Measuring the energetics of a hydrogen bond through vibrational spectroscopy

1 Introduction

Hydrogen bonds impact many different areas of science, including, but not limited to, chemistry, biology, and biophysics. In particular, they play a major role in determining the properties of water, and defining the structure of proteins and DNA. They are relatively weak bonds forming and breaking on an ultrafast time scale; i.e, 10^{-15} seconds.

At a given temperature, a polar molecule in water will be in equilibrium between its free form and its water-bonded form. The constant for this equilibrium may be inferred by measuring the relative concentration of each molecular species at equilibrium. In the present experiments, Fourier-Transform Infrared Spectroscopy is used to determine the energetics between methyl acetate (CH_3COOCH_3) and deuterium oxide (D_2O) by measuring the ratio between the D-bonded form and the free form.

2 Background

The carbonyl (C=O) stretch vibrational band of methyl acetate ($\sim 1700\ cm^{-1}$) in deuterium oxide is composed of two overlapping bands due to H-bond formation with one [$1HB$] or two [$2HB$] water molecules. Scheme 1 below shows the equilibrium between the $2HB$ and $1HB$ forms.

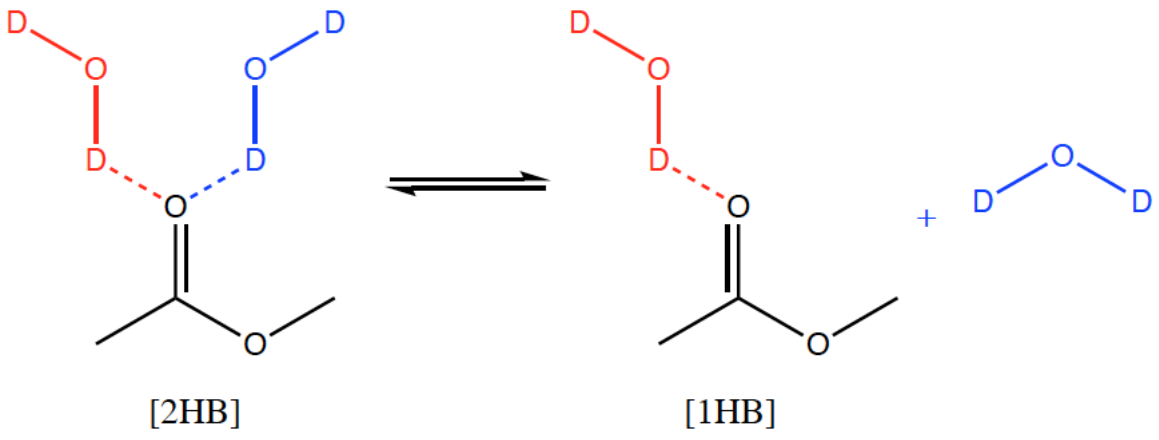

Figure 1: Hydrogen bond making and breaking of methyl acetate in deuterium oxide.

At equilibrium, the ratio between the $[2HB]_{eq}$ and $[1HB]_{eq}$ concentrations defines the

equilibrium constant, K_{eq}, for the inter-conversion between the two species, following:

$$K_{eq} = \frac{a^{eq}_{1HB} \cdot a^{eq}_{D_2O}}{a^{eq}_{2HB}} = \frac{[1HB]_{eq}}{[2HB]_{eq}} \tag{1}$$

where the a^{eq}_i are the reactants and the products activities at equilibrium. The change in standard Gibbs-free energy for the reaction is given by:

$$\Delta_r G^o = \Delta_r H^o - T\Delta_r S^o = -RTln(K_{eq}) \tag{2}$$

where $\Delta_r H^o$ and $\Delta_r S^o$ are the enthalpy and the entropy changes at standard conditions, T is the temperature in Kelvin, and R is the ideal gas constant. From equations (1) and (2) we infer the temperature dependence of the ratio of the $1HB$ and $2HB$ concentrations:

$$ln\left(\frac{[1HB]_{eq}}{[2HB]_{eq}}\right) = -\frac{\Delta_r H^o}{RT} + \frac{\Delta_r S^o}{R} \tag{3}$$

When using absorption spectroscopy, the concentration of the $1HB$ and $2HB$ species are given by the Lambert-Beer law:

$$A_i = \mathcal{E}_i \cdot b \cdot [i] \tag{4}$$

where $A_i, \mathcal{E}_i$, and $[i]$ are the absorbance, the molar absorptivity coefficient, and the concentration of the i species, respectively and b is the path length of the absorption cell. Combining equations (3) and (4) we obtain:

$$ln\left(\frac{A_{1HB}}{A_{2HB}}\right) = -\frac{\Delta_r H^o}{RT} + \frac{\Delta_r S^o}{R} + ln\frac{\mathcal{E}_{1HB}}{\mathcal{E}_{2HB}} \tag{5}$$

Equation (5) relates the relative absorbance of the hydrogen-bonded species with the thermodynamics of the chemical process. If a plot of $ln\left(\frac{A_{1HB}}{A_{2HB}}\right)$ as a function of $\frac{1}{T}$ is plotted, equation 5 should produce a straight line with $y = ln\left(\frac{A_{1HB}}{A_{2HB}}\right)$, $m = -\frac{\Delta_r H^o}{R}$, and $b = \frac{\Delta_r S^o}{R} + ln\frac{\mathcal{E}_{1HB}}{\mathcal{E}_{2HB}}$. Since $\mathcal{E}_{1HB} \approx \mathcal{E}_{2HB}$, then the y-intercept, $b = \frac{\Delta_r S^o}{R}$.

