

SpectraSuite Spectrometer Operating Software

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About This Manual

Document Purpose and Intended Audience

This document provides you with an installation and configuration instructions to get your system up and running. In addition, user interface for SpectraSuite is provided.

Document Summary

Chapter	Description
Chapter 1: SpectraSuite Introduction	Provides an overview of the SpectraSuite operating software.
Chapter 2: SpectraSuite Installation	Contains instructions for installing and configuring the SpectraSuite software.
Chapter 3: File Menu Functions	Contains descriptions of the functions on the File Menu.
Chapter 4: View Menu Functions	Contains a description of the functions on the View Menu.
Chapter 5: Spectrometer Menu Functions	Contains descriptions of the functions on the Spectrometer Menu.
Chapter 6: Processing Menu Functions	Contains descriptions of the functions on the Processing Menu.
Chapter 7: Strip Charts	Provides instructions for using the Strip Charts feature.
Appendix A: <u>Experiment Tutorials</u>	Contains instructions for performing sample experiments using an OceanOptics spectrometer and SpectraSuite software.

Product-Related Documentation

- External Triggering Options
- Various USB spectrometer documents such as the *HR4000 and HR4000CG-UV-NIR Series Installation and Operation Manual.*

You can access documentation for Ocean Optics products by visiting our website at <u>http://www.oceanoptics.com</u>. Select *Technical* \rightarrow *Operating Instructions*, then choose the appropriate document from the available drop-down lists. Or, use the **Search by Model Number** field at the bottom of the web page.



You can also access operating instructions for Ocean Optics products on the *Software and Technical Resources* CD included with the system.

Engineering-level documentation is located on our website at *Technical* \rightarrow *Engineering Docs*.

Upgrades

Occasionally, you may find that you need Ocean Optics to make a change or an upgrade to your system. To facilitate these changes, you must first contact Customer Support and obtain a Return Merchandise Authorization (RMA) number. Please contact Ocean Optics for specific instructions when returning a product.



Chapter 1

SpectraSuite Introduction

Product Overview

SpectraSuite is a completely modular, Java-based spectroscopy software platform that operates on Windows, Macintosh and Linux operating systems. The software can control any Ocean Optics USB spectrometer and device, as well as any other manufacturer's USB instrumentation using the appropriate drivers. The SpectraSuite interface looks and feels the same on all operating systems yet retains the familiar appearance of an application native to each OS. Ocean Optics is the first to offer such a flexible, feature-packed application with this level of cross-platform capability.

The SpectraSuite Java-based framework is modular and every function in it can be altered or replaced. For instance, the data acquisition functions, the scheduling functions, the data processing functions and the rendering functions are all separate modules. You can add or delete modules to create a proprietary user interface or functionality; create modules to perform calculations; automate experiment routines and more. You or an Ocean Optics application developer can easily customize SpectraSuite through Java code.

SpectraSuite is the platform for all future Ocean Optics application software development. Current software applications including OOIChem, OOISensors, OOIColor and OOIIrrad-C will be migrated to the SpectraSuite platform in the near future.

USB Spectrometer and Device Control

SpectraSuite easily manages multiple USB spectrometers – each with different acquisition parameters – in multiple windows, and provides graphical and numeric representation of spectra from each spectrometer. Using SpectraSuite, you can combine data from multiple sources for applications that include upwelling/downwelling measurements, dual-beam referencing and process monitoring.

SpectraSuite can be used with the following Ocean Optics spectrometers when they are interfacing to a PC through their USB port:

- USB2000 Spectrometer
- USB4000 Spectrometer
- HR2000 High-resolution Spectrometer
- HR4000 High-resolution Spectrometer



- HR2000+High-resolution Spectrometer
- QE65000 Scientific-grade Spectrometer
- NIR-512 Near-IR Spectrometer
- NIR256 Near-IR Spectrometer

Features

- **Modular Framework** The SpectraSuite framework is modular and every function in it can be altered or replaced. For instance, the data acquisition functions, the scheduling functions, the data processing functions and the rendering functions are all separate modules. You can add or delete modules to create a proprietary user interface or functionality; create modules to perform calculations; automate experiment routines, and more. You or an Ocean Optics application developer can easily customize SpectraSuite through Java code.
- **Platform Independent** The SpectraSuite is a platform-independent application that provides Graphical and numeric representation of spectra in one window. SpectraSuite can operate on a Windows, Linux, or Macintosh operating system.
- Advanced Data Capture Control SpectraSuite provides the user with advanced control of episodic data capture attributes. For instance, a user can acquire data for a fixed number of scans or for a specific interval. Initiation of each scan can be externally triggered or event-driven. Captured data is quickly stored into a systems memory at speeds as fast as 1 scan per msec with speeds limited by hardware performance.
- 21 CFR Part 11 Compliant SpectraSuite is 21 CFR Part 11 compliant with an encoded binary file. This binary data format tracks the complete history of all processing steps that are performed on the data. Spectra suite stores and provides data in a variety of other formats including tabdelimited ASCII (for Excel or other analysis packages), Grams SPC, and JCAMP. Additionally, Spectra Suite offers a database module where arbitrary data can be stored in any user-selectable format. Another benefit of the software's binary data tracking function is that it offers a user "process do-overs." A user can change the values of various parameters in a process and apply those changes to data without having to recreate an entire process.
- **Fully Internationalized** All of the software's menus, dialog boxes, prompts, messages and files can reflect a native language by simply changing a single file. Currently, modules that support English, German, Japanese and Russian are available.

Spectroscopic Functions

OOIBase32 allows you to perform the three basic spectroscopic experiments – absorbance, reflectance and emission – as well as signal-processing functions such as electrical dark-signal correction, stray light correction, boxcar pixel smoothing and signal averaging. Scope mode, the spectrometer operating mode in which raw data (signal) is acquired by the detector, allows you to establish these signal-conditioning parameters. The basic concept for the software is that real-time display of data allows users to evaluate the effectiveness of their experimental setups and data processing selections, make changes to these parameters, instantly see the effects and save the data. Most spectrometer-system operating software does not allow such signal-conditioning flexibility.



With OOIBase32, you can perform time-acquisition experiments for kinetics applications. As part of the time-acquisition function, you can monitor and report up to 6 single wavelengths and up to 2 mathematical combinations of these wavelengths. In addition, you can perform reference monitoring in a variety of ways: single wavelength (1 or 2 channels), integrated intensity (starting and ending wavelengths for 1 or 2 channels) and wavelength-by-wavelength (2 channels).

OOIBase32 gives you complete control of setting the parameters for all system functions such as acquiring data, designing the graph display, using spectra overlays and configuring the cursor. You can also save and then retrieve all of these system parameters for future experiments. OOIBase32 has the benefit of providing various software-controlled triggering options for external events such as laser firing or light source pulsing.

Other advanced features give you several data-collection options. You can independently store and retrieve dark, reference, sample and processed spectra. All data can be saved to disk using autoincremented filenames. You can save data as ASCII files or in the native GRAMS/32 SPC format. One feature prints the spectra and another copies spectral data into other software such as Excel and Word.

Other OOIBase32 extras include the ability to monitor the status bar for each spectral window, which reflects numerous parameters set by the user; the ability to manipulate the placement of an array of dockable toolbars; and the ability to choose sound cues for a variety of spectroscopic events. You can also designate how to receive data acquisition warnings such as when the Scope mode signal has saturated in absorbance, transmission and irradiance modes. In addition, the time-normalization function allows you to designate separate integration times for reference and sample scans.

Getting Updates

SpectraSuite software features one year of free, web-based automatic upgrades when you buy the software. This service requires that you register the first time you connect. You can purchase renewals for half the software price per year. All customers (whether they have a free upgrade plan or not) will be able to get free bug fixes.

To obtain updates, select **Tools | Update Center** from the SpectraSuite menu.



Chapter 2

SpectraSuite Installation

Overview

The following sections will guide you in installing SpectraSuite on either a Windows, Macintosh or Linux operating system.

Note

Do NOT connect the spectrometer to the PC until you install the SpectraSuite software. Follow the instructions contained in this chapter to properly connect and configure your system.

Installing SpectraSuite

This section contains instructions for installing SpectraSuite on each of the following operating systems:

- Microsoft Windows Windows 98, 2000, XP, Me
- Apple Macintosh OS X version 10.0 or later
- Linux Red Hat 9 or later, Fedora (any version), Debian 3.1 (Sarge), and SUSE (9.0 or later)

Installing on a Windows Platform

Total download is approximately 30 MB.

Procedure

- 1. Close all other applications running on the PC.
- 2. Type <u>ftp://spectrasuite.oceanoptics.com</u> into the browser.
- 3. Right-click on any white space in the browser window and select **Login As...**. The login and password dialog box appears.



2: SpectraSuite Installation

4. Enter the following:

Username: photonics

Password: spectrasuite

- 5. Double-click on **Installations**.
- 6. Drag WindowsSetup.exe to the local PC and execute.

Installing on a Macintosh Platform

Total download is less than 15 MB.

