

## BC367: Conducting a Kinetics Experiment (Absorbance vs. Time)

Power up and calibrate your spectrometer according to the previous Ocean Optics instructions from BC367 Experiment 2. Note that you will need to re-calibrate with the appropriate reference solution later on in this procedure. Simply warm up and calibrate with dH<sub>2</sub>O the first time (according to the previous directions).

### I. Running a Kinetics Trial

1. Click on the Configure Spectrometer Data Collection icon, , located on the right hand side of the toolbar to open the Configure Spectrometer Data Collection display.

2. Click Abs vs. Time (under the Set Collection Mode). Click on Treat Contiguous Wavelengths as a Single Range. The wavelength of maximum absorbance will be automatically selected. First, if you don't want to use this automatic wavelength, press the Clear button. We want to average over a range of wavelengths for better precision, Figure 1. To average over a range of wavelengths, drag the mouse on the spectrum to select the analysis wavelengths. You should select at least 5 wavelengths on both sides of the maximum. In choosing a wavelength range, you should choose a narrow enough range that the absorbance is relatively constant (i.e. a relatively "flat" region near the maximum). Alternatively you can Scroll the Select Wavelengths list box to a wavelength near your absorbance maximum and then click on about 5 wavelengths on both sides of the maximum. Click OK.

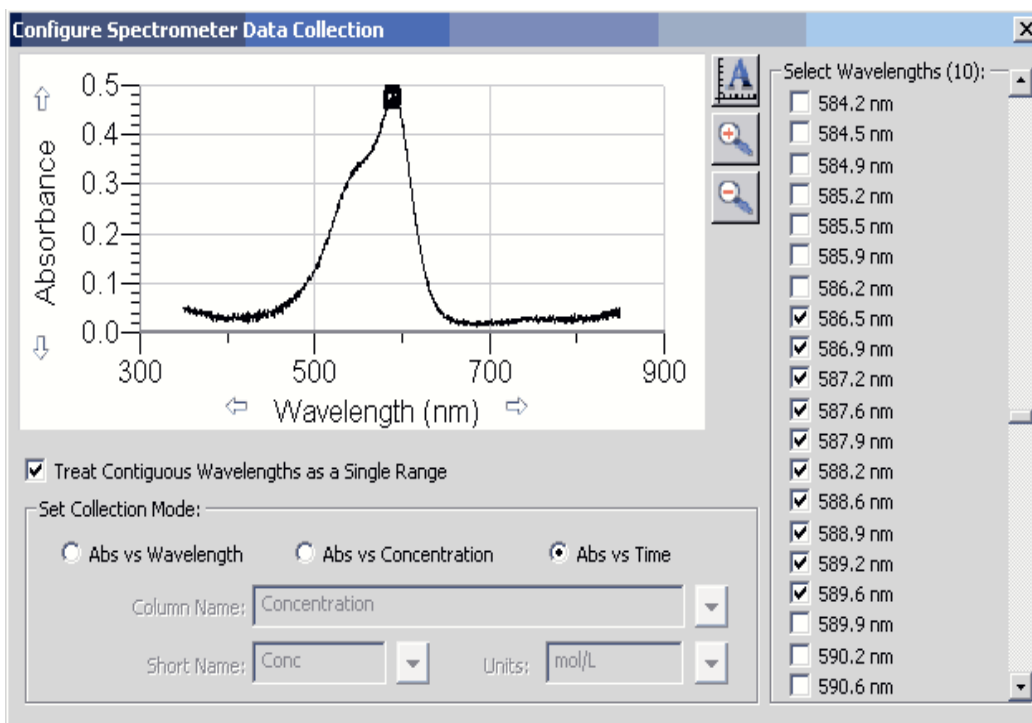



Figure 1. Averaging over several wavelengths to obtain better precision.

3. Click Data Collection . Choose an appropriate Length and Sampling Rate as suggested for your experiment (for example, 1 minute at 30 samples/minute) as in Figure 2. Click Done.

