Wavelength Calibration

How to Perform Wavelength Calibrations on a Modular Spectrometer

Quick Start Guide

This document provides information which allows you to perform your own wavelength calibration on your spectrometer. Each spectrometer is calibrated before it leaves Ocean Insight, however the wavelength for all spectrometers will drift slightly as a function of time and environmental conditions. Additionally, Ocean Insight recommends that a calibration be performed after changing the slit on a spectrometer as it will optimize the performance of the device.

An EEPROM flash memory chip in each spectrometer contains wavelength calibration coefficients, linearity coefficients, and a serial number unique to each spectrometer. The spectroscopy application reads these values directly from the spectrometer, enabling the ability to "hot-swap" spectrometers between computers without entering the spectrometer coefficients manually on each computer.

OceanView may be used to write calibration coefficients to the spectrometer.

Wavelength Calibration

You are going to be solving the following equation, which shows that the relationship between pixel number and wavelength is a third-order polynomial:

$$\lambda_p = I + C_1\,p + C_2\,p^2 + C_3\,p^3$$

Where:

λ = the wavelength of pixel p

I = the wavelength of pixel 0

C_1 = the first coefficient (nm/pixel)

C_2 = the second coefficient (nm/pixel^2)

C_3 = the third coefficient (nm/pixel^3)

You will be calculating the value for I and the three Cs.

Preparing for Calibration

To recalibrate the wavelength of your spectrometer, you need the following components:

- A light source capable of producing spectral lines

<div style="background-color:#434a7c; color:white; padding:10px;">

NOTE

Ocean Insight's AR-2 Argon lamp or HG-2 Mercury-Argon lamps are ideal for recalibration.

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- An Ocean Insight modular spectrometer

- An optical fiber (for spectrometers without a built-in slit, a 50-μm fiber works best)

- A spreadsheet program (Excel, for example) or a calculator that performs third-order linear regressions

> **NOTE**
>
> If you are using Microsoft Excel, choose **Tools** | **Add-Ins** and check **AnalysisToolPak** and **AnalysisToolPak-VBA**.

Use the Analysis ToolPak to perform complex data analysis

If you need to develop complex statistical or engineering analyses, you can save steps and time by using the Analysis ToolPak. You provide the data and parameters for each analysis, and the tool uses the appropriate statistical or engineering macro functions to calculate and display the results in an output table. Some tools generate charts in addition to output tables.

The data analysis functions can be used on only one worksheet at a time. When you perform data analysis on grouped worksheets, results will appear on the first worksheet and empty formatted tables will appear on the remaining worksheets. To perform data analysis on the remainder of the worksheets, recalculate the analysis tool for each worksheet.

The Analysis ToolPak includes the tools described in the following sections. To access these tools, click **Data Analysis** in the **Analysis** group on the **Data** tab. If the **Data Analysis** command is not available, you need to load the Analysis ToolPak add-in program.

Load and activate the Analysis ToolPak

1. Click the **File** tab, click **Options**, and then click the **Add-Ins** category.

    a. If you're using Excel 2007, click the **Microsoft Office Button**, and then click **Excel Options**

2. In the **Manage** box, select **Excel Add-ins** and then click **Go**.

    a. If you're using Excel for Mac, in the file menu go to **Tools** > **Excel Add-ins.**

3. In the **Add-Ins** box, check the **Analysis ToolPak** check box, and then click **OK**.

    a. If **Analysis ToolPak** is not listed in the **Add-Ins available** box, click **Browse** to locate it.

    b. If you are prompted that the Analysis ToolPak is not currently installed on your computer, click **Yes** to install it.

> **NOTE**
>
> To include Visual Basic for Application (VBA) functions for the Analysis ToolPak, you can load the Analysis ToolPak - VBA Add-in the same way that you load the Analysis ToolPak. In the **Add-ins available** box, select the **Analysis ToolPak - VBA** check box.

Calibrating the Wavelength of the Spectrometer

1. Place the spectrometer operating software into Quick View (Scope) mode and take a spectrum of your light source. Adjust the integration time until there are several peaks on the screen that are not off-scale.