The experiment will be performed using in situ FTIR spectroscopy in a temperature-regulated reactor. The absorption spectra will be analyzed using MATLAB in order to infer the energetics of the hydrogen bond.

3 Experimental

Supplies: methyl acetate, deuterium oxide (D_2O), tetrahydrofuran (THF), 0.1 M hydrochloric acid, methanol, acetone, lens paper, $20 - 200\ \mu L$ pipette, and sampling vials.

Instrument: In situ FTIR spectrometer (ReactIR) and temperature-controlled reactor (EasyMax).

Hazard: methyl acetate is an irritant and flammable, while methanol is toxic and flammable.

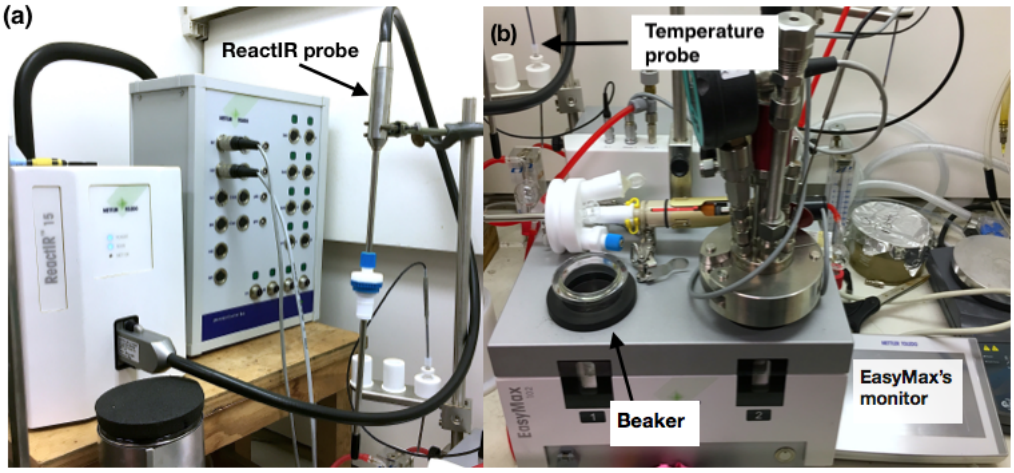

Figure 2: Pictures of the instruments that will be utilized in this experiment: (a) ReactIR and (b) EasyMax.

3.1 Initial instrument setup

1. Switch On EasyMax and set the reactor temperature (T_r) to 10^oC. Fill the beaker in the EasyMax with $\sim 50mL$ of water. Place EasyMax temperature probe into the glass flask of EasyMax.

2. Click of the iCIR software icon in the computer to launch the software. Then click on *Instrument* $\rightarrow$ *Configure*. The status of the instrument should be ready and make sure that the *Probe Interface* is **AgX Fiber**.

3. Under the 'Probe Tip' section, make sure that the *Probe Tip* used is DiComp (Diamond) — this is for ReactIR probe, the resolution is 4 wavenumbers, and the 'scan per sample' is 256.

4. Click on 'Collect Background' and a new window will pop-out. Make sure that the 'MCT detector' and the 'Alignment bar' is **Green**. Click on 'Collect Background' and then click on 'Done' once it is completed.

5. Click on *Clean Probe* and then click 'Start'. If the measurement is within range, and a flat line is produced, click 'Next' and collect another background and then click on 'Done'.

6. If the measurement is not flat nor within range, the probe need to be cleaned. Clean the ReactIR probe by immersing the probe tip in the following solution/solvent: 0.1 M hydrochloric acid, methanol, and acetone. Wipe the probe tip with Kimwipe.

3.2 Effect of solvent on carbonyl frequency

1. Place the sampling vial with 4 mL of solvent (THF) into the beaker in the EasyMax. Place the ReactIR probe into the sampling vial. Make sure that the tip of the ReactIR is completely submerged in the solvent and that the probe is **vertical**.

2. Start a new experiment by click on 'File' → 'New' → 'Quick Start'.

3. Change the file name to something appropriate, and make sure your file folder is in folder 'CHEM349' on 'Desktop'.

4. Change your 'Scan per sample' to 256 and change the 'Duration' to 15 minutes. Then click 'Create'. Once the T_r reached the targeted temperature click on the 'Play' button. The instrument will scan the sample 256 times before it generate a spectrum. Get 3 spectra of the solvent before proceed.

5. Add 100 μL of methyl acetate into the sampling vial to obtain a solution of 0.3 M of methyl acetate. **Be careful not to touch the probe**. Get 3 spectra of methyl acetate in THF solution.

