

## Appendix #1<sup>1</sup>

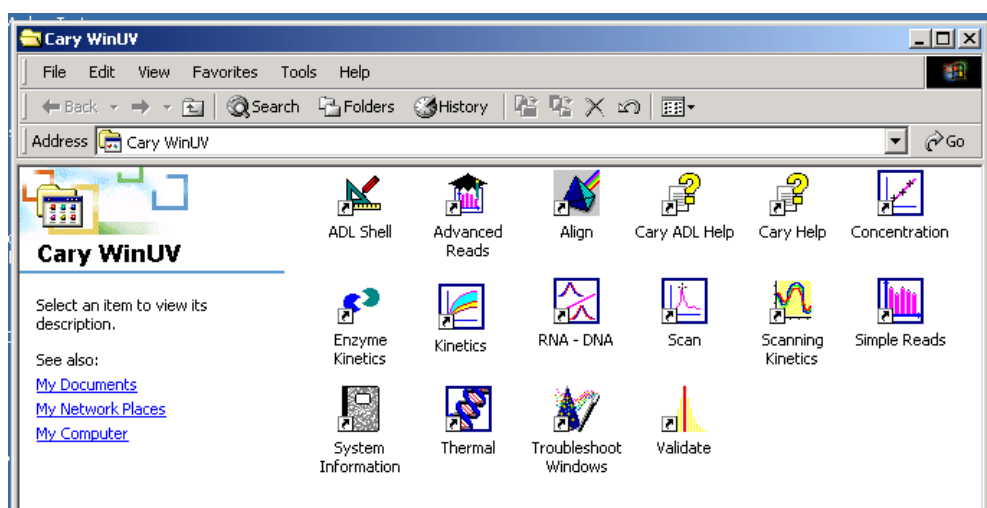
### Guidelines for measuring an UV-Vis spectrum using the Cary 100 Scan UV-Visible Spectrophotometer and Lamber-Beer law.

Mircea D. Gheorghiu

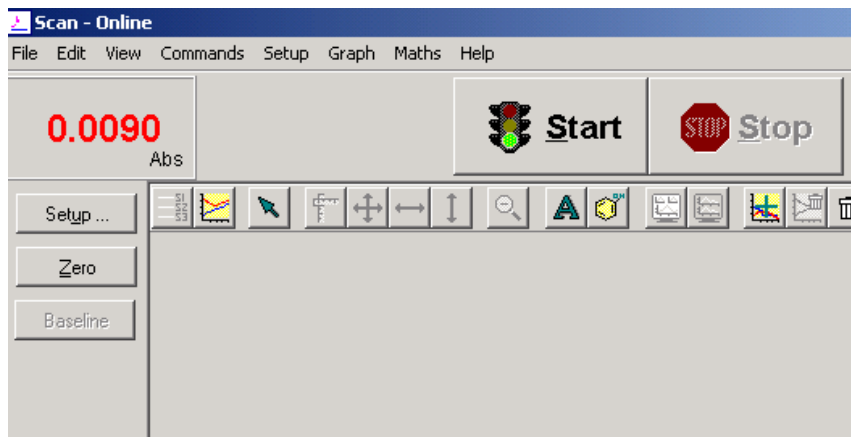
1. On the Dell Optiplex GX 150 Monitor, if CaryWinUV is not launched, click on the icon:



then click on **Scan** icon.

