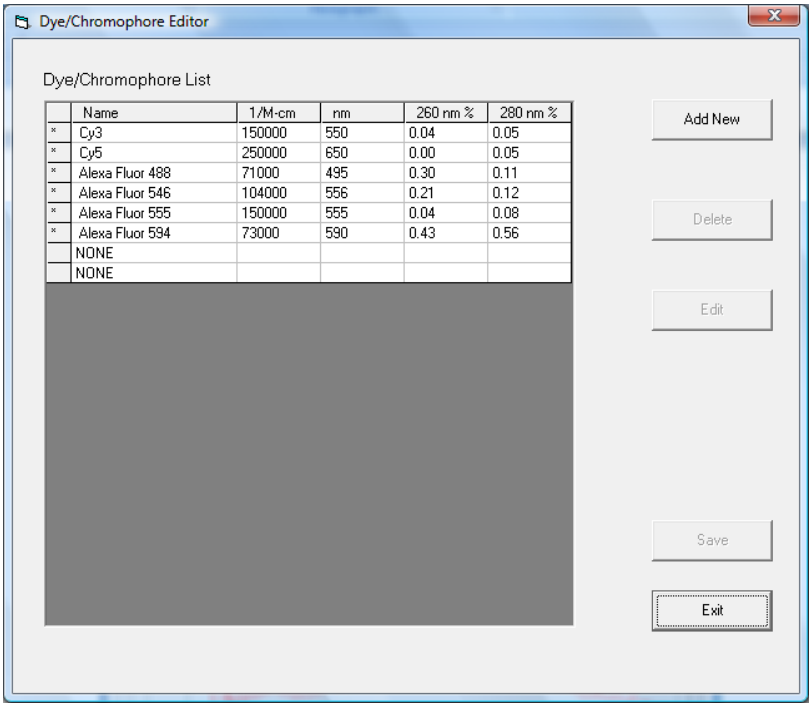
- **Graph Display**: The Y-axis indicates the absorbance value and the X-axis is for the wavelength. The maximum absorbance value is automatically

adjusted to the highest absorbance value. The user is also allowed to manually enter the absorbance value.

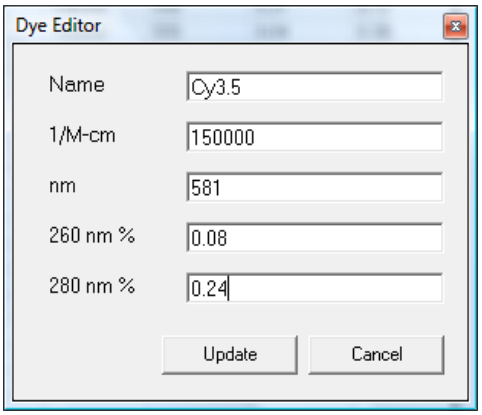
- **Sample ID - #:** there are two windows that display the sample ID information. The left window displays the sample ID input by the user. The default is "My Sample". The right window displays the ID number for that sample ID. The ID number begins at 1 and increases by 1 for each measurement. Changing the Sample ID will automatically reset the ID number to 1.
- **600 nm Abs (1mm):** absorbance at 600nm
- **Baseline:** if selected, the absorbance value of the baseline is subtracted from the absorbance at 600nm.
- **Selected nm:** user select wavelength.
- **Abs (1mm):** absorbance at selected nm.
- **Green vertical line:** indicates the peak position of the wavelength for selected nm.

13. THE PREDEFINED FLUORESCENT DYES LIST

The list works with both the Microarray and Labeled Proteins modules. This list contains a pre-defined list of fluorescent dyes as shown below.

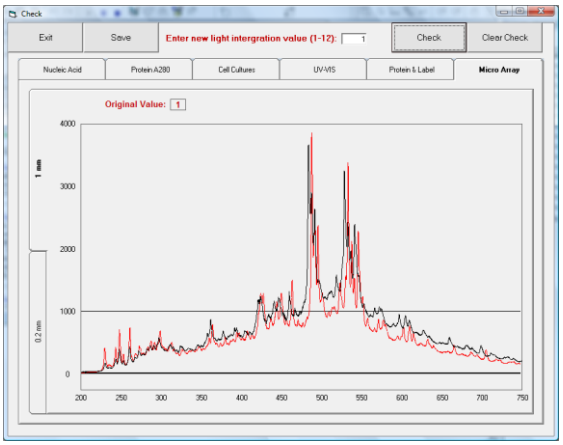


Additional fluorescent dyes can be added by the user as needed.