6. Click on the 'Stop' button to stop taking measurement. Go to 'Notes' and "pin spectra" by clicking on the 'pin' buttons. Go to 'Spectra' tab and choose 'Export all pinned spectra'. On the selection menu, choose 'pinned spectra', choose 'CSV' for format, 'raw' for data, and click on "Export".

7. Dispose of the methyl acetate-THF solution, clean the ReactIR probe following the procedure described above, and take a new background. Repeat procedures 1 to 5 in subsection 3.2 with deuterium oxide as the solvent. Click on the 'Pause' button after you have obtained 3 spectra of methyl acetate in D_2O solution at 10^oC. Proceed to subsection 3.3.

3.3 Effect of temperature on the carbonyl spectrum

1. Write down the name of the files that corresponds to the spectra of methyl acetate-D_2O solution at 10^oC.

2. Set the T_r on EasyMax to 20^oC. Once T_r reached the targeted temperature click on the 'Play' button to get spectra of methyl acetate-D_2O solution at 20^oC.

3. Pause the measurement after 3 spectra has been obtained. Repeat procedure 1 and 2 of subsection 3.3 at 30^oC, 40^oC, and 50^oC.

4. Dispose of the methyl acetate-D_2O solution, and export all of the IR spectra data for methyl acetate-D_2O solution (follow step 6 of subsection 3.2). Clean the ReactIR probe (follow step 5 of subsection 3.1). Turn off EasyMax and remove the water in the glass flask in EasyMax. Cover the tip of ReactIR with its cover.

4 Data Analysis

The data for these experiments will be analyzed using MATLAB. This handout was written using data for $T = 10^oC$ for tutorial purposes. The same protocol can be used to process data for other temperatures. Figure 3 shows a typical MATLAB window. The dashed box shows the current directory of MATLAB.

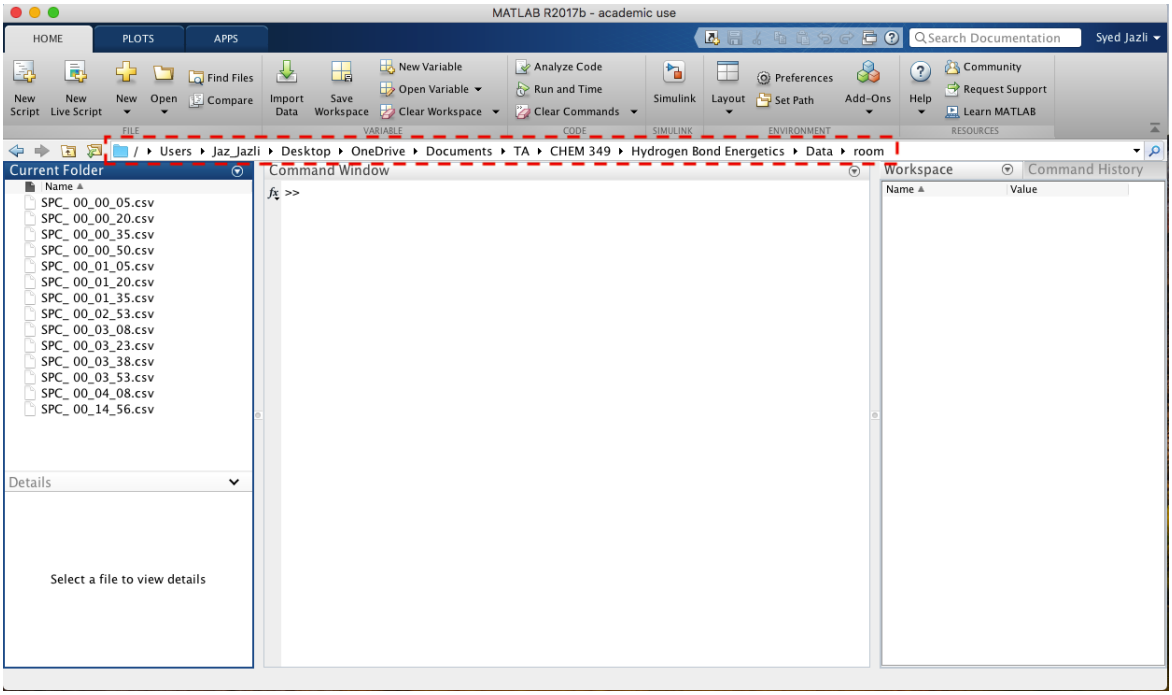

Figure 3: Typical MATLAB window. The dashed box in the figure shows the current directory of MATLAB.

4.1 Importing Data

In order to import experimental data into MATLAB, make sure the data are in the current directory of MATLAB. If the data are not in the current directory, copy the data to this directory. The data obtained from the IR instrument should be in a '.txt' format. In the left column of the MATLAB window, under the section called "Current Folder", you should see your experimental data file. Right-click on that file, and choose "Import Data..." option.