2. Move the cursor to one of the peaks and position the cursor so that it is at the point of maximum intensity.

3. Record the pixel number that is displayed in the status bar or legend (located beneath the graph). Repeat this step for all the peaks in your spectrum.

4. Use the spreadsheet program or calculator to create a table like the one shown in the following figure. In the first column, place the exact or true wavelength of the spectral lines that you used.

   In the second column of this worksheet, place the observed pixel number. In the third column, calculate the pixel number squared, and in the fourth column, calculate the pixel number cubed.

Independent Variable		Dependent Variables		Values Computed from the Regression Output	
True Wavelength (nm)	**Pixel #**	**Pixel # ^{2}**	**Pixel # ^{3}**	**Predicted Wavelength**	**Difference**
253.65	175	30625	5359375	253.56	0.09
296.73	296	87616	25934336	296.72	0.01
302.15	312	97344	30371328	302.40	-0.25
313.16	342	116964	40001688	313.02	0.13
334.15	402	161604	64964808	334.19	-0.05
365.02	490	240100	117649000	365.05	-0.04
404.66	604	364816	220348864	404.67	-0.01
407.78	613	375769	230346397	407.78	0.00
435.84	694	481636	334255384	435.65	0.19
546.07	1022	1044484	1067462648	546.13	-0.06
576.96	1116	1245456	1389928896	577.05	-0.09
579.07	1122	1258884	1412467848	579.01	0.06
696.54	1491	2223081	3314613771	696.70	-0.15
706.72	1523	2319529	3532642667	706.62	0.10
727.29	1590	2528100	4019679000	727.24	0.06
738.40	1627	2647129	4306878883	738.53	-0.13
751.47	1669	2785561	4649101309	751.27	0.19

5. Use the spreadsheet or calculator to calculate the wavelength calibration coefficients. In the spreadsheet program, find the functions to perform linear regressions.

   - If using Excel, look under **AnalysisToolPak**

6. Select the true wavelength as the dependent variable (Y). Select the pixel number, pixel number squared, and the pixel number cubed as the independent variables (X). After executing the regression, you will obtain an output similar to the one shown below. Numbers of importance are noted.

Regression Statistics

Multiple R	0.999999831
R Square	0.999999663
Adjusted R Square	0.999999607
Standard Error	0.125540214
Observations	22

R Square 0.999999663 ← R Squared

		Coefficients	Standard Error
Intercept		190.473993	0.369047536
X Variable 1	First coefficient	0.36263983	0.001684745
X Variable 2	Second coefficient	-1.174416E-05	8.35279E-07
X Variable 3	Third coefficient	-2.523787E-09	2.656608E-10

7. Record the Intercept, as well as the First, Second, and Third Coefficients. Additionally, look at the value for R squared. It should be very close to 1. If not, you have most likely assigned one of your wavelengths incorrectly.

    Keep these values at hand.

Saving the New Calibration Coefficients

You can overwrite old calibration coefficients on the spectrometer if you are communicating with the spectrometer via OceanView. Extreme caution must be used when performing this step. Contact Technical Support at Ocean Insight for assistance to obtain the necessary password and steps to perform this function in OceanView.
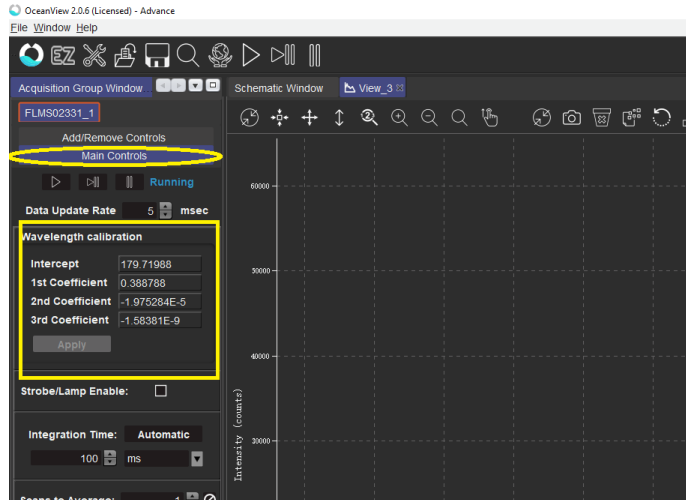
OceanView 2.0