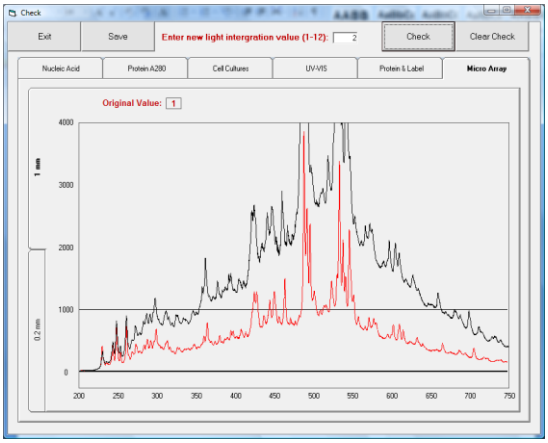


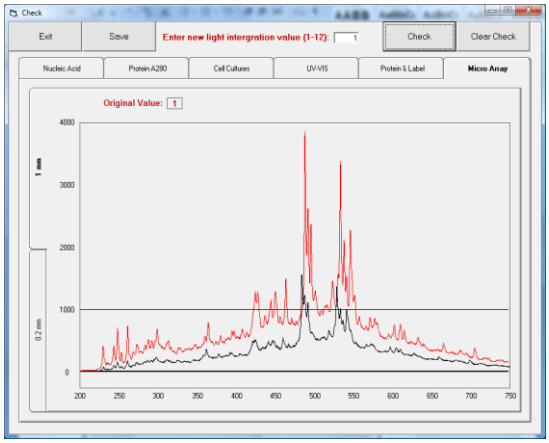
14. LIGHT INTEGRATION

This module is used for adjusting the light integration. The integration number is set to normal before shipping. The light integration and detector systems are functioning normally if the red and black spectra appear as in the image below.



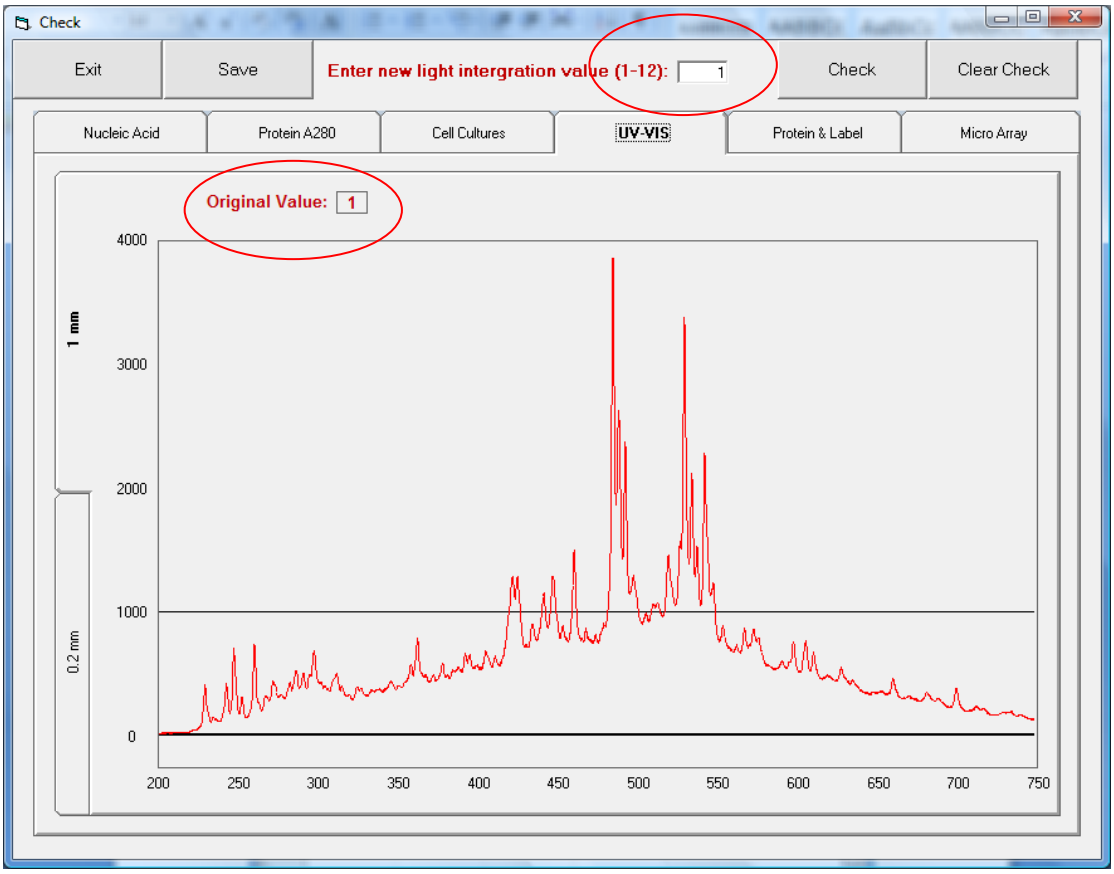
However, if the image appears as below, with the light integration spectrum too high or too low, the light integration can be adjusted with this feature.