► Procedure

- 1. Open a terminal window (Finder | Applications | Utilities | Terminal).
- 2. Paste the following text into this terminal window:

```
ftp ftp://photonics:spectrasuite@spectrasuite.oceanoptics.com
hash 10000
lcd ~/Desktop
cd Installations
get SpectraSuite.dmg
quit
```

When this command is finished, a file named **SpectraSuite.dmg** appears on the desktop.

- 3. Double-click on the SpectraSuite.dmg file to mount the disk image.
- 4. Drag the SpectraSuite icon from the disk image to where you want to install the application.

Installing on a Linux Platform

Total download is approximately 30 MB.

Procedure

- 1. Open a terminal window.
- 2. Paste the following text into the terminal window:

ftp ftp://photonics:spectrasuite@spectrasuite.oceanoptics.com hash 10000 lcd ~/Desktop cd Installations



get SpectraSuiteSetup_Linux.bin quit

A SpectraSuiteSetup_Linux.bin file appears on your desktop.

3. Start a terminal window and enter the following commands:

chmod 755 ~/Desktop/SpectraSuiteSetup_Linux.bin sudo ~/Desktop/SpectraSuiteSetup_Linux.bin

4. You are prompted for your password, which allows you to execute the setup as root. Contact your system administrator if you do not have password.

If the sudo command does not work (it may not be set up for your user account), then enter the following:

su (enter password for root) ~/Desktop/SpectraSuiteSetup_Linux.bin

Notes

The default installation directory is /usr/local/OceanOptics/SpectraSuite.

A symbolic link is put in /usr/bin so that you can enter **spectrasuite** on any command line to start the program.

The SpectraSuite icon () location varies by installation, but will be under either Applications or Other under the Application Launcher menu.



Chapter 3

File Menu Functions

Overview

This chapter details the various options and functions available from the File menu in SpectraSuite.

Where applicable, each section contains the associated toolbar icon below the section heading. Click on these icons in SpectraSuite to perform the described function.

New

The New menu selection displays the following list of functions that also are available from the Data Display toolbar:

- Spectrum Graph
- Spectrum Table
- High Speed Acquisition
- Absolute Irradiance Graph
- Strip Chart

Spectrum Graph

The Spectrum Graph menu option displays a new curve of spectral data on the open graph based on the parameters set in the Acquisition toolbar. Each new curve you create appears as a different color in the graph. Graphs appear in scope mode by default.

If you click on a curve, its corresponding parameters appear in the Acquisition toolbar. The data source (spectrometer) is listed in the Source box.



Procedure

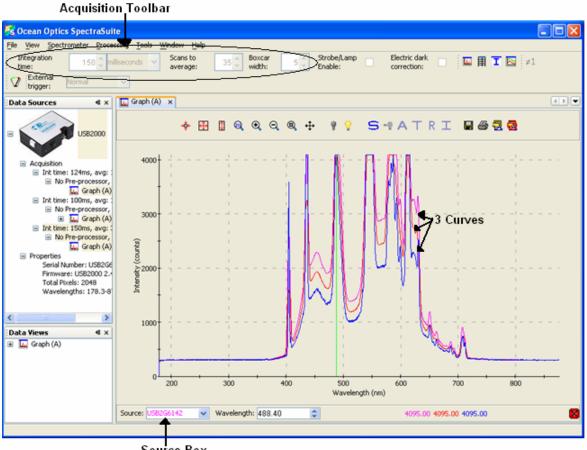
To display a spectral graph, follow the steps below:

- 1. Set the desired parameters (integration time, scans to average, and boxcar width).
- Select File | New | Spectrum Graph 2.

Or

Click in the Data Display toolbar. The curve displays in the graph pane.

The sample below shows a graph with three spectral curves (blue, pink and orange).



Source Box



Spectrum Table

围

The Spectrum Table menu selection displays a spectral graph in tabular format. Select File |

Spectrum Table from the menu or click in the Data Display toolbar. The spectrum table appears.

File View Spectrometer Proce	ssing Tools Window Help		
Integration time: 150 0 External trigger: Fiormal	Alseconds V Scans to average: 35.0 Boxcar 5.0	Strobe/Lamp Electric dark Enable: Correction:	
Data Sources 4 ×	Graph (A) x Table (B) x		
Acquisition Acquisition Acquisition Int time: 124ms, a. No Pre-process No Pre-proce	Wavelength (nm) 178.32 178.70 179.06 179.46 179.84 180.22 180.60 180.96 181.36 181.74 182.12 182.50 182.67 183.63 184.77 185.15 185.53 185.91 186.29 186.67 187.04 187.42	Intensity (counts) 0.00 284.00 289.00 289.00 291.00 293.00 293.00 294.00 294.00 293.00 294.00	
	187.80 188.19 188.56 188.94 189.32	294.00 299.00 297.00 301.00 298.00	-

High-Speed Acquisition

^{SUse} this menu selection if you want to capture every scan at a very fast rate (20 ms or faster). SpectraSuite performs this high-speed data acquisition by capturing raw data without slowing it down by processing it. This captured high-speed data is called a spectra collection.

A spectra collection can be viewed in either a graph or table format. You can also record the captured data in a 3-dimensional graph, and then run other measurements on this captured data.

It's best to store your reference and dark spectra using the regular graph window before performing a high-speed data acquisition. The following procedure assumes that you do so.



High-Speed Data Capturing

► Procedure

- 1. Save reference spectrum to a file. See Reference.
- 2. Save a dark spectrum to a file. See Dark.

Caution

To ensure accurate data, be sure that you have selected the same parameters (integration time, scans to average, and boxcar width) for your high-speed acquisition that you used when capturing your dark and reference spectra.

3. Select **File** | **New** | **High Speed Acquisition**. If you have other acquisitions running, a warning message appears telling you that all other acquisitions will be stopped when you are acquiring high speed data. Either stop all your other acquisitions, or click **OK** on this message and SpectraSuite will temporarily stop them. The **High-speed Acquisition** dialog box appears.

🔀 High-speed Acquisition 🛛 🛛 🔀	
Integration Time (usec): 15000	
Scans to Average: 1	\sum
Boxcar Width: 0	Parameters
External Trigger Mode: Normal	/
Electric Dark Correction: 🔲	
Strobe/Lamp Enable: 🗌 🚽	
Filename:	Enter Filename
Number of Scans: 100	
Capture Period (usec): 5000000	>Select one
Show progress 🗹	
Done. 100 scans took 15.2 ms each.	
(**************************************	Acquisition Progress
	riogress
	Click
Go Close	-



The top third of the form lists the parameters selected in the main graph window(???).

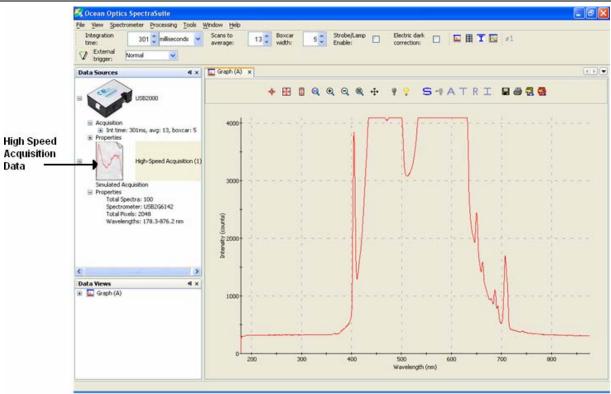
4. Enter the following information:

Field	Selection	
Integration Time (µsec)	Specify the integration time in µsec.	
Scans to Average	Enter the number of scans to average	
Boxcar Width	Enter the boxcar width	
External Trigger Mode	Select the external trigger mode (Normal, Software, Synchronization, External Hardware)	
Electric Dark Correction	Enable this option for electric dark correction	
Strobe/Lamp Enable	Enable this option to turn on the lamp	
Filename	Enter the name or browse to the file where you want the high speed acquisition data stored.	
	Choose one of these methods of data acquisition	
Number of Scans	Acquire a fixed number of scans (fastest method)	
Or		
Capture Period (µsec)	Specify a time period for data capture	
Show progress	Enable this option to show the progress of the acquisition.	

