

General Operating Procedure for: Cary 60 UV/Vis spectrophotometer

Department of Health and Environmental Sciences

By Yili Cheng

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1. Purpose

The purpose of this GOP is to regulate the operation, usage and maintenance of Cary 60 UV/Vis spectrophotometer.

2. Scope

This GOP applies to all personnel using and managing Cary 60 UV/Vis spectrophotometer.

3. Procedure

3.1 Switching on the machine

- 3.1.1 Turn on the instrument. The switch is at the front panel.
- 3.1.2 Turn on the computer.
- 3.1.3 The indicator lamp will flash with orange light while the instrument is initializing. Wait till the instrument emit green light.
- 3.1.4 Double click on the **Cary WinUV** icon on the desktop.
- 3.1.5 Choose the desired application.

3.2 Scan

3.2.1 Set up a method

Click on the scan icon to open the scan application.

Select **File/Open** Method from the menu to load a previously defined method or select the Setup... Setup button to display the Setup dialog and specify the method parameters for a new method.

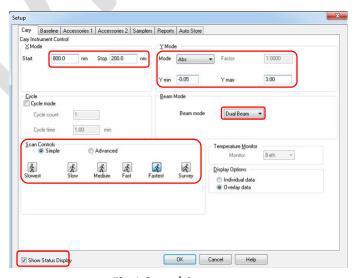


Fig.1 Scan / Cary page

3.2.1.1 Set the instrument parameters

Setup dialog box / Cary page (fig.1)

- a. Enter wavelength Start and Stop. The wavelength range is 190-1100 nm.
- b. Set the Y Mode to "Abs". Enter an upper range and lower range value in the Y min and Y max entry fields to specify the displayed ordinate range.
- c. OPTIONAL- Select Cycle mode and enter cycle count and Cycle time for repetitive scans, e.g. for kinetics run.
- d. Beam Mode MUST be set to Dual Beam.
- e. In the Scan Controls group, select 'Simple' and click a scan speed button. Alternatively, you can select
- 'Advanced' and enter an Ave Time and Data Interval.
- f. Select the Status Display check box so that you can view various instrument parameters during the scan to setup visual system monitoring.
- 3.2.1.2 Set up the baseline correction

Setup dialog box / Baseline page (fig.2)



Fig.2 Scan / Baseline page

Select 'Baseline Correction'. This will force the Cary to perform a baseline correction on the sample data. The correction will be performed on each point before it is displayed. You can either collect a new baseline or use a stored one.

- 3.2.1.3 Make sure that no accessories are selected on Setup dialog box / Accessories 1/2 page.
- 3.2.1.4 Once you are satisfied with your method setup select OK to confirm any changes you have made and close the Setup dialog. Save the method if you plan to use it regularly.

3.2.2 Sample measurement

a. Place the blank solution in the sample compartment. Make sure not to touch the side of the cuvette while doing so. Click to zero the system.

b. Click baseline to set up the baseline correction the system. When prompted, insert the blank sample into the sample compartment and press "OK".

The Cary will collect the baseline scan. After the collection, the word 'baseline' will appear in red in the ordinate status box, indicating that you are in baseline correction mode and you have a valid baseline file for the correction.

c. Insert the sample into the sample compartment. Select the Start button to commence a data collection. The Sample Name dialog is displayed. In the Sample Name dialog, enter the appropriate name and select OK. The scan will commence and the trace will appear in the Graphics area.

3.2.3 Exporting data

a. Use **File/Save Data as...** to save data. File type Data (.DSW) saves a single scan. File type Batch (.BSW) saves all displayed data. File type Method (.MSW) saves only parameters. Save as .CSV text is also available.

b. To copy all displayed graphs to the Windows clipboard, right click in the graph, select Copy Graph from the context menu.

3.3 Simple reads

3.3.1 Set up a method

Open the simple reads application. Click the Setup button . Select 'Read at Wavelength' and enter the required wavelength; type a blank space between wavelengths if you need to read at different wavelengths. Set the Y Mode to "Abs" (fig.3). Click OK to confirm the setting.

