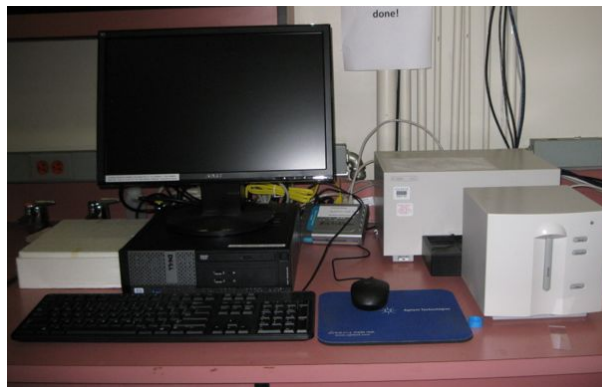
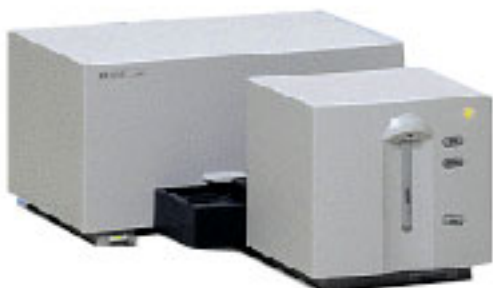


Agilent 8453 UV-vis Spectrometer

Introduction

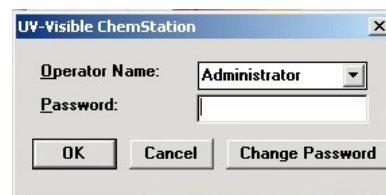
The Agilent 8453 UV-Vis instrument is a simple but powerful diode-array spectrophotometer capable of quickly acquiring data in the spectral range from 190 to 1100 nanometers. The instrument was purchased in 2006.



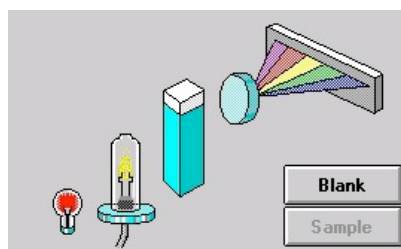
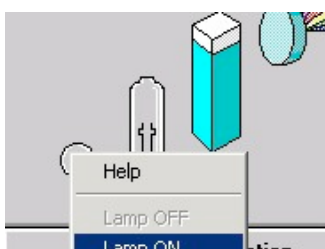
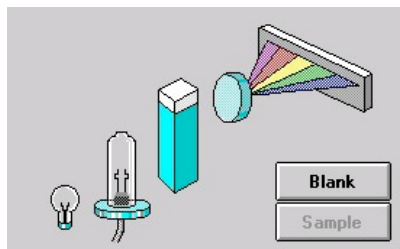
Agilent 8453 UV-Vis Spectrophotometer

Instrument Startup

1. Turn on the PC and monitor.
2. Login as Agilent 8453 with password bi019.
3. Turn on the instrument. The switch is at the lower left corner.
4. Wait until the spectrometer has started to make some clicking noises.
5. Double-click on the UV-Vis program icon to start the program. A program login box will appear. We do not use this feature of the software. **Simply press “cancel” or “ok”.**



6. The software should now open in the standard view.



7. Click on each lamp icon in succession to turn them on. The lamps take a few seconds to light. For critical work, you should allow the lamps to stabilize for about 15 minutes. Routine

measurements can be performed immediately.

Lamp ignition sequence.

8. Use only the visible lamp (blub with red filament when lit) for wavelengths between 350 and 1000 nm, the UV lamp for wavelength below 350nm and both for the full spectrum window.

Acquiring Data

1. With your solvent in a cuvette in the sample holder, select "Blank" from the source pane. If the lamps have been allowed to stabilize, the resulting "instrument blank" spectrum should be straight, with the noise level below ± 0.002 AU in the range from 190 nm to 1000 nm. The noise will be higher in the region above 1000 nm.
2. After running an instrument blank, the previously grayed out "Sample" icon will now be accessible. Place your sample in the cuvette and select "Sample". The measurement will only take a few seconds.

