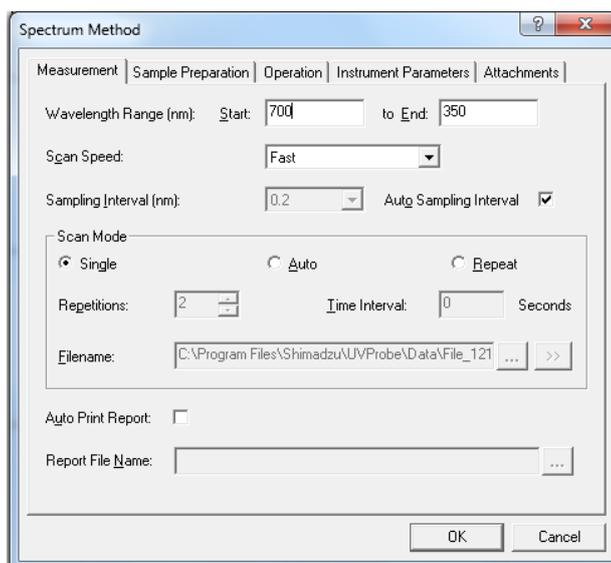


Shimadzu UV-2600 Spectrometer Operation Guide

Recording a scanning spectrum

1. Select the spectrum mode  from the tool bar.
2. Enter the required parameters for your experiment in the method manager  (or **Ctrl+M**).



Set the appropriate wavelength range (this must always run high to low). Leave all other settings unchanged as in the screenshot above. When you have selected your wavelength range, press **OK**.

3. Ensure the 'active' tab is selected:  Active  Overlay  Stacked
4. Insert two cuvettes filled with solvent into the spectrometer (ensure there are no air bubbles, and wipe the optical faces of the cuvettes with a tissue).

The rear holder is the reference compartment, and the front holder is for the sample. (Sometimes a multi-position sample holder will be fitted, in this case, check which sample position is illuminated on the CPS controller and place your cuvette in the appropriate slot. The optical sides of the cuvettes need to be facing the sides.)

5. Select baseline  **Baseline** and ensure the wavelengths are correct (If you wish to change them, return to the method manager ) , then click ok.

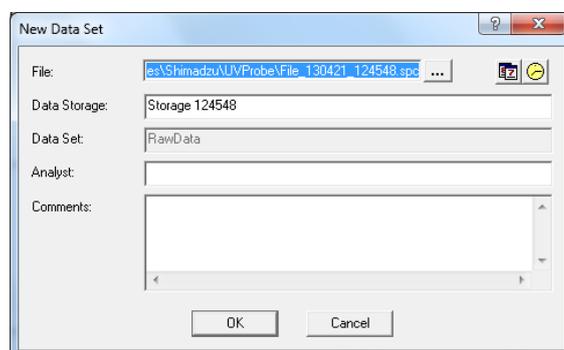
A baseline will be recorded. Whilst the spectrum is running you will see the wavelength values changing and the instrument control buttons will be greyed out:



Once the baseline is complete, the start button will become available.

6. Remove the sample cuvette of solvent and replace with the desired sample.
7. Press **Start**  **Start** and the spectrum will be recorded.

You should see the line being plotted from right to left. When the sample collection is complete a window will appear:



8. Click **OK** to this window, then press **Ctrl+L** to save the data.
9. Choose **Peak Pick** in the operations menu. A table should appear (top left) showing the wavelengths and absorption values. Alternatively, you can use **Point Pick** and drag the marker to the peak you want to select, and then **double click** it for the peak to be added to the table.
10. To print the data, first adjust the scales on the graph. Click on the top value in the y axis and the software will allow you to enter a new upper limit. Choose something appropriate for your data. Do the same for the lower limit (this should usually be close to zero!). To print the spectrum and peak table, right

click on the peak table and press Print. If you just want the spectrum, right click on the spectrum and press Print.

11. If you want to overlay multiple spectra on a single graph:
 - a. Record each of your spectra, as described above, and then select the overlay tab.
 - b. On the right hand side there will be a list of recorded spectra, tick the ones you wish to display.
 - c. When you have selected the desired spectra, adjust the overlay y axis as appropriate again, then right click on the graph and choose print.

It is not possible to print a peak table with the overlays.

