

# Quick Reference How-To Guide for the DSM CD Models

## Turning on the Olis DSM CD spectrophotometer

- Turn on the lamp cooling box.
- Turn on nitrogen flow to the lamp and monochromator to at least 6 L per min. This ensures protection of the optics from ozone produced by the lamp. Higher nitrogen flow rates should be used if the instrument is to be used in the ultraviolet region.
- Turn on the power supply for the lamp. Allow the power supply to warm up about 5-10 sec. Press and hold the ignite button until lamp ignites. Ensure that the current to the lamp does not exceed 7.5 Amps. Note there is a switch on the back of the power supply to automatically ignite the lamp when the power is turned on.
- Turn on the main power switch at the power strip.
- Turn on the computer and open Globalworks program.
- Select the Data Collection tab and double click on "Conv CD".
- The spectrophotometer will initialize and calibrate.

## Changing modes of detection

- Chosen from in the "Parameters" tab under the "CD Units" menu.
- Milliabsorbance:** Reports data as the difference in absorbance of left circularly polarized light and right polarized light (AbsLeft-AbsRight).
- Molar Extinction:** Reports The difference in extinction coefficients for left and right polarized light (eLeft-eRight).
- Millidegrees:** Reports ellipticity in units of millidegrees
- Molar ellipticity:** Reports molar ellipticity in units of deg cm<sup>2</sup>/dmol. The user must provide the program with protein concentration (g/L), cell pathlength (cm), and mean residue weight (g/mol).
- Single beam:** This is available in the "Modes" tab as an alternative to Circular Dichroism. It is a diagnostic mode, which reports the outputs of the PMT. PMT high volts and slit widths are specified by the user.

## Taking a wavelength scan

- Open "Mode" window and select "scan" data collection mode.
- Ensure that the proper data reduction mode is selected (ie. Circular Dichroism).
- Go to Live display tab.
- Change "wavelength scan range" to desired range (click on the number to highlight).
- Enter the desired number of points to be taken and the integration time per datum (the higher this number is the better the signal to noise ratio, but the longer the scan will take).
- Click on the "collect data" button to begin scan.

## Taking repeated scans

- Under "Repeated Scans" tab, change number of scans to the desired number. Scans can be made automatically as a function of time, or manually. In the auto mode, the time selected is the total time to complete all scans. Manual scans are started by pressing the spacebar.
- Select "Manual" or "Auto" scans on this same page.
- Ensure that time units are correct. These can be changed in the "Mode" tab.
- All repeat scan data will be saved as a single, 3-D data set.

## Taking repeated scans as a function of a temperature script

- In the "Temperature Controller" tab, select the desired temperature script by entering or browsing to the correct file. Files will have the extension '.osf'.
- To edit a script file, click on "Edit Script" and change the appropriate number of scans, temperatures, and integration times. Ensure that the number of scans (Under [Scans]) is less than or equal to the number of command lines (Under [Scan Data]). When using the default temperature file as a template be sure to select "save as" to give your file a new filename.
- Check that temperature controller is set to "On" (In the "Temp Controller" tab)
- In the "Repeated Scans" tab, select "Repeat Scans as a function of Temperature Script". The "Number of Scans" should change to be equal to the number of scans in the temperature script.
- Select "Scan Range", "Number of Points", and "Bandwidth" in the "Live Display" tab.
- Click on "Collect Data" to begin scans.

## Determining mode options for a CD scan

- Detector Options:
  - Manually shut off HV:** High volts to the PMTs are turned on/off in the "Live Display" tab only, when this option switch is selected. Alternatively, HV are shut off at the end of a scan or when the escape key is pressed.
  - Reverse Beam Mode:** Used for testing purposes only.
- Scan Modes:
  - Constant Bandwidth:** Not applicable to the RSM system.
  - Fixed Slit Width:** Slits are fixed at a user-defined value (0-3 mm) and PMT HV are adjusted to keep signal approximately 2 V.
  - Fixed PMT HV:** Not applicable to the RSM system.
- Time Options:
  - Constant Integration Time:** In this option, the user selects a data collection time (0.1-25.5 s/datum). This time spent collecting data is independent of current wavelength or PMT HV, resulting in relatively noisy regions where light intensity is low.