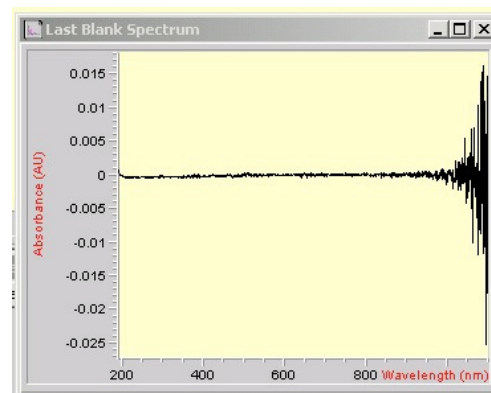


Figure 1. Blank Spectrum

Cuvette mounted in the sample holder

NOTE: Often the software will show a region with missing data when it shows the spectrum. This is a bug in the program however all the data is there. If you export that data you will have all the data. If you zoom the spectra you can see the portion that is not showing in the normal plotting of the spectrum.

Processing Data

Data is not stored automatically. Each successive "Sample" measurement is overlaid in the Overlaid Sample Spectra view and added to the Results Table.

You may manipulate the current data; including zooming in, annotating more or fewer peaks, and printing results, but when you exit the program your data will not be automatically saved.

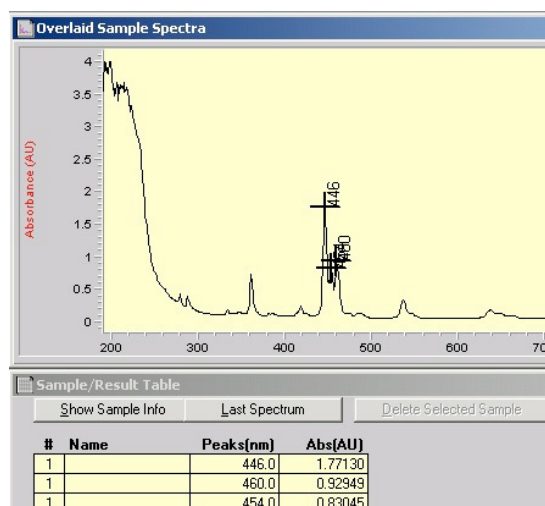
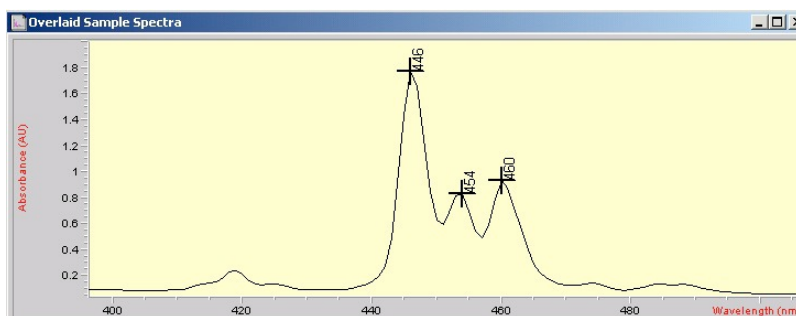


Figure 2. Typical Sample Spectrum

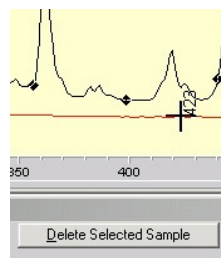
1. A spectral region may be selected by dragging a box around the area of interest.



Use **View -> Reset Current View** to return to the full spectral display.

2. Additional spectra may be acquired and overlaid. To delete a spectrum from the view, select it by clicking on the appropriate trace in the Sample Spectra view. Diamond-shaped points will appear on the selected trace, and a “Delete Selected Sample” button option will appear below the Sample Spectra view. The selected trace may now be deleted.

3. To save data files, click on the **Spectrum to Disk** icon located in the tool bar. Alternatively, select **File -> Save -> Samples As** from the File pull down menu. Only store your files in the data directory.



Finishing Up

1. Exit the ChemStation software.
2. Turn off the instrument power unless you plan to return the same day.
3. Shutdown the computer and turn off the monitor.