5. Click . Your high-speed acquisition data appears in the **Data Sources** pane.



3: File Menu Functions





- 6. Right-click on . A menu displays the following selections:
 - Spectrum Graph Displays a graph of the spectra

View Spectrometer Processing Tools Window										
External Normal	rage:	Boxcar width:	0 Strob Enable	e/Lamp e:	Electric dar correction:	^k 🔲 📗		≠1		
ta Sources 4 ×	🛴 Graph (A) 🗙									•
Acquisition Int time: 20ms, avg: 1, boxcar: 0		÷ 🕀 🖸	Q Q	ର୍ ଷ୍ ↔	Y Y	S-1 A	TRI	864	2 2	
No Pre-processor, Scope Mode	†/									
	140	1. Carlos (1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1	n an aire an an		e e ele el	1 				
High-Speed Acquisition (1)	t And									
	120									
High-Speed Acquisition (2)	<mark>-</mark>)									
Tightspeed Acquisition (2)	100	1	E.			1			1	
	100									
	i (j									
High-Speed Acquisition (3)	Intensity (counts)									
	Aji .									
 Simulated Acquisition Spectrum 100/100 	luter									
No Pre-processor, Scope Mode	60min		е е е езе 12							
🕀 🔛 Graph (A)	÷,									
ta Views 4 ×	40									
🖾 Graph (A)	10 1									
	t l									
	20-1									
	T)									
	0 <mark>+++++</mark> 200	300	400	500	600 Wavelenç	700 1th (nm)	800	900	1000	110



Wavelength (nm)	Intensity (counts)	Wavelength (nm)	Intensity (counts)
178.32	0.00	178.32	0.00
178.70	283.00	178.70	288.00
179.08	282.00	179.08	289.00
179.46	284.00	179.46	283.00
179.84	282.00	179.84	287.00
180.22	286.00	180.22	288.00
180.60	288.00	180.60	287.00
180.98	294.00	180.98	289.00
181.36	293.00	181.36	292.00
181.74	291.00	181.74	294.00
182.12	289.00	182.12	291.00
182.50	291.00	182.50	293.00
182.87	294.00	182.87	292.00
183.25	289.00	183.25	289.00
183.63	292.00	183.63	292.00
184.01	296.00	184.01	293.00
184.39	295.00	184.39	295.00
184.77	291.00	184.77	292.00
185.15	293.00	185.15	294.00
185.53	293.00	185.53	292.00
185.91	292.00	185.91	293.00
186.29	297.00	186.29	287.00
186.67	288.00	186.67	292.00
187.04	294.00	187.04	292.00
187.42	293.00	187.42	290.00
187.80	295.00	187.80	291.00
188.18	295.00	188.18	289.00
188.56	293.00	188.56	293.00
188.94	287.00	188.94	294.00

• Spectrum Table – Displays spectra data in a table format

- Save Spectra Collection Stores the high-speed spectra in a file. See <u>Creating a 3-D Chart</u> <u>of High-Speed Acquisition Spectra</u> for more information about what can be done with stored high-speed spectra data.
- **Remove Spectra Collection** Deletes the captured high-speed spectra collection data. You can also select **Spectrometer** | **Remove Spectra Collection**.

Creating a 3-D Chart of High-Speed Acquisition Spectra

You can create a 3-dimensional representation of the data over time in graph form. The navigation controls enable you to view the graph from different angles. Click on any point in the graph to display its value and wavelength.

► Procedure

To create a 3-D graph,

1. Right-click on the spectrometer in the Data Sources pane and select High Speed Acquisition

Or

Select **File** | **New** | **High Speed Acquisition** from the menu. The **High-speed Acquisition** dialog box appears.



Kigh-speed Acquisition	×			
Integration Time (usec): 15000]			
Scans to Average: 1				
Boxcar Width: 0				
External Trigger Mode: Normal	~			
Electric Dark Correction: 📃				
Strobe/Lamp Enable: 📃				
Filename:				
 Number of Scans: 100 				
Capture Period (usec): 5000000				
Show progress 🔽				
Done. 100 scans took 15.2 ms each.				
Go Close 🔗				

2. Enter the parameters and click *M*. The 3-dimensional graph appears.

Using High-speed Acquisition Spectra

A spectra collection that you have stored using the high-speed acquisition feature have a variety of uses such as the following:

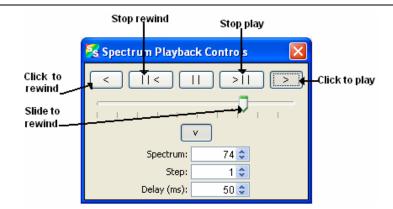
- You can replay the spectra, fast forward, rewind, etc. just like any recorded data
- You can also conduct further experiments on the recorded data (absorbance, transmission, etc.) by clicking on the appropriate icon (A, T, R, or I).

► Procedure

To replay recorded data,

- 1. Right-click on the desired Spectrum listed under **Simulated Acquisition** in the **Data Sources** pane. A menu appears.
- 2. Select Spectrum Playback Controls. The Spectrum Playback Controls dialog box appears.



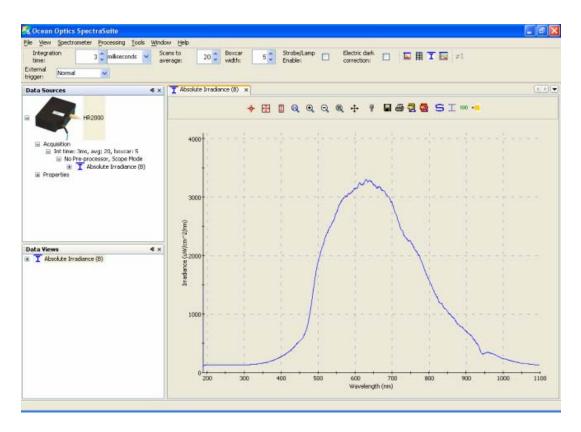


3. Rewind the recording, then press to play the recorded spectrum graph.

Absolute Irradiance Graph

The Absolute Irradiance Graph menu selection displays the Absolute Irradiance graph. Select File

Absolute Irradiance Graph from the menu or click on the Irradiance toolbar. The Absolute Irradiance Graph in Scope mode appears. See <u>Absolute Irradiance</u> in Appendix A: Experiment Tutorials for more information.



3: File Menu Functions



Strip Chart

SpectraSuite allows you to use Strip Charts to track processes, perform kinetic analyses, and monitor spectral events all as a function of time. The Strip Chart shows you how the value that you selected appears over time. This value can be any of the following:

- One pixel (a single wavelength)
- The average of a range of pixels
- An integral calculated by one of three methods representing the area beneath the graph's trend line

You can acquire this data using any processing mode (transmission, absorbance, etc.).

► Procedure

- 1. Place SpectraSuite in Scope mode by clicking the Scope (S) icon in the Experiment mode toolbar or selecting **Processing | Processing Mode | Scope** from the menu.
- 2. Store a reference spectra and dark spectra.
- 3. Choose the measurement mode (absorbance, transmission, etc.).
- 4. Click the Strip Chart button (\square)

OR

Select File | New | Strip Chart from the menu. The Chart Trend Settings dialog box opens.

🔀 Chart Trend Settings	×
# Spectrometers Spectrum Type 0 HR2B1231 Processed	Wavelength Selection One wavelength: 188.80 mm Average from 188.80 mm to 188.80 mm
Display Options Trend Line Color: Scale Trend Value:	O Integrate over 188.80 ♀ to 188.80 ♀ nm Method: Rectangular ♥
Multiply by 1 then add 0 Minimum delay between updates: 0 🗢 ms	Accept Cancel

5. Enter the following data to create your Strip Chart:





Field	Selections	
Wavelength Selection	Select the type of data for which you want to view the time trend. Select from the following:	
	 One wavelength – For one pixel (used in fluorescence, for example). Select he wavelength. 	
	 Average from – For a range of pixels. Select the starting and ending wavelengths. 	
	 Integrate over – For the area underneath the trend line. Select the starting and ending wavelengths. Then, select the method for calculating the continuous area underneath the graph curve: Rectangular, Simpson's, or Trapezoid. 	
Display Options	Select a color for the graph's trend line.	
Scale Trend Value	If you would like to adjust your data, enter the appropriate information in this field.	
Minimum delay between updates	Set the amount of time (in milliseconds) between data points on the graph.	
	NOTE: The longer the time between updates, the less computer memory that is used.	

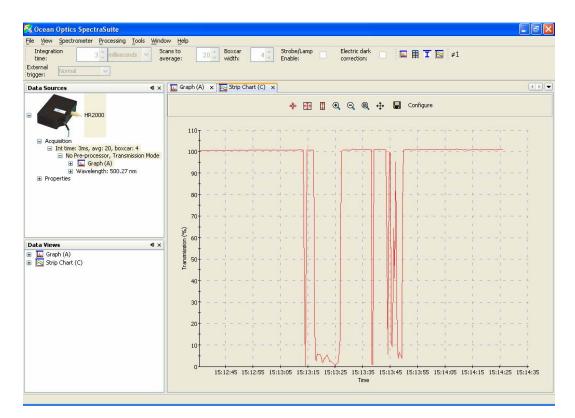
6. Click Accept . The Strip Chart Options dialog box appears. Strip Chart Options lists all of the Strip Charts that you have created, along with the options that you have set for each one. Use this dialog box to add, edit, remove, or clear trends.

Note

Highlight the desired trend to make the **Clear**, **Remove Trend**, and **Edit Trend** buttons active.



3: File Menu Functions



Open

Selecting the **Open** menu option allows you to load previously stored dark spectra, reference spectra, or all stored all spectra files. Select the type of spectrum to load, then browse to where the file is stored.

Dark

A dark spectrum is a spectrum taken with the light path blocked.

Select File | Open | Load Dark Spectrum to select and open one or more dark spectra.

Reference

A reference spectrum is a spectrum taken with the light source on and a blank in the sampling region.