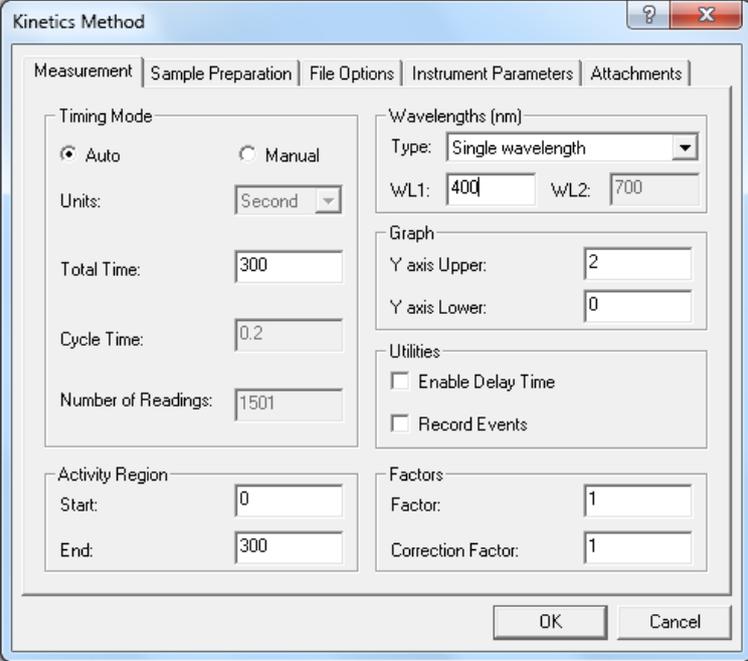
Recording photometric data

1. Select the photometric mode  from the tool bar.
2. Select go to wavelength  and enter the desired wavelength.
3. Press **OK** and the spectrometer will adjust to that wavelength.
4. Insert your background solution in the front cell holder (you do not need a reference solution).
5. Press **Auto zero** .
6. Remove the blank and insert your first sample.
7. The spectrometer will give you a live absorbance readout.

Note down the value in your lab script or lab book.

Recording a kinetics plot

1. Select the kinetics mode  from the tool bar.
2. Select the required parameters for your experiment in the method manager  (or **Ctrl+M**).



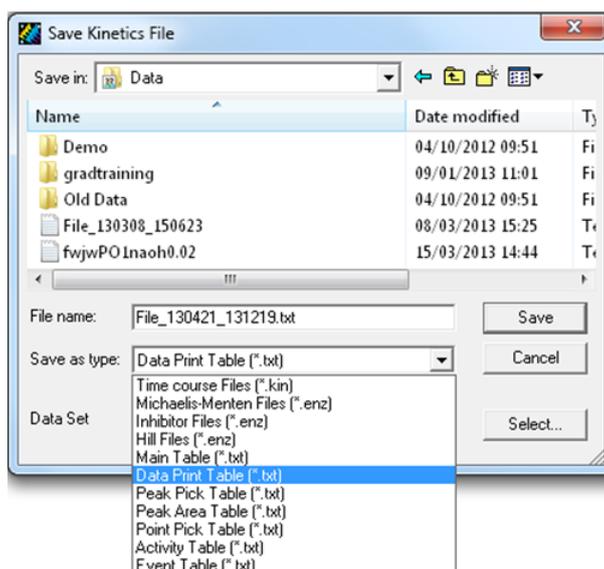
The screenshot shows the 'Kinetics Method' dialog box with the following settings:

- Timing Mode: Auto, Manual
- Units: Second
- Total Time: 300
- Cycle Time: 0.2
- Number of Readings: 1501
- Activity Region: Start: 0, End: 300
- Wavelengths (nm): Type: Single wavelength, WL1: 400, WL2: 700
- Graph: Y axis Upper: 2, Y axis Lower: 0
- Utilities: Enable Delay Time, Record Events
- Factors: Factor: 1, Correction Factor: 1

Ensure single wavelength mode is selected and in the WL1 box, enter your desired wavelength. Adjust the total time and activity region (this is how long the kinetic run will measure for) then click **OK**. You should see the spectrometer displaying your desired wavelength. All other settings should be as above.

3. Insert your blank solution in the front cell holder (if you are measuring the formation of product, this will be your starting solution, if you are measuring the decay of starting material, this will be a solution of your product). You do not require a reference solution.
4. Click **Auto Zero**  and the spectrometer will zero itself.
5. Remove the blank solution and insert your kinetic sample.
6. Click **Start** to begin the run. You should see a graph being plotted. On completion a dialogue box will appear. Click finish, then press **Ctrl+L** to save the data.

- To print the kinetics graph, adjust the y axis to an appropriate scale, by clicking on the top value and replacing it, and similarly with the lower figure (this should usually be set to zero). Then right click on the graph and select Print.
- Saving the data in excel. It is not usually necessary to print the raw point data, but you will often want to save this in a format usable in excel. To do this click **File, Save As**, and then change the file type to **Data Print Table**.



Ensure the data is being saved in C:\Program Files\Shimadzu\UVProbe\Data (check). You should normally accept the default filename, as this is what will be listed on your graph printout. You can access this data by following the instructions for data access on the teaching labs homepage (allow 15 minutes). This enables easy import into excel.

Saving and opening Shimadzu data in excel

- Ensure data is saved as a Data Print Table (.txt)
- Go to [ftp://chemtl.york.ac.uk](http://chemtl.york.ac.uk). Find the name of computer used to record your UV spectrum and click to open the file you want. **Right click** and click **Save As**.
- Open excel and click **Open**.
- Find your (.txt) file and open it.
- Click **Next**.
- Tick the **Comma** box.
- Click **Finish**.