1. Ensure the spectrometer is properly connected.

2. Open OceanView and go to the Acquisition Group Window.

3. Select Add/Remove controls.
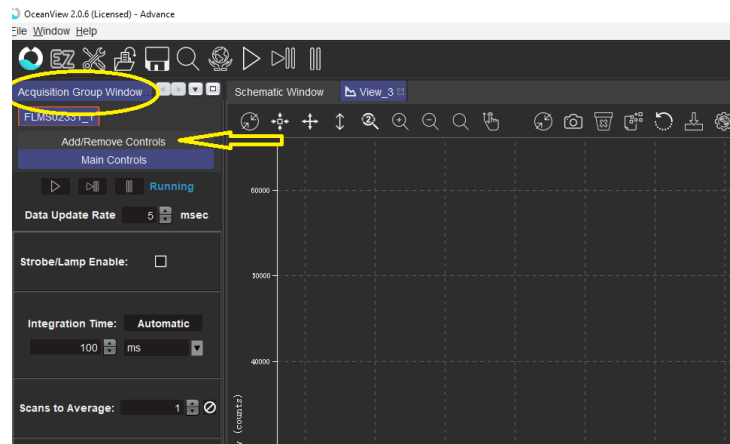
4. Select Wavelength Coefficients and enter the password.

5. Enter the password 'oceanfactory' and click OK.
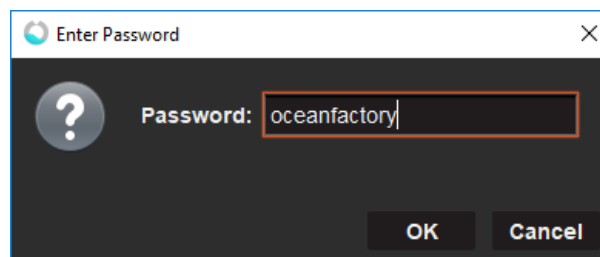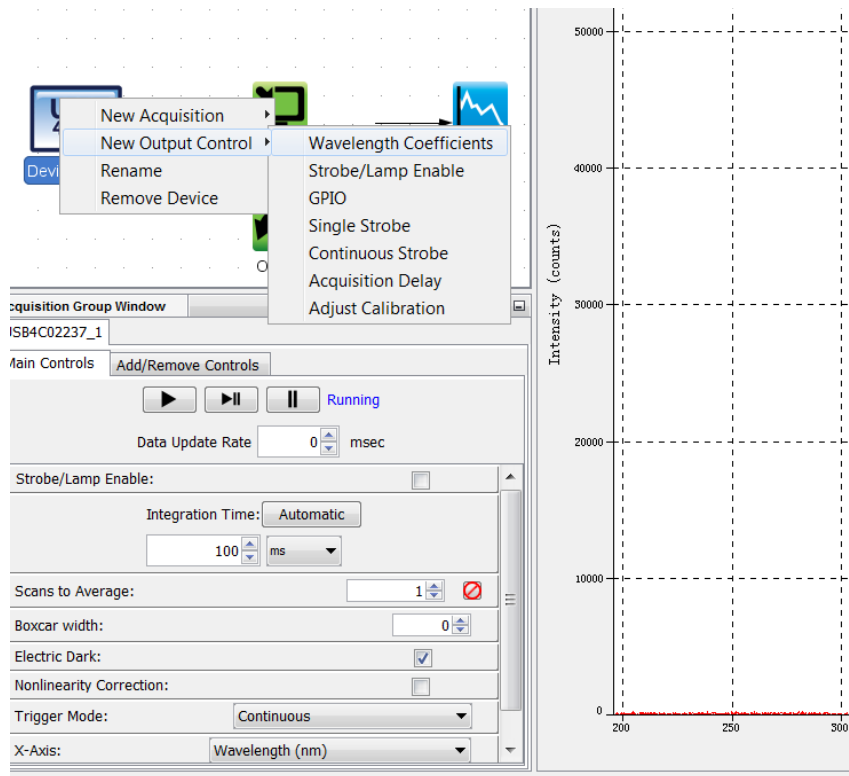