Select File | Open | Load Reference Spectrum to select and load one or more reference spectra.



Spectrum Collection

A spectrum collection is created when you perform a high-speed acquisition. Select **File** | **Open** | **Load Spectrum Collection** to browse to and open a spectrum collection from a high-speed acquisition.

Save

This menu option allows you to save your spectra collection to a file. Select **File** | **Save** | **Save Spectra Collection** from the menu and enter a file name.

Store

Use this menu selection to store dark and reference spectra before running your experiment.

Dark

A dark spectrum is a spectrum taken with the light path blocked.

Select **File** | **Store** | **Store Dark Spectrum** from the menu to save a dark spectrum to a file. You can also click **?** in the graph toolbar.

Reference

A reference spectrum is a spectrum taken with the light source on and a blank in the sampling region.

Select **File** | **Store** | **Store Reference Spectrum** to store a reference spectrum file. You can also click in the graph toolbar.

Exit

Select **File** | **Exit** from the menu to exit the SpectraSuite application. A shutdown confirmation dialog box appears. Click **Yes** to exit SpectraSuite.

Alternately, you can click the top right X box of the application's display window.



Chapter 4

View Menu Functions

Overview

This section details the various options and functions available from the View menu in SpectraSuite.

Scale

Empty

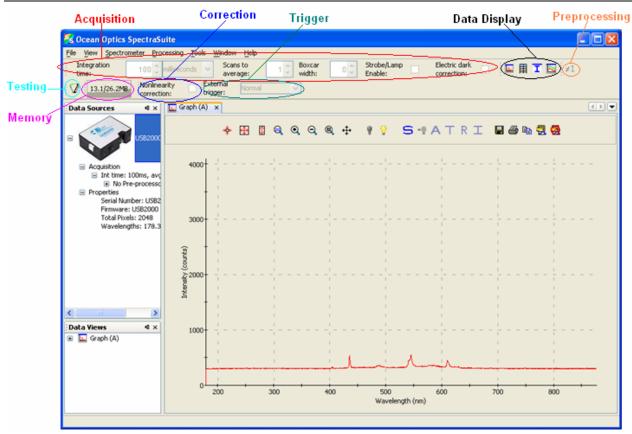
This menu selection is for future use.

Toolbars

This menu item controls which toolbars are visible on the SpectraSuite window. Click on the following selections to toggle the toolbar on and off. When a toolbar is visible, a checkmark appears next to the menu selection. The toolbars available from this menu item are shown on the SpectraSuite window in the figure below:



4: View Menu Functions



Acquisition

Select **View** | **Acquisition** to display/hide the tools related to the data being acquired and displayed in SpectraSuite.

Tool	Function
Integration time: 3 milliseconds	Specifies the integration time of the spectrometer, which is analogous to the shutter speed of a camera. The higher the integration time, the longer the detector monitors the incoming photons. If your Scope mode intensity is too low, increase this value. If the intensity is too high, decrease this value. Adjust the integration time so that the greatest amount of light that you anticipate for your application causes a signal of about 3500 counts. While watching the graph trace, adjust the integration time until the signal intensity level is approximately 3500 counts. The integration time specified controls enabled spectrometer channels in the active spectral window.



Tool	Function
Scans to 1	Specifies the number of discrete spectral acquisitions that the device driver accumulates before SpectraSuite receives a spectrum. The higher the value, the better the signal-to- noise ratio (S:N). The S:N will improve by the square root of the number of scans averaged.
Boxcar 0	Sets the boxcar smoothing width, a technique that averages across spectral data. This technique averages a group of adjacent detector elements. A value of 5, for example, averages each data point with 5 points to its left and 5 points to its right.
	The greater this value, the smoother the data and the higher the signal-to-noise ratio. If the value entered is too high, a loss in spectral resolution will result. The S:N will improve by the square root of the number of pixels averaged.
Strobe/Lamp Enable:	Enables or disables the spectrometer strobe control function.
	This function toggles the S0 line of the spectrometer.
Electric dark correction:	Enables or disables the correction of the spectral data for electrical dark signal.
	The first 24 pixels in the spectrometer, while producing an electrical signal, do not respond to light. This option subtracts the average value of these first 24 pixels from the entire spectrum.

Correction

This toolbar has one tool: the nonlinearity correction checkbox. This tool enables/disables the detector linearity feature. The Correction toolbar only displays when you select it with **View** | **Toolbars** | **Correction**; it is absent from the screen by default.

Data Display

The Data Display toolbar contains the icons for the following graph functions:

lcon	Function
	Spectrum Graph . Displays a new spectrum curve on the current graph with the parameters selected in the Acquisition toolbar. Each spectrum curve appears on the graph in a different color. The Acquisition parameters for each curve are listed in the Data Sources pane.

4: View Menu Functions



lcon	Function
Ħ	Spectrum Table . Displays the data acquisition in a table format. A tab for the table appears at the top pf the graph pane. See <u>Spectrum Table</u> for more information.
Ī	Absolute Irradiance Graph. Displays a graph of absolute irradiance data. This graph appears on its own page with a tab at the top of the graph pane. It is also listed in the Data Views section.
2	Strip Chart. Allows you to track track processes, perform kinetic analyses, and monitor spectral events all as a function of time. See <u>Strip Charts</u> for more information.

Memory

The Memory toolbar consists of a display of the memory being used by the SpectraSuite application

(7.1/22.6MB). Click this icon to force SpectraSuite to clean unneeded data from memory. The Memory toolbar only displays when you select it with **View** | **Toolbars** | **Memory**; it is absent from the screen by default.

Preprocessing

The Preprocessing toolbar contains the nonunity correction feature $(\neq 1)$. Nonunity correction is a preprocessing function that performs a calculation to compensate for a reflection standard that is not perfectly white. See <u>Preprocessing</u>.

Testing

The Testing toolbar consists of the speed benchmark (D). This feature allows you to see how fast the SpectrSuite application is running (how long it takes from the time the data is acquired until all the calculations have been performed. The Testing toolbar only displays when you select it with **View** | **Toolbars** | **Testing**; it is absent from the screen by default.

Trigger

The External Trigger selection box on the Trigger toolbar enables you to set the following trigger mode of the spectrometer:

- Normal free running
- Software Integration time is controlled by frequency of triggers
- External Hardware Hardware fixed or jumpered integration time



Small Toolbar Icons

For future use.



Chapter 5

Spectrometer Menu Functions

Overview

This chapter details the various options and functions available from the **Spectrometer** menu in SpectraSuite.

Acquisition

This menu selection consists of options that control starting and stopping SpectraSuite's acquisition of spectral data.

Pause Acquisition/Resume Acquisition

These menu selections toggle to pause and restart data acquisition. When you start the SpectraSuite application, it comes up acquiring spectral data in Scope mode from the attached spectrometers. If you are monitoring more than one spectrometer, select the desired spectrometer in the Data Sources pane, and choose either **Spectrometer** | **Acquisition** | **Pause Acquisition** to temporarily pause the acquisition. Select **Spectrometer** | **Acquisition** | **Resume Acquisition** to resume the acquisition.

Terminate Acquisition

To completely stop spectral acquisition on a spectrometer, select the desired spectrometer in the Data Sources pane (if you are monitoring more than one), then select **Spectrometer** | **Acquisition** | **Terminate Acquisition**.

Spectrometer Features

This option provides access to additional functionality available with the attached OOI spectrometers. See the documentation for your spectrometer to learn more about the available options and their settings. You must select a spectrometer in the **Data Sources** pane to make this menu selection active.



HR4000/HR2000+ Features

This option provides access to additional functionality available with HR4000, HR2000+, and QE65000 Spectrometers.

Available options on the **HR4000/HR2000+ Features** dialog box include the following:

Option	Description		
	The options in this field enable you to set the timing for a single strobe from the spectrometer's light source. The timing of the single strobe is based on the beginning of the integration period.		
Single Strobe Timing	 High Transition Delay Counter – This option determines a single strobe's delay from the start of the acquisition period expressed in units of one master clock tick. 		
	• Low Transition Delay Counter – This option determines how long the single strobe single will remain high by setting the delay from the start of an acquisition period until the single strobe signal goes low.		
Continuous Strobe Timing	This field determines the length of time between 2 flashes of the continuous strobe signal. The Base Clock Divisor divides down the 48 MHz master clock and feeds the Strobe Divisor . The resulting delay between flashes is shown next to the Strobe Divisor edit box. For the spectrometer to acquire accurate data, the integration time must be a multiple of the time shown next to the Strobe Divisor box.		
External Trigger	This setting sets the delay between the assertion of an external trigger and the start of the spectrometer integration period. The base frequency of this counter is the master clock frequency.		
	 This field corresponds to the 10 GPIO pins in the spectrometer. Alt. Function – Allows OOI functionality to take control of the enabled pin(s). A pin is enabled if its corresponding box is checked. 		
General Purpose	 Pin is Output – If the box is checked, then the pin is configured as output. 		
Input/Output (GPIO)	 Value – If the corresponding pin in the Pin is Output field (above) is configured as output (checked), then a check in the Value field for the same pin indicates that the pin's value is high. Otherwise, the pin's value is low. 		
	If the corresponding pin in the Pin is Output field (above) is configured as input (not checked), then a check in the Value field for the same pin indicates that the external value being applied to the pin is high. Otherwise, the external value is low.		
Analog Input and Output	Set the analog output $(0 - 5 \text{ volts})$ in the Analog Out Value field. This field is invalid for QE65000 Spectrometers.		