A new window similar to Figure 4 should pop-out. Change the "Output Type" using the drop down menu at the top of this new window (dashed box in Figure 4). Change the "Output Type" from 'Table' to 'Column Vectors'. In this case, there are 2 variables – wavenumber and absorbance in column A and B, respectively. Change the name of the variables to "WavNum" and "T10_1" by double-clicking on the variables' name. Next, select column A to highlight the entire column. Change the range from "A1:A940" to "A2:A940", and double click the "Import Selection" button to import your data into MATLAB. If you have done this successfully, you should see the variable 'WavNum' in the "Workspace" window. Repeat this procedure with column B. Then repeat the import procedure for all your data at 10^oC. You may skip importing the *Wavenumbers* since the *Wavenumbers* should be the same for all of your data. When importing the data for *Absorbance*, you have to give different name for each *Absorbance* data, for example, 'T10_1', 'T10_2', 'T10_3', etc.

Since the data you measured at 10^oC were obtained over time, you need to average the *Absorbance* data. To average the *Absorbance* data, type the following command into MATLAB's "Command Window"

$$T10Avg = (T10\_1 + T10\_2 + T10\_3 + T10\_4 + T10\_5)./5; \tag{6}$$

The above command tells MATLAB to add 'T10_1' to 'T10_5', then divide the sum by 5 and assign the values to 'T10Avg'.

4.2 Data Plotting

T10Avg is the data average for *Absorbance* at 10^oC. Plot the *Average Absorbance* as a function of *Wavenumbers* by typing the following command into the "Command Window"

$$figure;\ plot(WavNum, T10Avg) \tag{7}$$

To label the axes and put a title to the plot, type the following commands

$$xlabel(`Wavenumber\ (cm\hat{}\{-1\})'); \ ylabel(`Average\ Absorbance'); \tag{8}$$

$$title(`Plot\ of\ Average\ Absorbance\ vs.\ Wavenumber\ (cm\hat{}\{-1\})')) \tag{9}$$

If everything has been done correctly, you should get a figure similar to Figure 5. For this experiment, the region of interest is $\sim 1600\ cm^{-1}$ to $\sim 1800\ cm^{-1}$, which is highlighted with a dashed

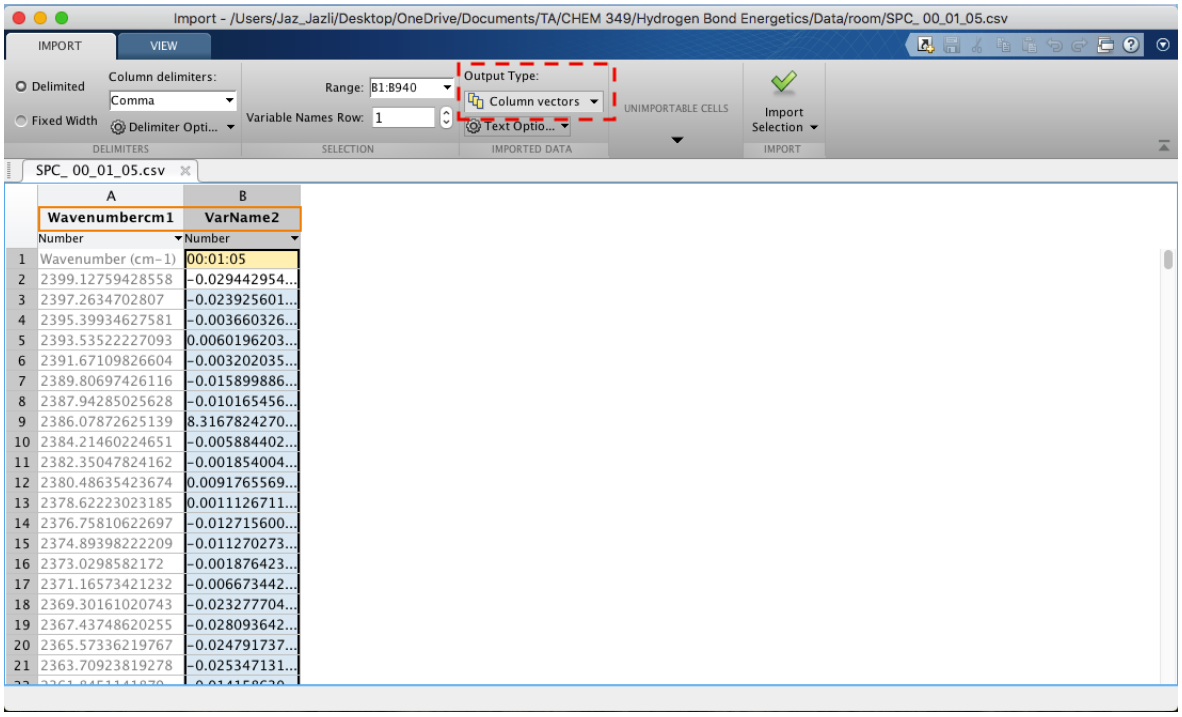

Figure 4: A picture of the "Import Data" window. The name of the variables are boxed with solid line while "Output Type" drop-down window is boxed with dashed line.

box in Figure 5.

4.3 Data Extraction

Find a variable called "WavNum" in the "Workspace" section and double click on it. A new window called "Variables" should pop-out (see Figure 6). Scroll down to find two values: $\sim 1800\ cm^{-1}$ and $\sim 1650\ cm^{-1}$. Record the index number corresponding to these two values. For this handout, the chosen values were $1800.7\ cm^{-1}$ and $1649.7\ cm^{-1}$ with the corresponding index numbers of 322 and 403 respectively. You may choose different values if you wish.

