# GILFORD<sup>®</sup> RESPONSE I Spectrophotometer



#### — STANDARD OPERATING PROCEDURE —



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## **Introduction**

The Gilford Response I spectrophotometer ► is still a useful teaching instrument, despite its age (early 1980s). The spectrophotometer has excellent optics, utilizing both a quartz-halogen lamp for the visible spectrum and a deterium lamp for the ultraviolet spectrum. The Response unit is the large blue instrument presently located in CB 325. Currently, the Response unit does not have a printer, so all data must be hand-tabulated until a suitable printer can be found.



## System Specs

- The Response Unit uses a quartz-halogen lamp for the visible region (about 400 nm to 750 nm), and a deuterium lamp for the ultraviolet region (200 nm to about 400 nm)
- The system is equipped with a six-position, water-heated cuvette holder, which is designed for applications that require temperature control. No temperature control unit is presently equipped, but the holder has the necessary adaptor to accommodate one.
- The wavelength range of the Response is **200 nm to 750 nm**.
- <u>Only</u> 10-mm square cuvettes can be used. Round cuvettes will not fit.
- The spectrophotometer comes with a set of two aperture adjustors: 1 mm x 2 mm, and 2 mm x 4 mm. Scans can be run with either slit in place or wide open.
- The user can select spectral bandwidths of 0.5nm, 1.0nm, or 4.0nm.

### DANGER

Do not operate Response in an atmosphere containing explosive concentrations of oxygen, hydrogen, or other flammable gases. Components of the instrument include switches, relays, and other electrical parts that may generate sparks.

## DANGER

The instrument is designed to be grounded through the power cord for safe operation. To ensure continuing safety of operating personnel the instrument must be connected to an electrical outlet that has an effective ground connection.

#### WARNING

As with any instrument, use extreme caution not to spill samples! Spilled solutions may result in damage to electrical or optical components. If an accidental spill does occur, promptly clean it up.

#### WARNING

Do not leave sample cuvettes in the compartment for longer than is needed to take readings, especially samples prone to evaporation or fuming. Vapors may damage operational components.

### CAUTION

Always let the lamp warm up for 15 minutes before the first reading is taken.

## CAUTION

Always hold cuvettes by their frosted sides. Make sure the clear sides are completely free of dust and scratches to ensure accurate light transmission.

### CAUTION

If you are scanning in the UV region, you *must* use quartz cuvettes. Glass cuvettes and plastic cuvettes are not fully transparent to ultraviolet light.

## CAUTION

Your reference sample must contain the same solvent as your samples.

## **Basic Operation**

#### Startup Procedure

- 1. Make sure the spectrophotometer is plugged in. The cord is located on the far backside.
- 2. Turn the unit on by flipping the power switch to the ON position.



- 3. When the screen warms up (after only a few seconds), you will see a blank screen with the word GILFORD flashing. The unit is now in standby mode. Press ENTER to continue.
- 4. The self-diagnostic will automatically start, and takes approximately 60 to 90 seconds to complete. The diagnostic is complete when USE DYNAMIC KEYS flashes at the bottom of the screen ▼. NOTE: all tests should read PASSED except for PRINTER and REMOTE COMM, which will read FAILED. This just means that the printer and remote computer are not connected.

| SELF DIAGNOSTICS                                    |       |
|---|-------|
| PROM - PASSED<br>RAM - PASSED<br>EAROM - PASSED     |       |
| Choose OPTICAL<br>CALI. By pressing<br>this button. |       |
|   | 10778 |

5. Choose OPTICAL CALI. to run the optical calibration  $\mathbf{v}$ .

**CAUTION** *Make sure you run the optical calibration every time you turn on the unit.* 

| SELF DIAGNOSTICS   |  |
|--|--|
| PROM – PASSED<br>Ram – Passed                            |  |
| EAROM - PASSED<br>Local I/O - Passed<br>Printer - Passed | Diagnostics are<br>complete when<br>this appears |
| REMOTE COMM - FAILED                                     | SKIP CALI.                                       |

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6. If the calibration is successful, then the operational guidelines screen is displayed  $\mathbf{v}$ .

| PREAMBLE                                       |                               |   |
|--|-------------------------------|---|
|  |                               |   |
| > OPERATOR =                                   | VIS LAMP                      | < |
| > DATE = 01-JAN-1985                           | U¥ LAMP                       | < |
| > TIME = 12:15                                 | UV AND VIS LAMPS              | < |
|  | DIAGNOSTICS                   | < |
| > CUVETTE OFFSETS                              | COMPUTER SETUP                | < |
| > ACCESSORY:<br>6 POSITION CUVETTE HOLDER (10) | MM.)<br>PRESS ENTER TO RESUME |   |