Option	Description	
Screen Update Interval (msec)	Set the frequency of screen updates (in msec). The status of the analog input value and the GPIO are updated if analog input is being read.	
	A 0 indicates that the screen is not updated in real time.	

Remove Spectra Collection

This menu selection deletes the captured high-speed spectra collection data. You can also right-click on the high-speed spectra collection icon in the **Data Views** pane and select **Remove Spectra Collection**. See <u>*High-Speed Acquisition*</u> for more information.

Show Devices

This feature allows you to redisplay the attached devices in the Data Sources pane if the tree has been collapsed.



Chapter 6

Processing Menu Functions

Overview

This section provides information on the various functions available under the Processing menu selection.

Pre-processing

Preprocessing functions currently include just nonunity correction. You can also remove the preprocessing function.

Remove Preprocessor

Select Processing | Pre-processing | Remove Preprocessor to ???

Select **Processing** | **Pre-processing** | **Non-unity correction** for reflection experiments if you need to perform a calculation to compensate for a reflection standard that is not perfectly white. You can also access this function with the $\neq 1$ icon in the Preprocessing toolbar (see <u>Preprocessing</u>).

Processing Mode

Processing mode functions include all of the modes necessary to conduct experiments. These modes are also available from the toolbar above the graph. See *Experiment Tutorials* for more information about how to conduct experiments using the processing modes.

Scope

Select **Processing** | **Processing Mode** | **Scope** to switch the current spectral window into Scope mode. You can also click S in the graph's toolbar.



8: Spectrum Menu Functions

The signal graphed in Scope mode is the raw voltage coming out of the A/D converter. This spectral view mode provides complete control of signal processing functions before taking absorbance, transmission, reflection, and relative irradiance measurements. This mode reflects the intensity of the light source, the reflectivity of the grating and mirrors in the spectrometer, the transmission efficiency of the fibers, the response of the detector, and the spectral characteristics of the sample.

Use Scope mode when configuring your setup, adjusting the integration time, and taking dark and reference scans.

Scope Minus Dark

Select **Processing** | **Processing Mode** | **Scope Minus Dark** to switch the current spectral window into Scope mode and subtract the stored dark spectra from each spectrometer channel before SpectraSuite displays it. You can also click in the graph's toolbar.

See the description of Scope mode (Scope) for more information.

Absorbance

Select **Processing | Processing Mode | Absorbance** to switch the current window into Absorbance mode.

You can also click A in the graph's toolbar. You must first store a dark and reference spectra in Scope mode before you can access Absorbance mode.

SpectraSuite uses an equation to determine the concentration of a species in solution (illustrated below). The software uses this equation to evaluate each pixel on the detector and produce the absorbance spectrum:

$$A_{\lambda} = -\log_{10} \left(\frac{S_{\lambda} - D_{\lambda}}{R_{\lambda} - D_{\lambda}} \right)$$

Where:

S = Sample intensity at wavelength λ

D = Dark intensity at wavelength λ

R = Reference intensity at wavelength λ

The concentration of a species in a solution directly affects the absorbance of the solution. This relationship, known as Beer's Law, is expressed as:

$$A_{\lambda} = \varepsilon_{\lambda} c \ell$$

Where:

A = Absorbance at wavelength λ ,

 ε_{λ} = Extinction coefficient of the absorbing species at wavelength λ

c = Concentration of the absorbing species and l is the optical path length of the absorption.

See *Experiment Tutorials* for information on conducting an experiment in Absorbance mode.





Transmission

Select Processing | Processing Mode | Transmission to switch the current window into Transmission

mode. You can also click in the graph's toolbar. This is also the spectral processing mode used for reflection spectroscopy, as the math necessary to compute reflection is identical to that required for transmission. You must first store a dark and reference spectra in Scope mode before you can access Transmission mode.

SpectraSuite calculates the transmission of a solution using the following equation:

$$\%T_{\lambda} = \frac{S_{\lambda} - D_{\lambda}}{R_{\lambda} - D_{\lambda}} \times 100\%$$

Where:

 S_{λ} = Sample intensity at wavelength λ

 D_{λ} = Dark intensity at wavelength λ

 R_{λ} = Reference intensity at wavelength λ

See *Experiment Tutorials* for information on conducting an experiment in Transmission mode.

Reflection

Select **Processing | Processing Mode | Reflection** to switch the current window into Reflection mode.

You can also click \mathbb{R} in the graph's toolbar. Reflection mode is also the spectral processing mode used for transmission spectroscopy, as the math necessary to compute transmission is identical to that required for reflection.

You must take a dark and reference spectra in Scope mode before you can access Reflection mode.

SpectraSuite calculates the reflection of a solution using the following equation:

$$\%T_{\lambda} = \frac{S_{\lambda} - D_{\lambda}}{R_{\lambda} - D_{\lambda}} \times 100\%$$

Where:

 S_{λ} = Sample intensity at wavelength λ

 $D_{\lambda} = \text{Dark}$ intensity at wavelength λ

 R_{λ} = Reference intensity at wavelength λ

See *Experiment Tutorials* for information on conducting an experiment in Reflection mode.

Relative Irradiance

Select **Processing | Processing Mode | Relative Irradiance** to switch the current window into Relative Irradiance mode. You can also click in the graph's toolbar.



8: Spectrum Menu Functions

Before you can access Relative Irradiance mode, you must take a reference spectrum in Scope mode of a blackbody of known color temperature. Additionally, you must obtain a dark spectrum by removing the fiber from the reference lamp and preventing light from entering it.

Relative irradiance spectra are a measure of the intensity of a light source relative to a reference emission source. SpectraSuite calculates relative irradiance using the following equation:

$$I_{\lambda} = B_{\lambda} \left(\frac{S_{\lambda} - D_{\lambda}}{R_{\lambda} - D_{\lambda}} \right)$$

Where:

 B_{λ} = Relative energy of the reference calculated from the color temperature

 S_{λ} = Sample intensity at wavelength λ

 D_{λ} = Dark intensity at wavelength λ

 R_{λ} = Reference intensity at wavelength λ

See *Experiment Tutorials* for information on conducting an experiment in Relative Irradiance mode.

Absolute Irradiance

Scope

Absolute Irradiance

Select **Processing** | **Absolute Irradiance** | **Absolute Irradiance** to create a new graph of spectral data Absolute Irradiance mode. You can also click **I** in the Data Display toolbar.

You do not need to store reference spectrum first since absolute irradiance is not relative to another measurement. However, you must store a dark spectrum and have a calibration file before you can measure absolute irradiance. A Wavelength Calibration Data Sheet containing the calibration file comes with your Ocean Optics spectrometer. Or, you can do your own calibration if desired.

See *Experiment Tutorials* for information on conducting an experiment in Absolute Irradiance mode.

Collection Area

Calibration

Combine Calibration Files



Chapter 9

Strip Charts

Overview

SpectraSuite allows you to use Strip Charts to track processes, perform kinetic analyses, and monitor spectral events all as a function of time. The Strip Chart shows you how the value that you selected appears over time. This value can be any of the following:

- One pixel (a single wavelength)
- The average of a range of pixels
- An integral calculated by one of three methods representing the area beneath the graph's trend line

You can acquire this data using any processing mode (tranmission, absorbance, etc.).

Creating Strip Charts

► Procedure

- 1. Place SpectraSuite in Scope mode by clicking the Scope (S) icon in the Experiment mode toolbar or selecting **Processing | Processing Mode | Scope** from the menu.
- 2. Store a reference spectra and dark spectra.
- 3. Choose the measurement mode (absorbance, transmission, etc.).
- 4. Click the Strip Chart button (\square)

OR

Select File | New | Strip Chart from the menu. The Chart Trend Settings dialog box opens.



9: Strip Charts

🌠 Chart Trend Settings	X
Spectrometers Spectrum Type HR2B1231 Processed	Wavelength Selection One wavelength: 188.80 nm Average from 188.80 nm to 188.80 nm
Display Options Trend Line Color: Select Color Scale Trend Value: Multiply by 1 then add 0 Minimum delay between updates: 0 \$ ms	○ Integrate over 188.80 to 188.80 m Method: Rectangular Accept Cancel

5. Enter the following data to create your Strip Chart:

Field	Selections	
Wavelength Selection	Select the type of data for which you want to view the time trend. Select from the following:	
	 One wavelength – For one pixel (used in fluorescence, for example). Select he wavelength. 	
	 Average from – For a range of pixels. Select the starting and ending wavelengths. 	
	 Integrate over – For the area underneath the trend line. Select the starting and ending wavelengths. Then, select the method for calculating the continuous area underneath the graph curve: Rectangular, Simpson's, or Trapezoid. 	
Display Options	Select a color for the graph's trend line.	
Scale Trend Value	If you would like to adjust your data, enter the appropriate information in this field.	
Minimum delay between updates	Set the amount of time (in milliseconds) between data points on the graph.	
	NOTE: The longer the time between updates, the less computer memory that is used.	