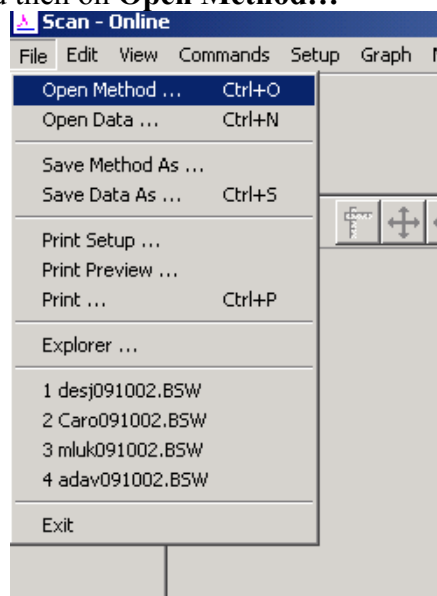


2. The **Scan** window pops-up:

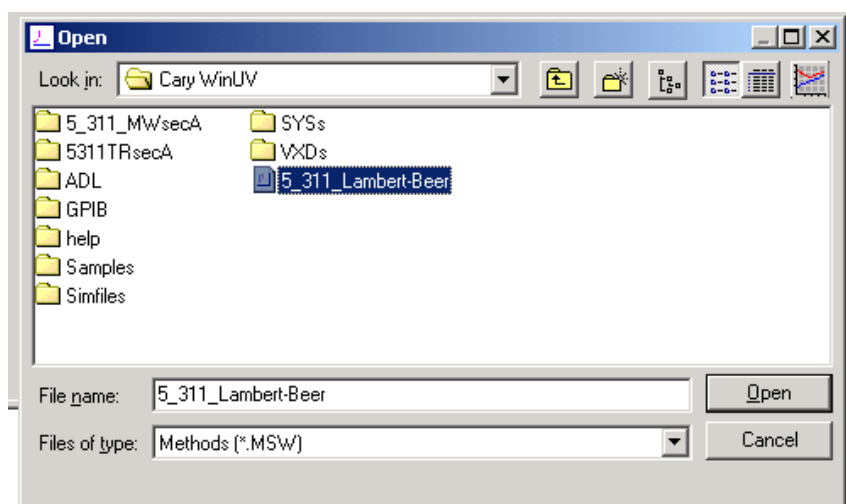


<sup>1</sup> Please address comments to Dr. Mircea D. Gheorghiu ([mircea@mit.edu](mailto:mircea@mit.edu)).

Click on **File** and then on **Open Method...**



Load the **5\_311\_Lambert-Beer** setup file.



3. Click on **Setup...** button. Click on **Cary** tab.

- In **X Mode** go to **Mode**, select **nanometers** from the drop down list. Type into **Start** window 360.00 nm and into **Stop** window 550.00 nm.
- In **Y Mode**, for **Mode** select **Abs**, for **Y min** 0.00 and **Y max** 1.20.

## 5.311 Kinetics - Appendix 1

**Setup**

Cary | Options | Baseline | Accessories 1 | Accessories 2 | Accessories 3 | Reports | Auto Store

Cary Instrument Control

☒ X Mode

Mode: Nanometers

Start: 360.00 nm Stop: 550.00 nm

☐ Y Mode

Mode: Abs Factor: 1.0000

Y min: 0.00 Y max: 1.20

☐ Cycle

☐ Cycle mode

Cycle count: 1

Cycle time: 1.00 min

Scan Controls

Ave time (s): 0.100

Data interval (nm): 1.000

Scan rate (nm/min): 600.000

Temperature Monitor

Monitor: Block

☐ Show Status Display

OK Cancel Help

Type as **Scan Controls**, **Ave Time (s)** 0.100, **Data intervals (nm)** 1.000, and **Scan Rate (nm/min)** as 600.000.

4. Click on the **Options** tab. Set the **SBW** to 2.0 nm. In the **Display Options**, click on the radio button **Overlay data**.

Cary | Options | Baseline | Accessories 1 | Accessories 2 | Accessories 3 | Reports | Auto Store