Adjusting the light integration:

1. Select the module from the horizontal tab and the path from the vertical tab.
2. The original integration number appears in the window "Original Value".
3. Enter the number in the window "Enter new light integration value (1-12)".
4. Click the "Check" and the black spectrum will display in the window.
5. Increase the integration value from 1 up to12, stopping when the black and red spectra are close.



If you have questions about the light integration, please contact your local distributer or Quawell at info@quawell.com.

15. POTENTIAL SOURCES OF ERROR

Sample Overlap

Sample residue on either measurement surface will affect the result. We suggest that the user:

- Clean the two surfaces with dry soft lab paper after each sample measurement.
- Clean the surfaces with 2 ul de-ionized after measuring highly concentrated samples.
- Thoroughly clean the surfaces and surrounding area with de-ionized water after completing all measurements.
- Before sample measurement blank the machine as described:
    1. Open the appropriate application module.
    2. Lift the upper arm and add 2.5 ul of blanking reagent to the lower surface.
    3. Lower the upper arm and click the Blank button.
    4. Lift the upper arm and clean the blanking reagent from both surfaces with de-ionized water.
    5. Add 2.5 ul blanking reagent to the lower surface.
    6. Lower the upper arm and click the Measure button.
    7. If the value in the Abs260 window is higher than 0.004, repeat these blanking steps.

Sample Homogeneity

Non-homogenous samples would cause significant deviation in the data generated by any measurement system, including the spectrophotometer.

Effect of Evaporation

Sample evaporation could cause a 1-2% deviation in the sample's concentration.

Insufficient Sample Volume

We recommend 2.5 ul for normal sample measurements.

16. TROUBLESHOOTING

USB Connection Error

This error screen usually indicates that the Q5000 and PC connection failed. To correct the error, unplug the USB cable, wait for 2-3 seconds, and plug it again.

Sample Accuracy and Reproducibility

If the sample data are inaccurate and/or not reproducible please refer to Section 13 "POTENTIAL SOURCES OF ERROR**".**

17. MAINTENANCE

The primary maintenance requirement of the Q5000 spectrophotometer is to keep the measurement surfaces clean. Upon completion of a measurement, wipe the sample from the upper and lower surfaces. Clean the surfaces and surrounding area with de-ionized water to prevent sample carryover and residue buildup.

18. APPENDICES

A. Instrument Specifications

Minimum Sample Size	0.5ul
Optic Path Length	0.2-1mm (auto selected)
Light Source	Xenon flash lamp
Wavelength Range	200-850 nm
Wavelength Resolution	1nm
Wavelength Accuracy	1 nm
Absorbance Range	0.01 -100
Absorbance Precision	0.002 Abs (1mm)
Absorbance Accuracy	1%
Detector Type	3648 element linear silicon CCD array
Detection Limit	1ng/ul (dsDNA)
Max Concentration	5,000 ng/ul (dsDNA)
Measurement Cycle	<8 seconds
Dimensions	145 mm x 210 mm
Weight	2 kg
Operating Voltage	12 vdc
Power Consumption	15 W (operating)
Power Consumption	1.5 W (standby)
surface Construction	303 stainless steel and quartz fiber
Software Compatibility	Windows® XP and Vista (32 bit)
CE Approval	Units sold in Europe

B. Absorbance Calculation

When the Q5000 Spectrophotometer is "blanked", a spectrum is taken of a reference material (blank) and stored in memory as an array of light intensities by wavelength. When a measurement of a sample is taken, the intensity of light that was transmitted through the sample is recorded. The sample intensities along with the blank intensities are used to calculate the sample absorbance according to the following equation:

$$Abs = -\log\left(\frac{\mathbf{I}_{sample}}{\mathbf{I}_{blanc}}\right)$$

Thus, the measured light intensity of both the sample and of the blank are required to calculate the absorbance at a given wavelength.

C. Concentration Calculation (BEER'S LAW)

The Beer-Lambert equation is used to correlate the calculated absorbance with concentration:

$A = E * b * c$

Where A is the absorbance represented in absorbance units (A), E is the wavelength-dependent molar absorptive coefficient (or extinction coefficient) with units of liter/mol-cm, b is the path length in cm, and c is the analyze concentration in moles/liter or molarities (M).

17. CONTACT INFORMATION

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