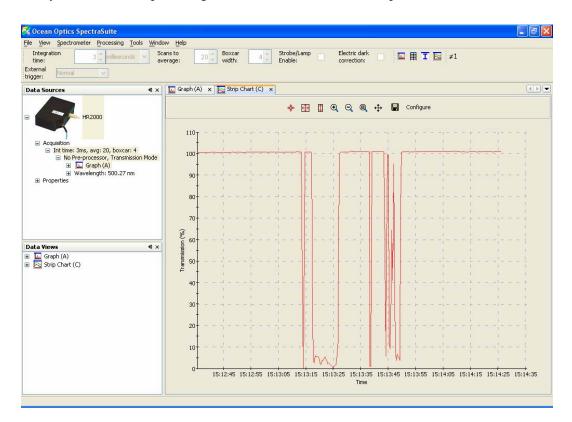
6. Click Accept . The Strip Chart Options dialog box appears. Strip Chart Options lists all of the Strip Charts that you have created, along with the options that you have set for each one. Use this dialog box to add, edit, remove, or clear trends.



Note

Highlight the desired trend to make the **Clear**, **Remove Trend**, and **Edit Trend** buttons active.

Source	Wavelength (nm	n) Mode	Offset	Color
HR2B1231 500.27		Value	0	



Appendix A

Experiment Tutorials

Overview

The following sections contain information on conducting sample experiments using a USB2000 Spectrometer and SpectraSuite.

For information on experiments with Ocean Optics spectrometers other than the USB2000, consult the operating instructions for your particular spectrometer model.

Preparing for Experiments

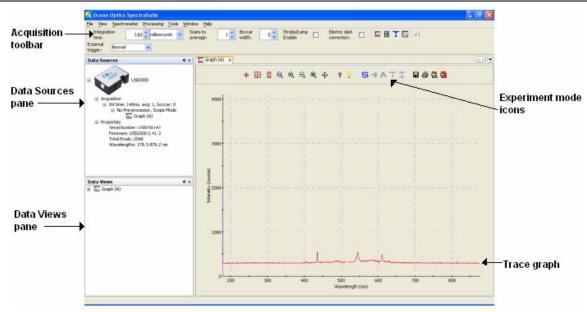
Verify that you have correctly installed the spectrometer, installed SpectraSuite, and configured the light source and other sampling optics.

If you followed the previous steps and started SpectraSuite, the spectrometer is already acquiring data in Scope mode. Even with no light in the spectrometer, SpectraSuite should display a dynamic trace in the bottom of the graph window. If you allow light into the spectrometer, the graph trace should rise with increasing light intensity. This indicates that you correctly installed the software and hardware.

Note the spectrometer(s) that you have installed are listed in the Data Sources pane. Information for each spectrometer listed in this pane consists of the following:

- Acquisition parameters that you set via the (integration time, scans-to-average, boxcar smoothing),
- Whether dark and/or reference spectra have been stored, the graph (A, B, C, etc.) associated with this spectrometer that appears in the right pane (important if you have installed more than one spectrometer),
- Spectrometer's properties (serial number, firmware level, number of pixels, and wavelengths).





Once you install the hardware and software, and establish your sampling system, you are ready to take measurements.

This section details the following types of experiments:

- <u>Absorbance Experiments</u>
- <u>Transmission Experiments</u>
- Irradiance Experiments

The type of measurement you will take determines the configuration of the sampling optics for your system. Furthermore, your choice of reference and data analysis determines how SpectraSuite presents the results.

Note

For each measurement, you must first take a reference and dark spectrum before the experiment mode icon (A, T, I) on the toolbar becomes active. After you take a reference and a dark spectrum, you can take as many measurement scans as needed. However, if you change any sampling variable (integration time, averaging, smoothing, fiber size, etc.), you must store a new dark and reference spectrum.

Application Tips

If the signal you collect is saturating the spectrometer (intensity greater than 4000 counts), you can decrease the light level on scale in scope mode by:

• Decreasing the integration time



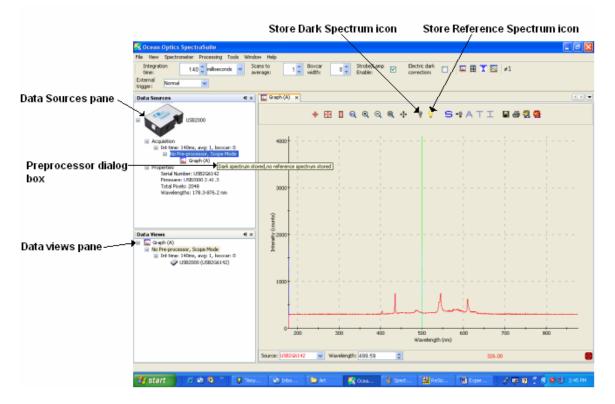
- Attenuating the light going into the spectrometer
- Using a smaller diameter fiber
- Using a neutral density filter with the correct optical density

If the signal you collect has too little light, you can increase the light level on scale in scope mode by:

- Increasing the integration time
- Using a larger diameter fiber
- Removing any optical filters

Taking Reference and Dark Spectra

Reference and dark spectra must be stored before collecting experiment data. If you pass the cursor over the **No-preprocessor** line in either the Data Sources or Data Views pane, a dialog box informs you whether reference and/or dark spectra have been stored.



A reference spectrum is taken when the light source is on and the there is no sample present. Click the Store Reference Spectrum icon $(\ref{eq:spectrum})$ to take a reference spectrum.



A dark spectrum is taken in the absence of light. Block the light path, uncheck the **Strobe/Lamp Enable** option on SpectraSuite's Acquisition toolbar, or turn off the lamp, and then click the Store Dark Spectrum icon () to take a dark spectrum.

Absorbance Experiments

Absorbance spectra are a measure of how much light a sample absorbs. For most samples, absorbance relates linearly to the concentration of the substance. SpectraSuite calculates absorbance (A_{λ}) using the following equation.

$$A_{\lambda} = -\log_{10} \left(\frac{S_{\lambda} - D_{\lambda}}{R_{\lambda} - D_{\lambda}} \right)$$

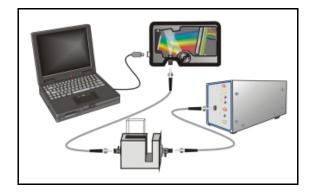
Where:

 S_{λ} = Sample intensity at wavelength λ

 D_{λ} = Dark intensity at wavelength λ

 R_{λ} = Reference intensity at wavelength λ

Typical absorbance setup: The light source (far right) sends light via an input fiber into a cuvette in a cuvette holder (bottom center). The light interacts with the sample. The output fiber carries light from the sample to the spectrometer (top center) connected to the PC (far left).



Absorbance is also proportional to the concentration of the substance interacting with the light (this is known as Beer's Law). Common absorption applications include the quantification of chemical concentrations in aqueous or gaseous samples.



► Procedure

Follow the steps below to take an absorbance measurement using SpectraSuite:

- 1. Place SpectraSuite in Scope mode by clicking the Scope () icon in the Experiment mode toolbar or selecting **Processing | Processing Mode | Scope** from the menu.
- 2. Ensure that the entire signal is on scale. The intensity of the reference signal should peak at about 3500 counts. If necessary, adjust the integration time until the intensity is approximately 3500 counts.
- 3. If you have not already done so, place a sample of the solvent into a cuvette and take a reference spectrum. You must take a reference spectrum before measuring absorbance.

Note

Do not put the sample itself in the path when taking a reference spectrum, only the solvent.

Click the Store Reference Spectrum (¹) icon on the toolbar or by select **File** | **Store** | **Store Reference Spectrum** from the menu bar to store the reference. This command merely stores a

reference spectrum in memory. You must click the Save Spectra () icon on the toolbar or select **File** | **Save** | **Save Spectra Collection** from the menu bar to permanently save the reference spectrum to disk.

4. If you have not already done so, store a dark spectrum reading. To do this, either block the light path to the spectrometer, uncheck the **Strobe/Lamp Enable** box in the Acquisition toolbar, or

turn the light source off. Then, take a dark spectrum by clicking the Store Dark Spectrum (**I**) icon on the toolbar or by selecting **File** | **Store** | **Store Dark Spectrum** from the menu bar. This

command merely stores a dark spectrum in memory. You must click the Save Spectra () icon on the toolbar or select **File** | **Save** | **Save Spectra Collection** from the menu bar to permanently save the spectrum to disk.

Note

If possible, do not turn off the light source when taking a dark spectrum. If you must turn off your light source to store a dark spectrum, allow enough time for the lamp to warm up again before continuing your experiment. After the lamp warms up again, store a new reference (Step 3).