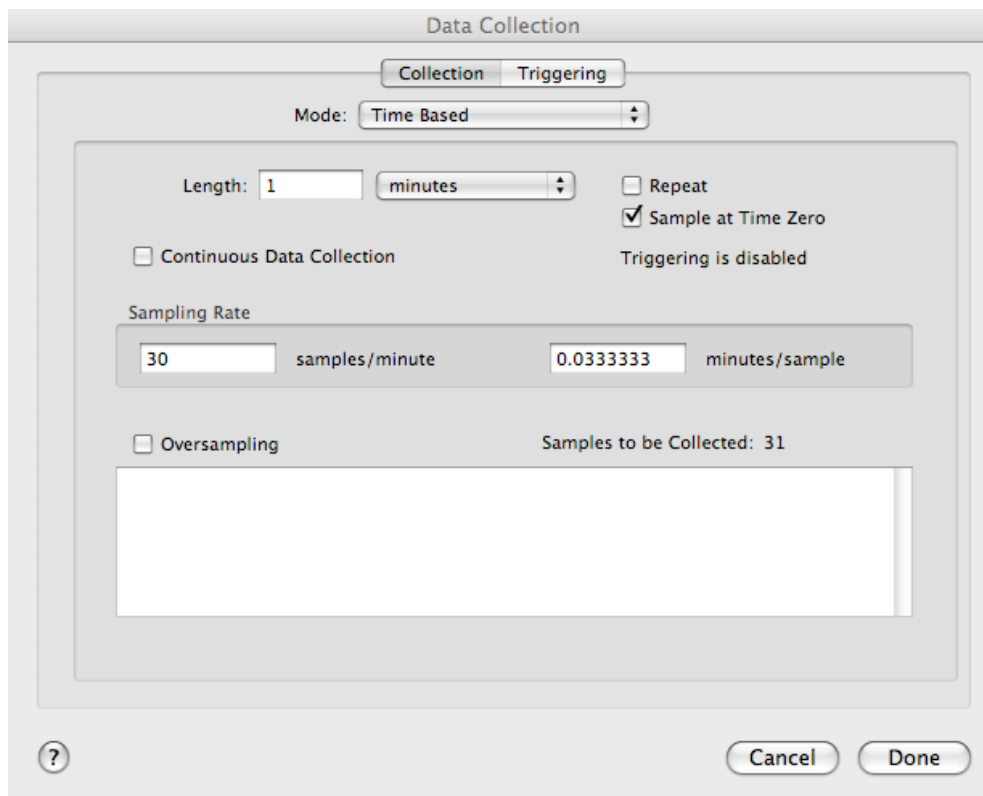





Figure 2. Setting the sampling rate and total experiment length.


4. You must calibrate the spectrophotometer using the correct blanking solution.


5. Add your reagents and a special cuvette stirbar to the cuvette. Adjust the stirrer to allow for quick mixing, but not for splashing. BE SURE THAT THE HEATING ELEMENT OF THE HOTPLATE IS TURNED OFF! Check for bubbles in the cuvette. If present, tap the cuvette gently to remove the bubbles as they may impede the light path.

6. Add the correct volume of your final reagent (sample to be tested) to the cuvette. Immediately click on the  button. You will can autoscale the y-axis by clicking on .

7. When an appropriate time has elapsed (in collection setup), collection of data is automatically stopped.

8. Click on the Linear Fit icon, . This will display the linear equation and the correlation coefficient of the line. The line can be modified by clicking on the black bracket at the end of the line. Sliding the bracket will highlight the desired “new” portion of the line. Go to File – Print graph, set up print options and click on OK to print your kinetics plot. The file can also be saved to the desktop and or copied into Microsoft Word.

9. To run another kinetics trial, close the linear fit window, prepare the next sample, click on the collect button , and choose erase and continue (or store latest run if you prefer to overlap the plots).

10. To exit the kinetics mode and return to taking full spectra, click on the Configure Spectrometer Data Collection icon, . Click Abs vs. Wavelength (under the Set Collection Mode). Click OK.

## II. Finishing up

1. Make sure to rinse your cuvettes three times with reagent grade water. Remember not to use paper towels to clean the glass or plastic surfaces, only use ChemWipes. Don't stick anything sharp into the cuvettes, including a test tube brush.

2. Make sure the area around the spectrometer is clean and dry.

3. Please unplug the spectrometer's transformer. The deuterium lamp in the spectrometer has a limited lifetime and replacements are very expensive.