Once you have the two index numbers, you need to create two new variables containing the spectra of interest using the selected index numbers. Type the following command into the MATLAB's "Command Window"

$$WavNumS = WavNum(322:403); \tag{10}$$

$$T10S = T10SAvg(322:403); \tag{11}$$

If you have different index numbers than this handout uses, replaced the numbers in command (10) and (11) with your own index numbers. Command (10) loosely translates to copy all values from index number 322 to 403 of "WavNum" and paste it to variable "WavNumS". Next, make a new plot of *Absorbance* vs. *Wavenumbers* using command (7). You should get a figure similar to Figure

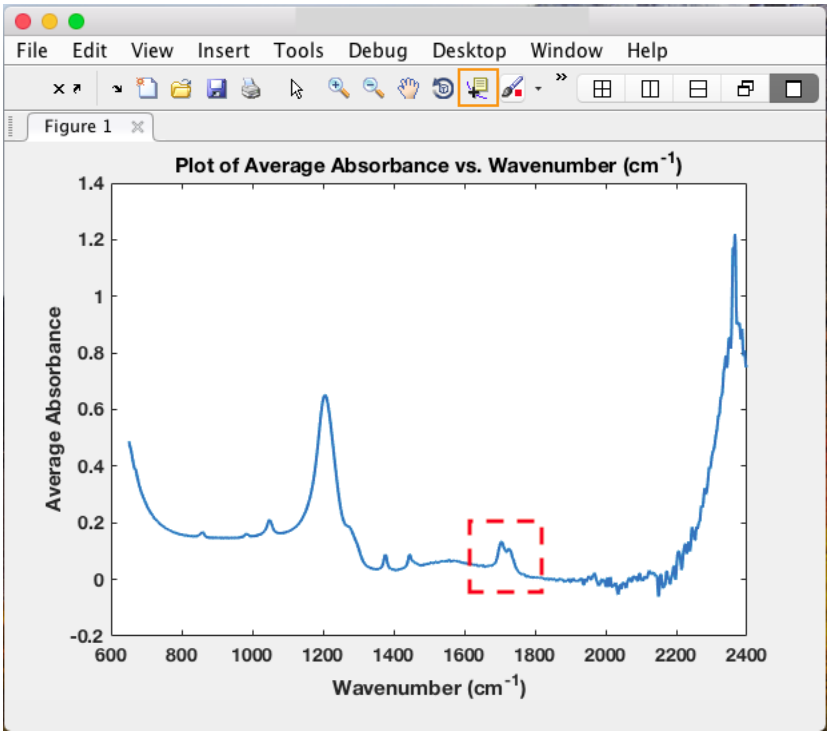

Figure 5: A plot of Average Absorbance vs. Wavenumber (cm^{-1}) for T = 10^oC. The region of interest are within the dashed box, which corresponds to wavenumbers from 1600 cm^{-1} to 1800 cm^{-1}.

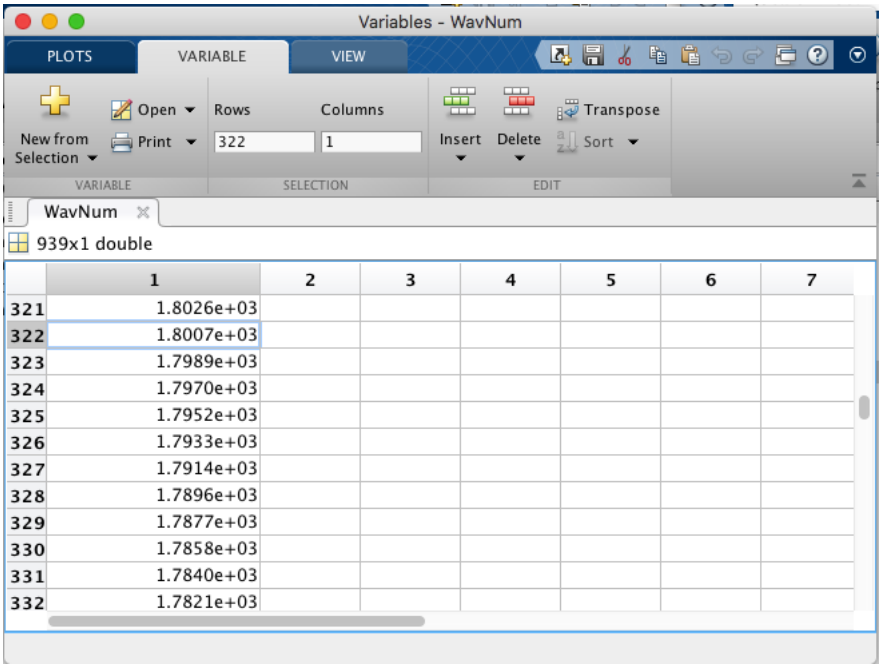

Figure 6: A picture of "Variables" window that shows the *Wavenumber* values. Column 1 has all of the *Wavenumbers* and the numbers to the left of column 1 are the index numbers for each row of column 1.