Advanced Settings

SBW/Energy

SBW (nm): 2.0

Beam mode: Double

Energy: 1.00

Signal-to-Noise

☐ Signal-to-noise mode

Acceptable S/N: 10000.00

S/N timeout (s): 0.100

Source

☒ Auto lamps off

UV Vis UV-Vis

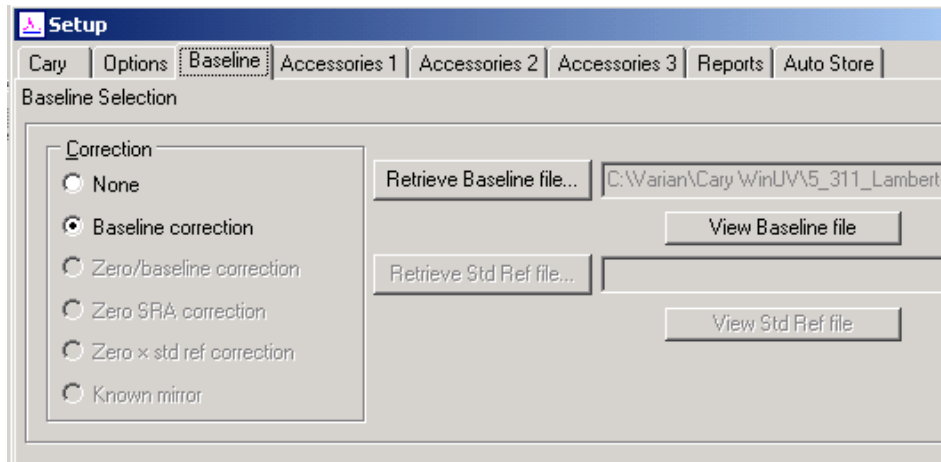
Source changeover (nm): 350.00

Display Options

☐ Individual data

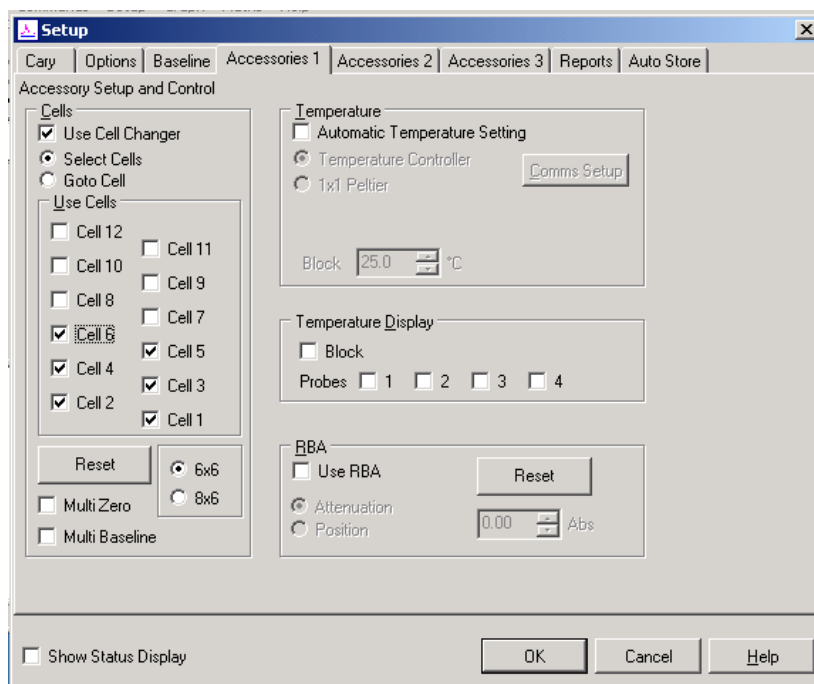
☒ Overlay data

- Go to the **Baseline** tab. Select the radio button **Baseline correction**.

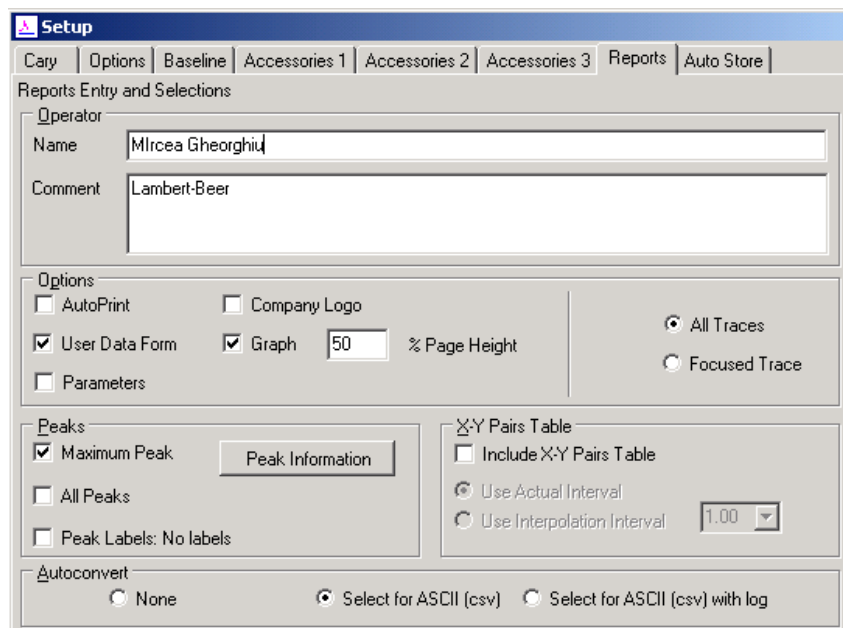


- In the **Accessories 1** window check the following:

- Accessory Setup and Control:** Use Cell Changer, Select Cells, Cell 1 through Cell 5 (for the five concentrations of  $K_3Fe[(CN)_6]$ ).



- Click on the **Reports** tab. Provide the **Operator's Name** and the pertinent **comment**. Check for **Options** on **User Data Form**, **Graph**, **All Traces**. In the **Peaks**, check on the **Maximum Peak**. Click on the radio button **Select for ASCII (csv)**.



