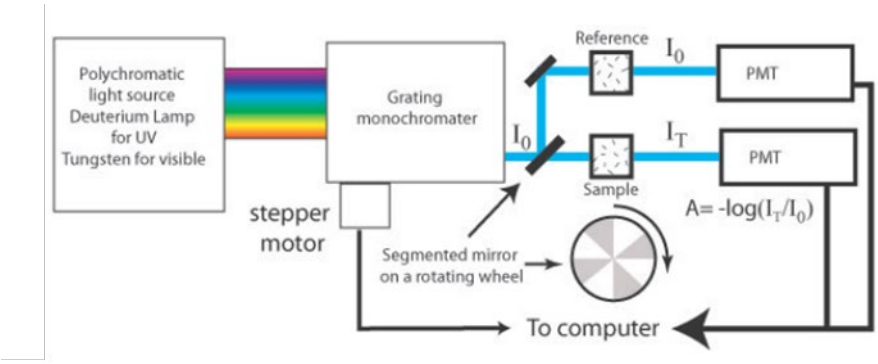
Imaging Facility
Biotechnology Resource Center
Cornell University
BRC_Imaging@cornell.edu

Cary 300 UV-Vis Spectrophotometer User Guide

The Cary UV-Vis spectrophotometer has two light sources: a tungsten lamp to provide 350-800 nm light and a deuterium lamp for the UV. In the UV, the 190-350 nm region is covered. Note that use below 350 nm requires quartz cuvettes because regular glass (borosilicate) does not transmit the UV.



Schematic of a Dual-beam UV-Vis Absorption Spectrophotometer

Turn on Sequence

1. Turn on spectrophotometer. Allow to warm up for 15 minutes.
2. Log in with your net ID and password.
3. Start the Cary Win UV software.

Setup Data Collection and Instrument Parameters

1. Click the Setup button or choose Setup from the Menu line to display the ‘Setup’ dialog box
2. In the ‘Wavelength’ field, enter the relevant wavelength. Range scans from high to low.
3. Set the speed of the data collection by setting the ‘Ave. Time’ and ‘Data Interval’.
    a. The Data Interval is the wavelength increment between data points.
    b. The ‘Scan Rate’ will automatically update when selected.
    c. In the ‘Ave. (averaging) Time’ field, enter the required value. A good starting point is 0.1 seconds.
4. In the ‘SBW’ field, enter the required spectral bandwidth. Unless your method specified another value, use the maximum setting of 2 nm.

5. Select the ordinate mode you require.
    a. Click ‘Abs’ to specify absorbance mode or ‘%T’ to specify percent transmittance.
    b. Enter an upper and lower range value in the ‘Y min.’ and ‘Y max.’ fields
    c. Select ‘Min.’ or ‘Sec.’ to set the abscissa (X).
6. Set up lamp (Setup > Options):
    a. Select *Auto lamps off* to automatically turn off the lamps at the end of the collection.
    b. Click the UV/Vis button to use both lamps.
    c. Enter the wavelength at which you would like the source lamp to change from UV to the visible lamp. The recommended changeover is 350 nm for lamps with a UV cutoff.
    d. Under ‘Beam Mode’, select double beam
7. Set up display options (Setup > Options) :
    a. Under ‘Display Options’, select ‘Individual Data’ to display the collected data of each sample in individual graph boxes, or
    b. Choose ‘Overlay Data’ to superimpose the collected data of each sample in the Scan run in one graph box.
8. Set up baseline correction (Setup > Baseline)
    a. Select Baseline Correction to perform a baseline correction on each sample data point.
9. Ensure no accessories are selected (Setup > Accessories)
10. Set up reporting and printing requirements (Setup > Reports)
11. Set up storage of collected data (Setup > Auto Store)
12. Finish setup.

Data Acquisition (Scan)

1. Zero the instrument: Click Zero. Alternatively, choose Zero from the Commands menu.
2. Measure a baseline:
    a. Click baseline.
    b. Place a blank in the sample compartment and click OK.
    c. After the baseline is collected, the word ‘baseline’ will be displayed in red in the ordinate status box, indicating that you are in baseline correction mode and have a valid baseline file for the correction.
3. Perform the scan:
    a. Click the **Start** button or choose Start from the Commands menu.
    b. The ‘Save As’ dialog box will be displayed. Enter the appropriate ‘File name’ for the data and click Save.
    c. The ‘Sample Name’ dialog box will be displayed. Enter the appropriate name for your sample and click OK.
    d. The Scan run will commence and the corrected trace will be displayed in the Graphics area.

4. At the end of the run, Cary will create the report and also print it, if ‘AutoPrint’ has been selected on the ‘Reports’ page of the ‘Setup’ dialog box.
5. Export your data
    a. On the File menu, click Export report (*.csv).
    b. Enter the ‘File name’ for this read.
    c. Click Save. The data will be stored as an ASCII spreadsheet, with a *.csv file name extension.

The database filename (.DSW) will save the scan that can be opened in the software and cut and pasted to other places. It also saves an excel spreadsheet with the same name (assuming you checked ASCII, above)