7. Notice that these peaks have a slanted baseline. Next, process the data so that these peaks have a flat baseline.

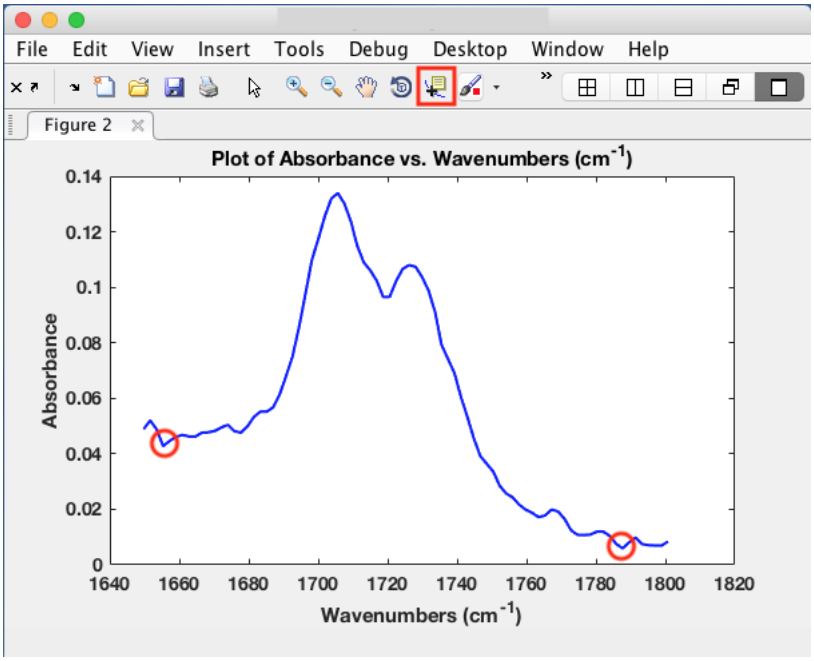

Figure 7: A plot of *Absorbance* vs. *Wavenumbers* using the extracted data. The button in the solid box at the top of the figure window can be used to retrieved the exact value of the points in the plot. The points in the circles on the plot are used to construct the straight line to make the baseline of this plot as flat as possible.

4.4 Baseline Correction

To make the baseline as flat as possible, we need to model the baseline by using a straight line fit. The straight line is constructed by choosing 2 points from the plot in Figure 7. The chosen points should be located near the ends of the plot — see Figure 7 for clarification. For this handout, the chosen points were (1655, 0.04235) and (1788,0.00627), and the resulting equation of straight line was $y = -\frac{0.03608}{133}x + 0.4913$. You have to calculate the equation of the straight line that describes the slanted baseline by yourself since MATLAB will not do this for you.

Next, using the straight line equation determined above, calculate the 'correction values' for your *Absorbance* and assign the 'correction value' to a new variable. You can do this by typing the following command into the "Command Window"

$$yFlat = -0.03608. * WavNumS./133 + 0.4913 \tag{12}$$

In this handout, the 'correction values' were assigned to a variable called *yFlat*. The command above is just the equation of a straight line that was determined earlier. Next, plot this straight

line together with your plot of *Absorbance* as a function of *Wavenumbers* by typing the following commands

$$figure;\ plot(WavNumS, T10S); \tag{13}$$

$$hold\ on;\ plot(WavNumS, yFlat); \tag{14}$$

It is important to include the command "hold on" in the above command. The command "hold on" tells MATLAB to plot a new graph onto an existing figure without removing the old graph. This feature can be turned off by typing *hold off*. To label the x-axis, y-axis, and the plot title, use the commands mentioned in the previous section (command (8) and (9)). If everything was done correctly, you should get a figure similar to Figure 8.

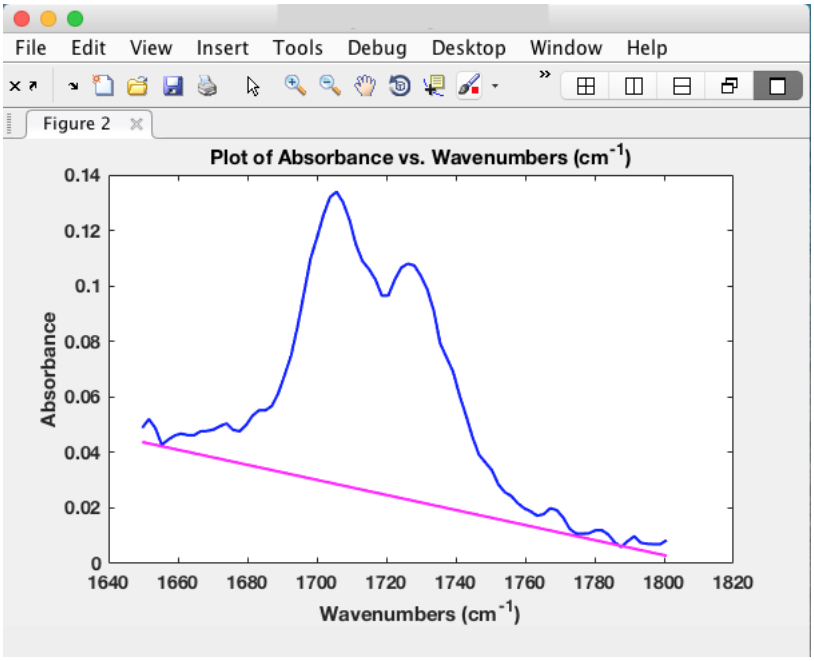

Figure 8: A plot of *Absorbance* vs. *Wavenumbers* with the correction values.