**Setup**

Carv | Options | Baseline | Accessories 1 | Accessories 2 | Accessories 3 | **Reports** | Auto Store

Reports Entry and Selections

**Operator**

Name: Mircea Gheorghiu

Comment: Lambert-Beer

**Options**

☐ AutoPrint ☐ Company Logo

☒ User Data Form ☒ Graph 50 % Page Height

☐ Parameters

**Peaks**

☒ Maximum Peak Peak Information

☐ All Peaks

☐ Peak Labels: No labels

**X-Y Pairs Table**

☐ Include X-Y Pairs Table

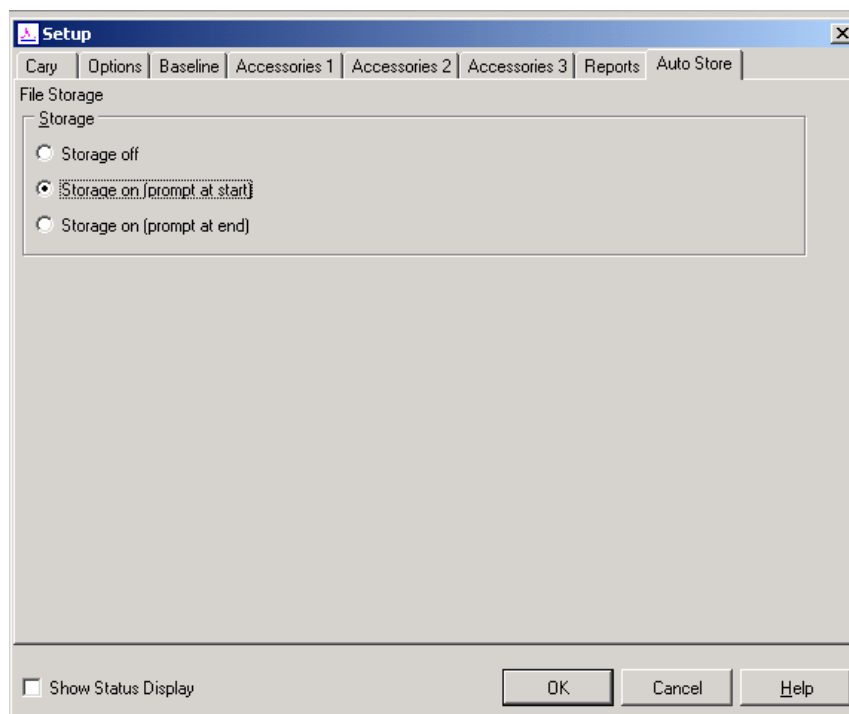
☒ Use Actual Interval

☐ Use Interpolation Interval 1.00

**Autoconvert**

☐ None ☒ Select for ASCII (csv) ☐ Select for ASCII (csv) with log

8. Click on the **Auto Store** tab, and choose the radio button **Storage on (prompt at start)**. Click **OK** button.



**Setup**

Carv | Options | Baseline | Accessories 1 | Accessories 2 | Accessories 3 | Reports | **Auto Store**

File Storage

**Storage**

☐ Storage off

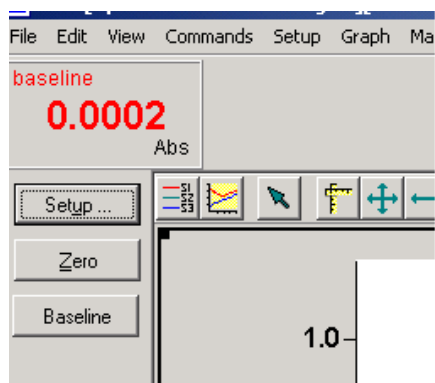
☒ Storage on (prompt at start)

☐ Storage on (prompt at end)