6. Go back to Main Controls and update your spectrometer coefficients.
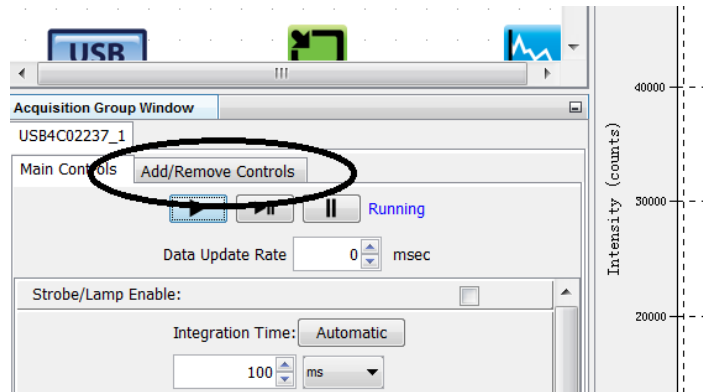
7.   Select Apply and then close OceanView.

8.   Relaunch OceanView and verify your new coefficients have been loaded.
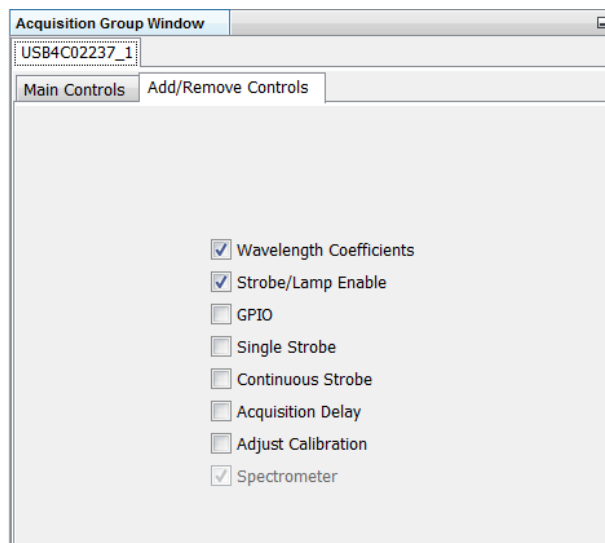
OceanView 1.6.3

1.   Open OceanView 1.6.3 and go to the schematic Window. Right-click on your spectrometer and go to New Output Control, and then select Wavelength Coefficients.
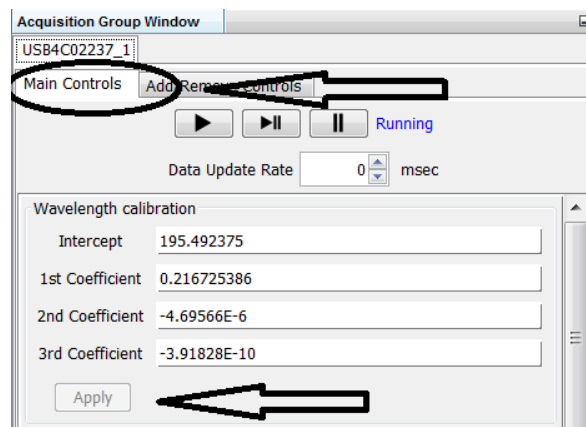

2.   In the Acquisition Group Window, go to Add/Remove controls.

3. On the Add/Remove Controls tab, check Wavelength Coefficients. When prompt for a password enter 'oceanfactory' and click OK.

4. Go to the Main Controls tab, and update your coefficients then select apply.

5. Now close OceanView and reopen it.

Unlock the Unknown

Ocean Insight exists to end guessing. We equip humanity with technology and data to make precisely informed decisions providing transformational clarity for human advancement in health, safety, and the environment.

Questions?
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