7. After reading the operating guidelines, press ENTER to continue. The preamble page is now displayed ▼.

| OPERATING GUIDELINES  |  |
|---|--|
| 1. USE DYNAMIC KEYS TO SELECT PROGRAM OR PARAMETER              |  |
| 2. COMPLETE NUMERICAL ENTRIES WITH THE "ENTER" KEY              |  |
| 3. "STATUS" KEY PROVIDES INSTRUMENT SETTINGS                    |  |
| 4. "LIBRARY" KEY PERWITS PROGRAM SELECTION                      |  |
| 5. "COPY" KEY PROVIDES PERMANENT RECORD OF<br>Programs and data |  |
| 6. "STOP" KEY INTERRUPTS DATA COLLECTION                        |  |
| PRESS ENTER TO RESUME   |  |
|   |  |

8. First, select VIS LAMP, UV LAMP or UV AND VIS LAMPS to select the lamps you will use.

**CAUTION** It is important to select lamps now so the lamps have adequate time to warm up.

- 9. If desired, select OPERATOR ▼ to enter an operator identification number (accepted values are integers 1 to 9999).
- 10. If desired, select DATE ▼ to change the date. The system only accepts dates from Jan 1, 1984 to Dec 31, 1999.

| PREAMBLE                                       |                            |                |
|--|----------------------------|----------------|
|  |                            |                |
| > OPERATOR =                                   | VIS LAMP                   |                |
| > DATE = 01-JAN-1985                           | UV LAMP                    | Select lamp(s) |
| > TIME = 12:15                                 | UV AND VIS LAM             |                |
| >  | DIAGNOSTICS                |                |
| > CUVETTE OFFSETS                              | COMPUTER SETUP             |                |
| > ACCESSORY:<br>6 POSITION CUVETTE HOLDER (10M | M.)<br>PRESS ENTER TO RESU |                |

| F   | PREAMBLE |                                 |   |                         |
|---|----------|---------------------------------|---|-------------------------|
| If desired, change<br>OPERATOR here<br>> OPERATOR = |          | VIS LAMP                        | < |                         |
| > DATE = 01-JAN-1985                                |          | UV LAMP                         | ( |                         |
| If desired, change<br>DATE here                     |          | UV AND VIS LAMPS<br>Diagnostics | ( |                         |
| <pre>&gt; CUVETTE OFFSETS &gt; ACCESSORY:</pre>     |          | COMPUTER SETUP                  | < | 1. You                  |
| 6 POSITION CUVETTE HOLD                             |          | ENTER TO RESUME                 |   | have<br>succe<br>ssfull |
|   |          |                                 |   | y set                   |

up the system. It is ready to begin sampling.

#### Running a Spectral Scan

1. Press the LIBRARY key **•**.



- 2. Select WAVELENGTH SCAN.
- 3. Select STARTING WAVELENGTH ▼, type in the maximum wavelength you want to scan, and press ENTER.
- 4. Select ENDING WAVELENGTH ▼, type in the minimum wavelength you want to scan, and press ENTER.

| WAVELENGTH SCAN 6 POSITION CUVETTE HOLDER (10MM.)<br>OPERATOR = DATE = 01-JAN-1985 TIME = 12:38<br>BANDWIDTH = 1.0 NM (                                     |  |
|---|--|
| Change starting = 0.5 NM<br>wavelength here = 1 PHOTOMETER = (  |  |
| wavelength here       =       1       PHOTOMETER =          DELHT TIME (MIN)       =       0.0       ABSORBANCE         > STARTING       REFERENCE(S) = ONE |  |
| WAVELENGTH = 750.0 NM<br>> ENDING SAMPLE(S) = ONE (   |  |
| Change ending<br>wavelength here; KEY PRESS ENTER TO SETUP  |  |
| TO EDIT PARAMETERS  |  |
|   |  |

- 5. If the bandwidth displayed is not the one you want to use, select BANDWIDTH, and select the bandwidth you want to use. The available choices are 0.5 NM, 1.0 NM, and 4.0 NM.
- If the scan increment displayed is not the one you want to use, select SCAN INCREMENT to change the resolution. If you select a small scan increment, you will get high resolution in your readings, but the scans will take a long time. The available choices for scan increment are 0.1 NM, 0.2 NM, 0.5 NM, and FAST SCAN.
- 7. If the photometer mode displayed is not the one you want to use, select PHOTOMETER to change to ABSORBANCE, TRANSMITTANCE, or DIFFERENCE SPECTRA.
- 8. To change the number of references you will use, select REFERENCE(S) = , type in the number of references, and press ENTER.
- 9. To change the number of samples you want to read, select SAMPLES(S) =, type in

| OPERATOR =       DATE = 01-JAN-1935       TIME = 12:33         BANDWIDTH =       1.0 NM         Any of these parameters may be changed by pressing the corresponding button to the right       SCAN INCREMENT         Methods       = 0.5 NM         PHOTOMETER =       (         ABSORBANCE       (         REFERENCE(S) = 0NE       (         VSE DYNAMIC KEY       STORE PROGRAM         TO EDIT PARAMETERS       PRESS ENTER TO SETUP | Any of these<br>parameters may be<br>changed by pressing<br>the corresponding<br>button to the right<br>USE DYNAMIC KEY<br>SCAN INCREMENT<br>= 0.5 NM<br>PHOTOMETER =<br>ABSORBANCE<br>REFERENCE(S) = ONE<br>STORE PROGRAM<br>(<br>PRESS ENTER TO SETUP |  |
|---|---|--|
|---|---|--|

the number of samples, and press ENTER.