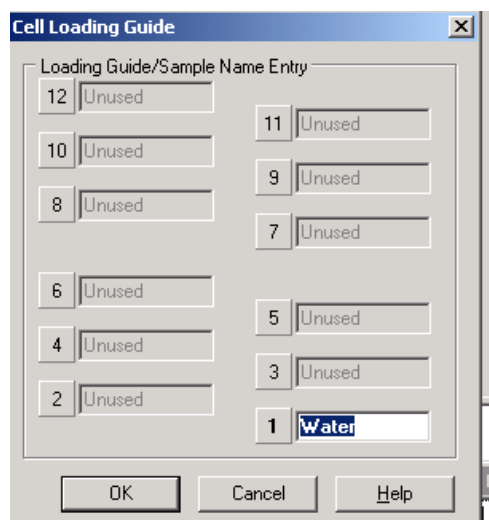
☐ Show Status Display

OK Cancel Help

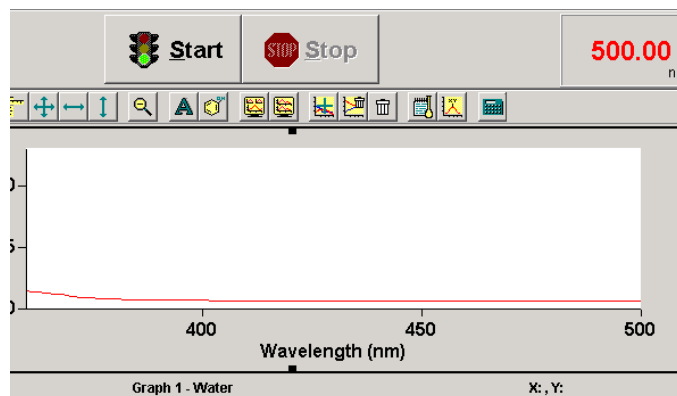
9. Click on **Baseline** button:



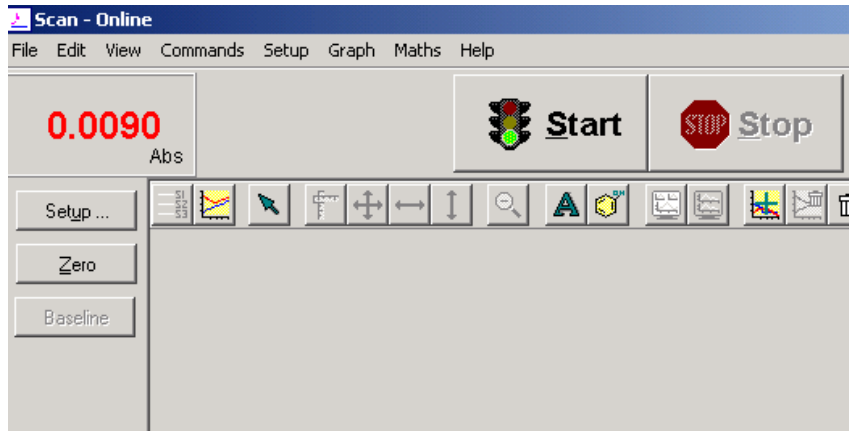
The **Cell Loading Guide** pops-up. Name the cell 1 as **Water**. Insert the cuvette with water, close the lid and click on **OK**.



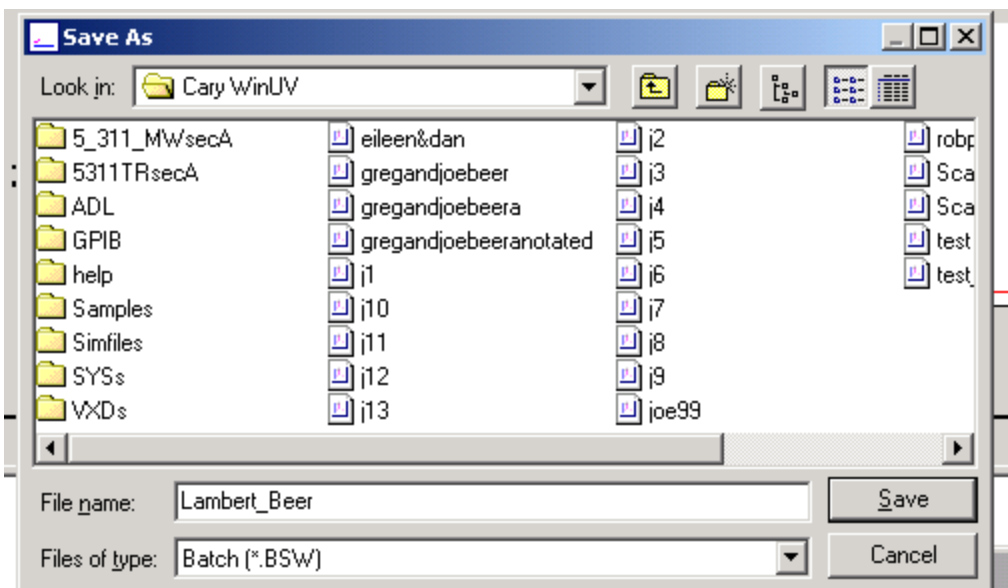
On the monitor screen the graph **Absorbance** (water + cuvette) versus **wavelength** curve is displayed.