You must take a dark spectrum before measuring absorbance.



5. Put the sample in place and ensure that the light path is clear. Then, take an absorbance

measurement by clicking on the Absorbance (A) icon on the toolbar or selecting **Processing** | **Processing Mode** | **Absorbance** from the menu. Note the following changes on the screen:

- The experiment mode listed in the Data Sources and Data Views panes changes to **Absorbance Mode**.
- The units listed on the Graph pane changes to Absorbance (OD).
- 6. To permanently save the spectrum to disk, click the Save Spectra (🕒) icon on the toolbar or select **File** | **Save** | **Save Spectra Collection** from the menu bar.

Note

If you change any sampling variable (integration time, averaging, smoothing, fiber size, etc.), you must store a new dark and reference spectrum.

Transmission Experiments

Transmission is the percentage of energy passing through a sample relative to the amount that passes through the reference. Transmission mode can also display the portion of light *reflected* from a sample, since transmission and reflection measurements use the same mathematical calculations. We express transmission as a percentage ((T_{λ})) relative to a standard substance (such as air). SpectraSuite calculates (T_{λ}) (or (R_{λ})) with the following equation.

$$\% T_{\lambda} = \frac{S_{\lambda} - D_{\lambda}}{R_{\lambda} - D_{\lambda}} \times 100\%$$

Where:

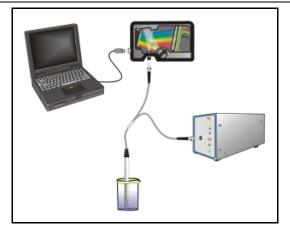
 S_{λ} = Sample intensity at wavelength λ

 $D_{\lambda} = \text{Dark}$ intensity at wavelength λ

 R_{λ} = Reference intensity at wavelength λ

Typical transmission setup: The light source (far right) sends light via the input leg of a transmission probe into a container (bottom center). The light interacts with the sample. The output leg of the transmission probe carries the information to the spectrometer (top center), which transmits the information to the PC (far left).





Common transmission applications include measuring light through solutions, optical filters, optical coatings, and other optical elements (such as lenses and fibers).

► Procedure

Perform the following steps to take a transmission measurement using SpectraSuite:

- 1. Place SpectraSuite in Scope mode by clicking the Scope (S) icon in the Experiment mode toolbar or selecting **Processing | Processing Mode | Scope** from the menu.
- 2. Ensure that the entire signal is on scale. The intensity of the reference signal should peak at about 3500 counts. If necessary, adjust the integration time until the intensity is approximately 3500 counts.
- 3. If you have not already done so, place a sample of the solvent into a cuvette and take a reference spectrum. You must take a reference spectrum before measuring transmission.



Note

Do not put the sample itself in the path when taking a reference spectrum, only the solvent.

Click the Store Reference Spectrum (¹¹) icon on the toolbar or by select **File** | **Store** | **Store Reference Spectrum** from the menu bar to store the reference. This command merely stores a

reference spectrum in memory. You must click the Save Spectra () icon on the toolbar or select **File** | **Save** | **Save Spectra Collection** from the menu bar to permanently save the reference spectrum to disk.

4. If you have not already done so, store a dark spectrum reading. To do this, either block the light path to the spectrometer, uncheck the **Strobe/Lamp Enable** box in the Acquisition toolbar, or

turn the light source off. Then, take a dark spectrum by clicking the Store Dark Spectrum (\mathbb{I}) icon on the toolbar or by selecting **File** | **Store** | **Store Dark Spectrum** from the menu bar. This

command merely stores a dark spectrum in memory. You must click the Save Spectra (\square) icon on the toolbar or select **File** | **Save** | **Save Spectra Collection** from the menu bar to permanently save the spectrum to disk.

Note

If possible, do not turn off the light source when taking a dark spectrum. If you must turn off your light source to store a dark spectrum, allow enough time for the lamp to warm up again before continuing your experiment.

You must take a dark spectrum before measuring transmission.

- 6. Put the sample in place and ensure that the light path is clear. Then, take a transmission measurement by clicking on the Transmission (T) icon on the toolbar or selecting Processing | Processing Mode | Transmission from the menu. Note the following changes on the screen:
- The experiment mode listed in the Data Sources and Data Views panes changes to **Transmission Mode**.
- The units listed on the Graph pane changes to **Transmission** (%).
- 7. To permanently save the spectrum to disk, click the Save Spectra () icon on the toolbar or select **File** | **Save** | **Save Spectra Collection** from the menu bar.





Note

If you change any sampling variable (integration time, averaging, smoothing, fiber size, etc.), you must store a new dark and reference spectrum.

Irradiance Experiments

Irradiance is the amount of energy at each wavelength emitted from a radiant sample. Absolute irradiance is the measure of light in absolute terms. Relative irradiance is a comparison of the fraction of energy the sample emits and the energy the sampling system collects from a lamp with a blackbody energy distribution (normalized to 1 at the energy maximum). SpectraSuite calculates relative irradiance with the following equation:

$$\mathbf{I}_{\lambda} = \mathbf{B}_{\lambda} \left(\frac{\mathbf{S}_{\lambda} - \mathbf{D}_{\lambda}}{\mathbf{R}_{\lambda} - \mathbf{D}_{\lambda}} \right)$$

Where:

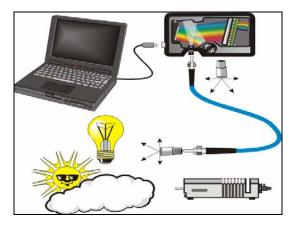
 B_{λ} = Relative energy of the reference (calculated from the color temperature) at wavelength λ

 S_{λ} = Sample intensity at wavelength λ

 $D_{\lambda} = \text{Dark}$ intensity at wavelength λ

 R_{λ} = Reference intensity at wavelength λ

Typical relative irradiance setup: Use a light source with a known color temperature (such as the LS-1 or LS-1-LL (lower right) to take a reference spectrum. The light to measure (lower left) accumulates through a CC-3 Cosine Corrector (or FOIS integrating sphere) into an input fiber, which carries the light information to the spectrometer. The spectrometer then transmits the information to the PC, which compares the measured spectra against the reference spectrum, thus removing wavelength-dependent instrument response from the measurement.





Common applications include characterizing the light output of LEDs, incandescent lamps, and other radiant energy sources such as sunlight. Relative irradiance measurements also include fluorescence measurements, which measure the energy given off by materials excited by light at shorter wavelengths.

Absolute Irradiance

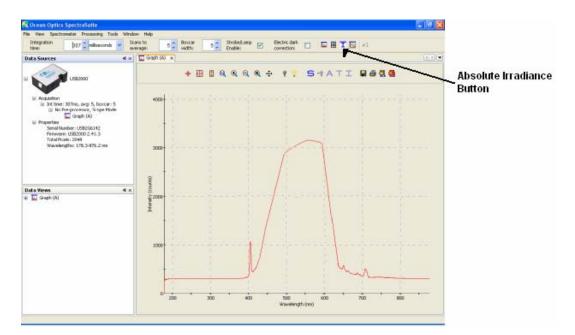
You do not need to store reference spectrum first since absolute irradiance is not relative to another measurement. However, you must store a dark spectrum and have a calibration file before you can measure absolute irradiance. A Wavelength Calibration Data Sheet containing the calibration file comes with your Ocean Optics spectrometer. Or, you can do your own calibration if desired.

Loading an Existing Calibration File

► Procedure

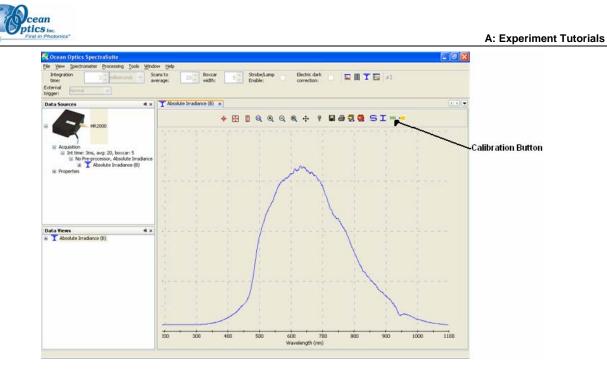
1. From the main graph, click **on the Irradiance toolbar**

OR



Select File | New | Absolute Irradiance Graph.

The Absolute Irradiance graph in Scope mode appears.



2. Click ¹⁰¹⁰. The **Absolute Irradiance Calibration** dialog box appears.

🔀 Absolute Irradiance Calibration	×
● Get Irradiance Calibration from File File Name: Browse File Type: OOI ASCII Format Collection Area: cm^2 Use Integrating Sphere:	Calibration Information
Get Irradiance Calibration from Spectrometer	
New calibration Calibrate	
	Apply Cancel

- 3. Select Get Irradiance Calibration from File.
- 4. Enter the calibration file name or browse to the desired .cal file.