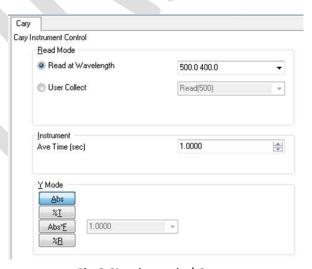


Fig.3 Simple reads / Cary page

3.3.2 Insert the blank into the sample compartment and zero the instrument by clicking the Zero button.

3.3.3 Insert the sample into the sample compartment. Wait while the instrument changes to the specified wavelength. Click the Read button Report area.

3.4 Concentration

3.4.1 Set up a method

3.4.1.1 Set up instrument parameters

Open the concentration application. Click Setup to display the 'Setup' dialog box.

Setup dialog box / Cary page (fig.4)

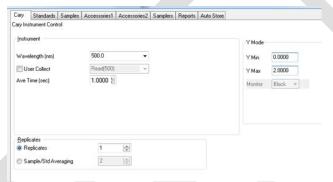


Fig.4 Concentration / Cary page

- a. In the Wavelength field enter the wavelength that you want monitored.
- b. In the Ave Time field enter the required value, usually we will choose 1s.
- c. Select 'Replicates' (if you require them) and set the value to the number of replicates that you are using.
- d. Enter an upper range and lower range value in the Y min and Y max entry fields to specify the displayed ordinate range.
- e. Select the Status Display check box so that you can view various instrument parameters during the scan to setup visual system monitoring.

3.4.1.2 Set up the calibration

Setup dialog box / Standards page (fig.5)

- a. Set the units and number of standards, In the 'Standards' table, enter the concentration of each standard in the 'Conc.' column.
- b. Select the type of curve fitting required for your calibration under 'Fit Type'.

c. Enter the required R2 value or correlation coefficient in the 'Min R2' field. The closer the number is to 1.000 the better the fit. Typically, 0.95 is used.

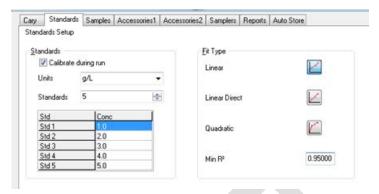


Fig.5 Concentration / Standards page

3.4.1.3 Set up your samples

Setup dialog box | Samples page (fig.6)

Enter the number of samples that you are going to use and the name of each sample. You can enter up to 20 characters for each name.

If you would like the samples to have the same name with a different numeric extension, enter the name in the first sample position and then click the 'Increment' button.

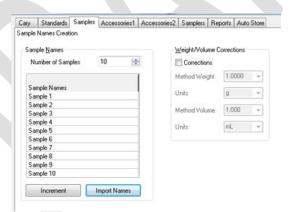


Fig.6 Concentration / Sample page

3.4.1.4 Make sure that no accessories are selected

Setup dialog box / Accessories page

3.4.1.5 Click OK to finish the method setup.

3.4.2 Perform the calibration and measure the samples

a. Insert the blank into the sample compartment and zero the instrument.

b. Click the Start button . Then select the standards and samples to be used in the analysis. Click OK to close The 'Standard/Sample Selection' dialog box.

- c. The 'Cell Loading Guide' dialog box will be displayed. Load your standards as indicated; then click OK to start reading the standards. If additional 'Cell Loading Guide' dialog boxes are displayed, continue to load your standards as indicated and click 'OK' to continue loading the standards. The instrument will measure the standards and calculate the calibration curve.
- d. A 'Cell Loading Guide' will be displayed for the samples. Load your samples as indicated and click OK. The instrument will then measure the samples and calculate their concentration.
- e. Save your data.

3.5 Cleaning

Any spills in the sample compartment should be immediately wiped up.

The exterior surfaces of the Cary 60 spectrophotometer should be kept clean. All cleaning should be done with a soft cloth. If necessary, this cloth can be dampened with water or a mild detergent. Do not use organic solvents or abrasive cleaning agents.

3.6 Switching off the instrument

Exit the Cary WinUV application;

Turn off the instrument and computer.

4. Additional Information

4.1 The WinUV software has extensive on-line Help files.

4.2 Video tutorials are available on the desktop.

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