10. If all of the settings suit you, load the cuvettes into the holder (refer to "Loading the Cuvettes" if needed.)

- 11. Press ENTER to set up the parameters. This will take 15 30 seconds. Note: once you do this, you will be unable to go back and edit any parameters without beginning a completely new scan.
- 12. When prompted, press ENTER to begin the run.
- 13. An empty graph will initially be displayed, but a curve will be drawn over the next



few minutes. It commonly takes 1-5 minutes to complete a scan. (Remember, the smaller the increment you selected, the longer the scan will take.)

- 14. If desired, select PEAK ID to see a list of the peaks, their associated wavelengths, and their maximum values. Select GRAPHICS to return to the graphical display when you are finished.
- 15. If desired, you can select GRAPHIC CURSOR to see any coordinate on the graph that you want to. The cursor starts on the left edge of the screen—hold the right arrow key vto move the cursor to the left and the left arrow key vto move the cursor to the left. The coordinate value will be displayed at the top of the screen. Select GRAPHIC CURSOR again to exit this mode and return to the graphical display.

16. If desired, select REFORMAT to change the scale ▼. When finished, select RETURN : GRAPHIC to return to the graphical display and apply the reformatting

| SAMPLE REFORMAT OPTIONS  |
|--|
| If you ran more than one sample, you can select<br>which graph to display here |
| ( SAMPLE NO.1 Choose between absorbance, % transmittance,                      |
| ( ABSORDER log absorbance, 1st derivative, and 2nd derivative.                 |
| ( WAVELENGTH = 340.0, 750.0<br>Changes the x-axis scale                        |
| ( PHOTOMETER = -0.01, 4.00<br>( CHANCE DATA FORMAT Changes the y-axis scale    |
|  |
| ( RETURN : GRAPHIC<br>Returns to graph   |

options you have changed.

17. The spectral scan is complete. You can reformat the graph many times until it suits your needs.





#### Setting up a Calibration Curve

- 1. Press the LIBRARY key  $\checkmark$ .
- 2. If needed, select NUMBER OF STANDARDS to change the number of standards you will use. NOTE: You must have at least 3 unique standards.
- 3. If needed, select WAVELENGTH to change the wavelength to scan.
- 4. If desired, select BANDWIDTH to change the bandwidth you want to use. The available choices are 0.5 NM, 1.0 NM, and 4.0 NM.

5. Load the reference cuvette and the standard cuvettes into the holder (refer to "Loading the Cuvettes" if needed.)

6. Note the position of each standard you insert. You will later be prompted to enter

the concentrations of standards in the order you positioned them.

- ANALYSIS = 1958  $3.000 - \frac{1}{0} -$
- 7. If all of the settings suit you, press ENTER to set up the parameters. This will take

15 - 30 seconds.

- 8. When prompted, press ENTER to begin the scan.
- 9. You will be prompted for the concentration of each standard, beginning with the second slot from the left and proceeding right. Type the concentration of the corresponding standard when you are prompted for it and press ENTER.

10. When all of the standards have been entered, press ENTER. A calibration curve such as the one below will be displayed  $\checkmark$ .

- 11. If you want to determine the concentration of unknown sample(s) based on this calibration, select SAMPLES. Load the samples into the cuvette holder (refer to "Loading the Cuvettes" if needed.) and press ENTER. The concentration of the sample(s) will be displayed. NOTE: If you have empty slots, enter a concentration of "0" when prompted, and ignore the concentration it determines. NOTE: An asterisk (\*) indicates the calculated absorbance is outside the range of your standards.
- 12. The calibration curve is complete.

#### Standby Mode

- 1. If you are going to be away for more than a few minutes, it is a good idea to put the instrument in <u>standby mode</u>. To do this, press the LIBRARY key, and select STANDBY.
- 2. To exit standby mode, press ENTER. The procedure is now the same as listed in "Startup Procedure".

#### Shutting Down the System

- 1. Make sure the inside compartment is clean and remove any cuvettes. Close the inside compartment lid.
- 2. Turn the power switch to the OFF position  $\checkmark$ .

- 3. Place the tan cover over the screen and keypad.
- 4. Make sure the general area is clean and free of spills.



1. Open the inside compartment  $\mathbf{v}$ .



2. Remove the cover from the cuvette holder  $\mathbf{v}$ .



3. The inside compartment will look like this: •



Load the cuvette containing the reference into the first (leftmost) slot ▼. Make sure that the clear sides of the cuvette are facing toward the front and back. The reference solution must <u>always</u> be in the first slot.



5. Load your sample(s) or standard(s) into the next slot(s) in the same manner. Leave no empty slots between the sample cuvette(s) and the reference ▼



6. If desired, insert an aperture adjuster into the slot  $\mathbf{v}$ .



7. Replace the cuvette holder cover and close the compartment. Proceed with scans.