🌠 Absolute Irradiance Calibration	X
 Get Irradiance Calibration from File File Name: aronCalibration.cal Browse File Type: OOI ASCII Format Collection Area: 7.854E-5 cm^2 Use Integrating Sphere: Get Irradiance Calibration from Spectrometer New calibration 	Calibration Information)ate Fri Nov 11 14:57:57 EST 2008 Spectrometer HR2B1231 .amp SN Sample nt. Time (usec) 3072 Average 20 Boxcar 5 Tiber (micron) 100 Pixels: 2048 Apply Cancel

- 5. If you are using an integrating sphere, you can check the **Use Integrating Sphere** option and change the collection area, if desired.
- 6. Click Apply

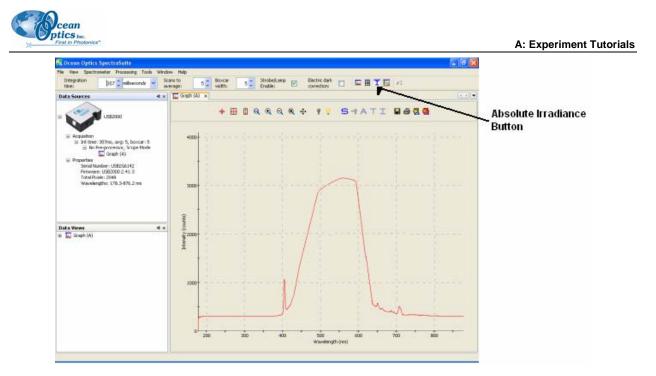
Creating a New Calibration File

► Procedure

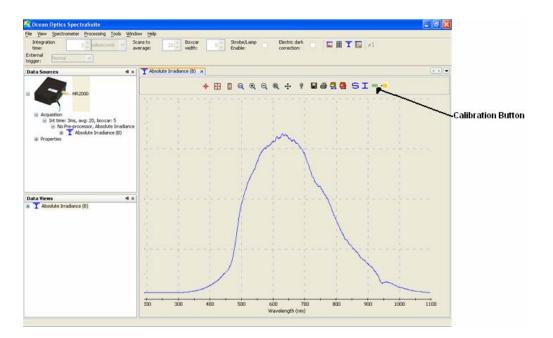
From the main graph, click on the Irradiance toolbar

OR

Select File | New | Absolute Irradiance Graph.



The Absolute Irradiance graph in Scope mode appears.



7. Click ¹⁰¹⁰. The **Absolute Irradiance Calibration** dialog box appears.



% /	Absolut	e Irradiance Cali	bration			
Fil Fi	 Set Irradiance Calibration from File File Name: Browse File Type: OOI ASCII Format Collection Area: cm^2 Use Integrating Sphere: □ 			Calibrat	ion Information	
\subset	Get Irradiance Calibration from Spectrometer					
C) New ca	libration	Calibrate			
					Apply Cance	

- 8. Select New Calibration.
- 9. Click Calibrate... The Spectrometer Irradiance Calibration wizard appears.

Spectrometer Irradiance Calibration				
Steps 1. Calibration Spectrum 2. Dark Spectrum 3. Lamp File 4. Final Calibration	wizard (1 of 4)Store CalibrationCalibration Spectrum Preview			
	< <u>Back</u> Next > Einish Cancel Help			

10. If you have not done so already, you must store a reference spectrum, a dark spectrum, and a lamp file. Follow the wizard's prompts.



If you already have these files, the **Next** button will be active. Click Next > to go on to the next step. When you have stored all of the necessary files, the final screen of the Spectrometer Irradiance Calibration wizard appears (screen 4 of 4).

Spectrometer Irradiance Cal	ibration		X
Steps 1. Calibration Spectrum 2. Dark Spectrum 3. Lamp File 4. Final Calibration	wizard (4 of 4) Wavelength (nm) 615.42 615.86 616.31 616.76	Calibration (uJ/count) 1.762E-7 1.769E-7 1.774E-7 1.782E-7	5.0† 4.5
B	Collection Area Using Integratin Fiber Diameter: Collection Area	micron : cm^2	g 3.5 g 2.5 2.0 1.5 1.5 1.5 0.0 0 400 800 Wavelength (nm) To Spectrometer
		< <u>B</u> ack Next >	<u>Finish</u> Cancel <u>Help</u>

11. Use this screen to specify whether you are using an integrating sphere. If you know the fiber diameter, you can enter it here and allow SpectraSuite to calculate the collection area. Or, if you know the collection area, you can enter that figure directly.

Note in the **Spectrometer Calibration Preview** that the lamp file curve (in yellow) is overlayed on the spectrometer's curve (red). Once you apply this calibration (in Step 7), the curves should be exactly the same.

12. Click Finish. The Absolute Irradiance Calibration dialog box reappears. Click Apply to apply this calibration to the spectrometer.



🄀 Absolute Irradiance Calibration	
○ Set Irradiance Calibration from File File Name: Browse File Type: OOI ASCII Format ✓ ✓ Collection Area: cm^2 Use Integrating Sphere: □ ✓ Get Irradiance Calibration from Spectrometer	Calibration Information New calibration ready. Collection area: 7.854E-5
	Apply Cancel

Measuring Absolute Irradiance

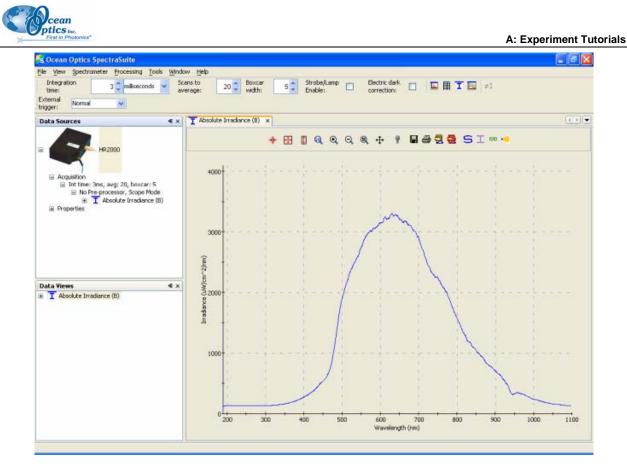
Once you have stored a dark reference and a calibration file, you are ready to measure absolute irradiance.

From the main graph, click on the Irradiance toolbar

OR

Select File | New | Absolute Irradiance Graph.

The Absolute Irradiance Graph appears.



Relative Irradiance

Relative irradiance is a measurement of light relative to the known color temperature of a blackbody light source. Common applications include characterizing the light output of LEDs, incandescent lamps, and other radiant energy sources such as sunlight. Relative irradiance measurements also include fluorescence measurements, which measure the energy given off by materials excited by light at shorter wavelengths. You must have stored dark and reference spectra before measuring relative irradiance.

Procedure

- 1. Place SpectraSuite in Scope mode by clicking the Scope () icon in the Experiment mode toolbar or selecting **Processing** | **Processing Mode** | **Scope** from the menu.
- 2. Ensure that the entire signal is on scale. The intensity of the reference signal should peak at about 3500 counts. If necessary, adjust the integration time until the intensity is approximately 3500 counts.
- 3. Take a reference spectrum using a light source with a black body of a known color temperature, such as the LS-1.
- 4. Click the Store Reference Spectrum (¹) icon on the toolbar or by select **File** | **Store** | **Store Reference Spectrum** from the menu bar to store the reference. This command merely stores a



reference spectrum in memory. You must click the Save Spectra () icon on the toolbar or select **File** | **Save** | **Save Spectra Collection** from the menu bar to permanently save the reference spectrum to disk.

5. If you have not already done so, store a dark spectrum reading. To do this, either block the light path to the spectrometer, uncheck the **Strobe/Lamp Enable** box in the Acquisition toolbar, or

turn the light source off. Then, take a dark spectrum by clicking the Store Dark Spectrum (**V**) icon on the toolbar or by selecting **File** | **Store** | **Store Dark Spectrum** from the menu bar. This

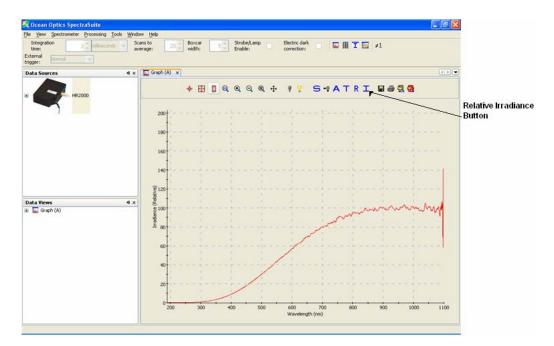
command merely stores a dark spectrum in memory. You must click the Save Spectra () icon on the toolbar or select **File** | **Save** | **Save Spectra Collection** from the menu bar to permanently save the spectrum to disk.

Note

If possible, do not turn off the light source when taking a dark spectrum. If you must turn off your light source to store a dark spectrum, allow enough time for the lamp to warm up again before continuing your experiment.

You must take a dark spectrum before measuring relative irradiance.

6. Position the fiber at the light source you want to measure. Then, click **I**. The Relative Irradiance graph appears.





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