**Integration Time as a Function of HV:** In this option, Time spent collecting data is a function of the current PMT HV, and thus, independently a function of wavelength (since light intensity varies with wavelength). This function is empirical and ensures that more time is spent collecting data under conditions where the PMT HV is high (i.e. low signal/noise). The goal is a constant signal/noise ratio for the entire CD spectrum.

**Time Units:** Time units (h, min, or s) can be chosen for assay mode (see Section 7) or scans as a function of time (see Section 4).

- End of Scan Options

**Return to Starting Wavelength at End of Scan:** This mode readies the spectrophotometer for the next scan after a scan is completed.

**Stop at the End of Scan:** This circumvents the monochromator movement at the end of a scan. This is recommended for photosensitive samples.

## Taking an assay

- Under Mode tab, select “assay” data collection mode.
- Select total time of data collection under live display tab.
- Enter the desired assay wavelength in “Current Wavelength”.
- Enter the number of data points and the integration time per data point.
- To subtract an offset from the data, click on “zero instrument” button. The software will subtract this single point value from all subsequent assays until the apply baseline button is unchecked.
- To begin the assay, click on the “Collect Data” button, and press spacebar when prompted.

## Fitting 2-D data set

- Click on data to be fit.
- If you desire to fit only a portion of this data, click on “Create Data Subset” from the “Tools” menu. Enter the range of the data set you wish to fit. Click on new dataset to select it.
- Select 2-D fits under the “Fits” menu and select the desired model to fit the data. If the model you would like to use is not there, please contact Olis and we will add it.
- The results of the fit will be presented.

## Fitting a 3-D data set

There is a tutorial under the help menu, which describes SVD data processing and fitting.

## Changing the axis scale on a data set

- Select desired data set
- Right-click on graph
- Select “Scale” and enter desired values.

## Viewing more than one set of data

- Open all desired sets of data.
- Select a dataset to be viewed (Move between data sets in the “Experiments” window on the right).
- Select “Copy Slice” under edit menu.
- Select second data set to view.
- Select “Paste Slice” under edit menu.
- To hide a slice from view (and from the printer), select it and select “Hide Slice” under the view menu.

- To switch between hidden slices and viewed slices, select “Swap Hidden/unhidden slices” under view menu.

## Smoothing a data set

- Highlight the data set in the experiment window.
- Left click on the data.
- Right click on the data.
- Select “Smoothing” under “Data Processing” menu.
- Select the appropriate (5-25 point) smooth.
- Alternatively, select the data filter option in the “Live Display” to filter during data collection. This is the same algorithm as used in smoothing.

## Assigning a baseline

- Highlight the desired data set in the experiment window,
- Left click on the data to select it.
- Right click on the data and select “Assign as Baseline” from the pop-up menu. The baseline will continue to be applied until the wavelength range, number of data points, or Assign baseline checkbox have been changed.

## Print a data set?

*Data can be printed as a simple graph:*

- Select chart by clicking on it.
- Select “Print Chart” in “Chart” menu.
- To print landscape see 14d.

*Data can also be printed as a report*

- Select chart by clicking on dataset.
- Select “Print Preview” under “File” menu.
- Select “Color” or “Black and White.”
- Click on “Print” to print.

*Graph can be pasted into a word processing program (i.e. Microsoft Word):*

- Select chart by clicking on dataset.
- Select “Send Chart to Clipboard” under “Chart” tab.
- Open Microsoft Word document.
- Choose “Paste Special” under “Edit” menu.
- Double click on graph to edit it using Microsoft Draw.
- To change printer to landscape, you must change the windows setting.
- Go to “Printers” in the “Start” and “Settings” menu.
- Double click on the desired printer.
- Click on “Properties” in the “Printer” menu.
- Click on the “Printing Preferences” button.
- Click on “Landscape” under the “Features” tab.

## Exporting a 2-D data set

- Select a data set to be exported.
- Right click on the chart and select “ASCII export”
- Enter the filename when prompted.
- Alternatively, data can be exported directly into Excel by selecting “Export to Excel” under the right click menu.

## Turning off the RSM instrument

- Exit the GobaIworks software by selecting “Exit” under the file menu.
- Exit windows and turn off main power switch.
- Turn off power to lamp.
- Turn off nitrogen flow.
- Leave lamp cooling box on for a few minutes to help cool the lamp before turning it off.