Next, you need to subtract the 'correction values' from the *Absorbance* values of the peaks by typing the following command

$$T10F\ =\ T10S\ -\ yFlat; \tag{15}$$

By plotting the 'corrected *Absorbance*' as a function of *Wavenumbers* you will get something like in Figure 9.

4.5 Curve Fitting

To fit two curves under the peaks, type the following command into the "Command Window"

$$ipf(WavNumS, T10F) \tag{16}$$

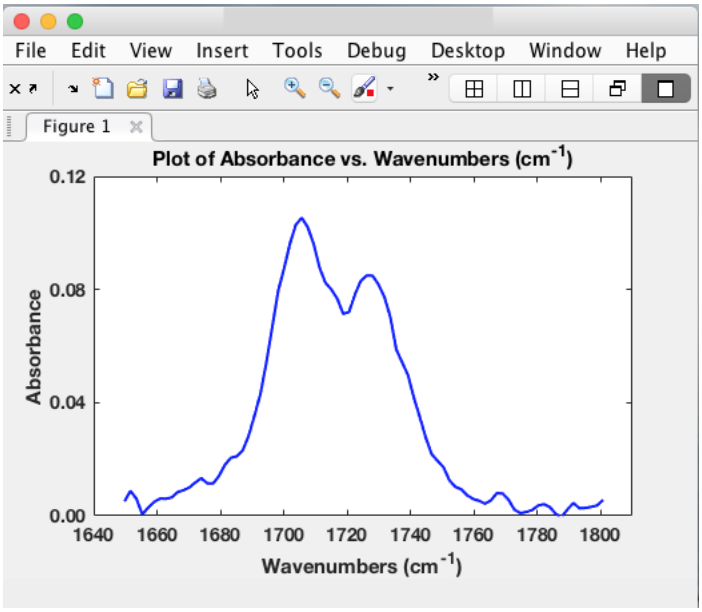

Figure 9: A plot of *Absorbance* as a function of *Wavenumbers* at T=10^oC with corrected baseline.

A new window similar to Figure 10 should pop-out. The default setting of this function is to fit a curve to one peak. To fit two curves under the two peaks, press '2' on your keyboard, and you will now have two 'pink dashed line' instead of one. Now, adjust the 'pink dashed line' so that each of the 'pink dashed line' coincide with each of the peak's maximum. You can adjust the position of the line by pressing the left and right arrow key on the keyboard. To zoom in or zoom out, you can press on up or down arrow key of your keyboard.

Once you are satisfied with the position of the 'pink dashed line', press the 'f' key of your keyboard to fit two curves under the peaks. Figure 11 shows an example of the end product after two curves have been successfully fitted under the peaks. The area under each curve is written at the bottom plot of Figure 11. You need this curve-fitted plot for your lab report. To save this plot, click the save button at the top of the "Figures" window. Now, repeat the entire process with all of the data at each temperature, i.e, 18^oC, 30^oC, 40^oC, and 50^oC.

4.6 Calculations

To determine the value $\Delta_r H^o$ and $\Delta_r S^o$, you need to plot $ln\frac{A_{1HB}}{A_{2HB}}$ as a function of $\frac{1}{T}$. To calculate $ln\frac{A_{1HB}}{A_{2HB}}$, create two new variables; one represents the area under the curve for 1 hydrogen bond and the other one represents the area under the curve for 2 hydrogen bonds. To do that, type the following command