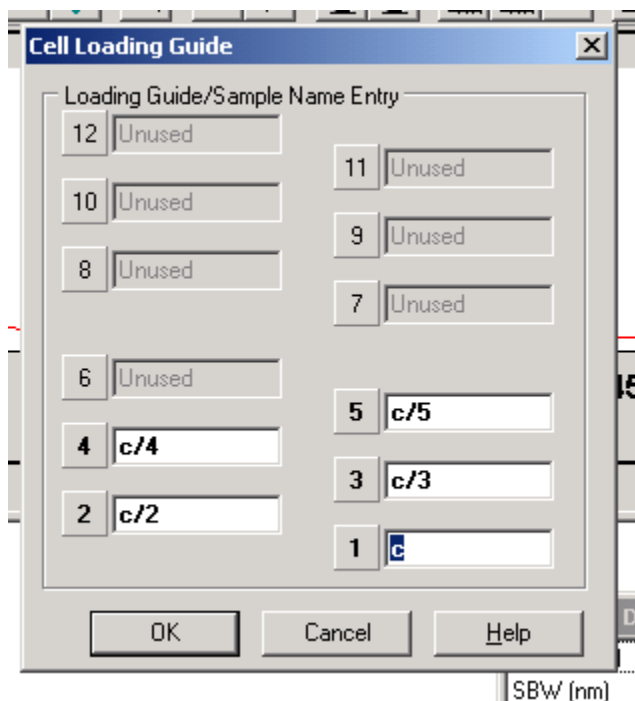
10. Click on **Start** button to begin the **Scan**.



a. **Save As..** your file into the directory of your section (MW or TR) and Group (A or B), or on your floppy disc or on both the harddrive and your floppy disk. Click **Save**.



b. In the **Cell Loading Guide** that pops-up provide the names of the samples that are subjected to the Scan. For example, I am suggesting **c**, then **c/2** (for the half diluted sample), **c/3**, **c/4** and **c/5**. Insert the samples in the cell holder in that order, close lid and click on **OK**.



**Cell Loading Guide**

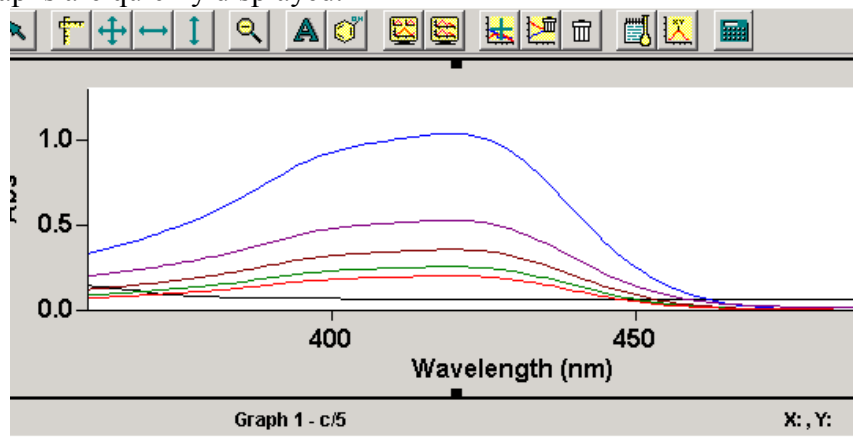
Loading Guide/Sample Name Entry

12	Unused	11	Unused
10	Unused	9	Unused
8	Unused	7	Unused
6	Unused	5	c/5
4	c/4	3	c/3
2	c/2	1	c

OK Cancel Help

SBW (nm)

The five graphs are quickly displayed.



In the Report section is printed information concerning the sample name and the measured maximum **Absorbance** at the corresponding wavelength.



Wavelength (nm)	Abs
420.00	1.0356
Sample Name: c/2	
Collection Time	9/13/2002 2:24:54 PM
Peak Table	
Peak Style	Maximum Peak
Peak Threshold	0.0100
Range	500.00nm to 360.00nm
Wavelength (nm)	Abs
419.00	0.5269
Sample Name: c/3	
Collection Time	9/13/2002 2:25:17 PM

Write down in your notebook all the five values of the respective maximum absorbances. Draw the Lambert Beer **Absorbance** versus **concentration** plot (include the 0,0 pair of points too). From the slope of the least squared straight line, calculate the  $\epsilon$  that will be used in the curve fitting the kinetics data to the second order kinetics equation.