$$A1HB = [2.013,\ 0.6061,\ 2.490,\ 2.285,\ 2.212]; \tag{17}$$

$$A2HB = [2.190,\ 0.4887,\ 2.502,\ 2.193,\ 1.957]; \tag{18}$$

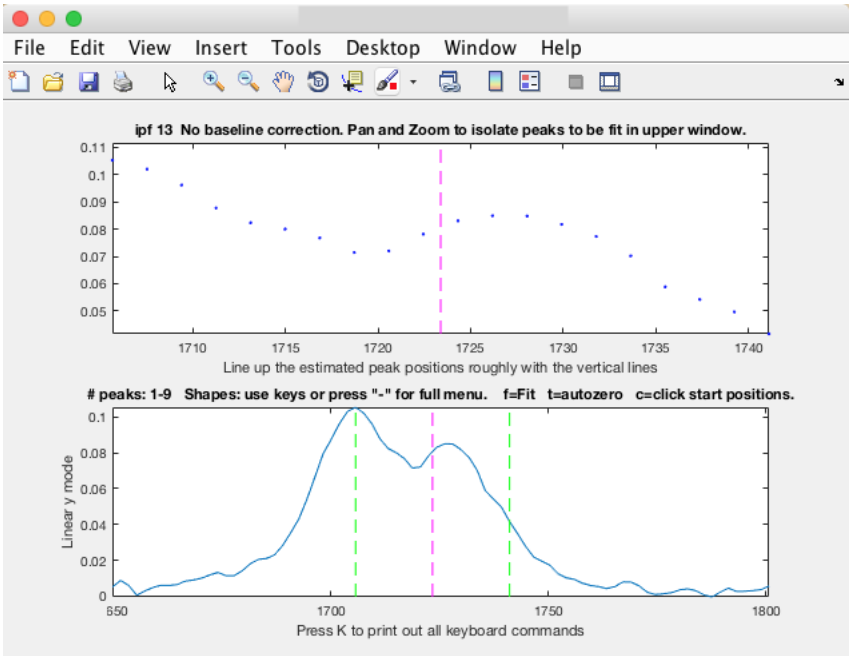

Figure 10: Interactive peak fitting (IPF) window. This figure shows the default setting of the IPF, which is used to fit a curve on a single peak. To fit 2 curves under 2 peaks, press '2' and adjust the position of the pink-dashed line so that the lines correspond to the maximum of respective peak.

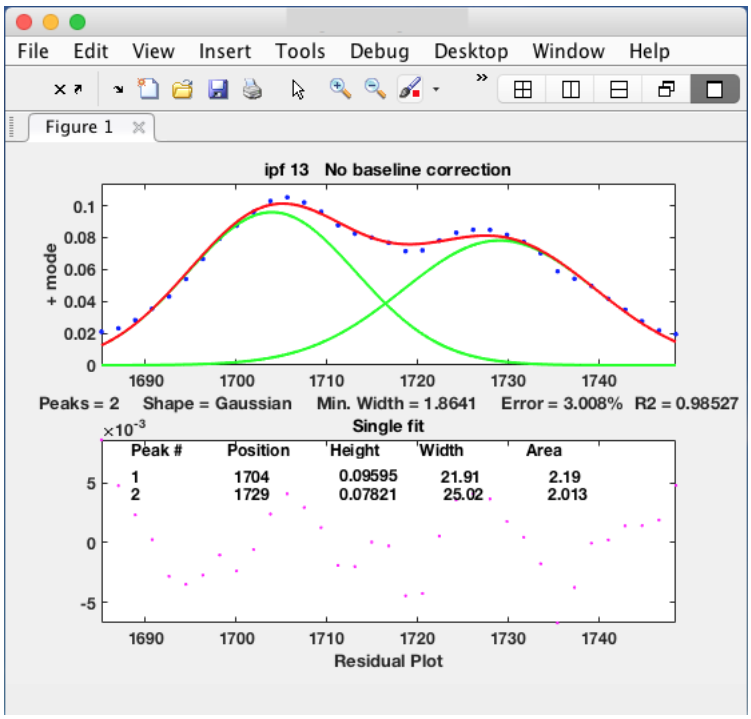

Figure 11: A plot of *Absorbance* as a function of *Wavenumbers* at T=10^oC and the associated curves under the peaks. The area under the curves is listed on the bottom plot of this figure.

The numbers in command (17) and (18) are the area under the respective curve for 1 and 2 hydrogens bond at different temperatures. You should type in your own area under the curves for command (17) and (18). Pay attention to the order of areas that you type in. For this handout, the area under the curves was type in ascending order of temperatures, i.e., area under the curves at temperature 10^oC, 18^oC, 30^oC, 40^oC, and 50^oC. Next, you will divide A1HB by A2HB and take the natural log of the number by typing the following command

$$lnK = log(A1HB./A2HB); \tag{19}$$

Next create a variable for $\frac{1}{T}$ by typing the following command

$$Temp = 1./[283.15,\ 291.15,\ 303.15,\ 313.15,\ 323.15]; \tag{20}$$

Make sure that the temperature is in Kelvin. To plot the graph, use the "scatter" function instead of the "plot" function. Type the following command

$$figure;\ scatter(Temp, lnK); \tag{21}$$

To fit a linear regression line, click on "Tools" and select "Basic Fitting" option. A new window should pop-out, and from this new window select "linear" and "Show equations" to add the linear regression line to your plot. To calculate the 'r^2' value, type the following command

$$mdl = \ fitlm(Temp, lnK) \tag{22}$$

Now that you have the equation of straight line from the linear regression line, determine the value of ΔH^o and ΔS^o based on the equation in the introduction.

5 Data Reporting

For your lab report make sure you have :

- Spectra of methyl acetate in THF and D_2O. Comment on the effect of solvent on IR spectra.
- Averaged spectra of methyl acetate in D_2O for all temperatures. Comment on the effect of temperature on IR spectra.
- Plot of $ln\left(\frac{A_{1HB}}{A_{2HB}}\right)$ vs. $\frac{1}{T}$ and determine the values of $\Delta_r H^o$ and $\Delta_r S^o$.

References

1. A Pragmatic Introduction to Signal Processing. https://terpconnect.umd.edu/%7Etoh/spectrum/ (accessed December 9, 2017).

2. Guerin, A. C.; Riley, K.; Rupnik, K.; Kuroda, D.G. Determining the Energetics of the Hydrogen Bond through FTIR: A Hands-On Physical Chemistry Lab Experiment. *J. Chem. Educ.* **2016**, 93, 1124-1129.