Appendix II:

Guide To NMR Data File Transfer and *Magnetic Resonance Companion* (MestRe-C)

(Throughout this guide, use the left mouse button unless stated otherwise).

Transfer of NMR data from the Gustavus NMR spectrometer computer to PC's in the computer lab on the first floor of Nobel Hall.

1. Choose the **LeechFTP** icon under the Programs and Internet directories.

2. In the FTP program, choose "Connect" from the "File" pull-down menu. For host, enter **bloch.chem.gac.edu**, for username, enter **organic** for password, enter **0rganic** (zero, not "oh" as first character. Choose okay. You will be connected to the NMR computer, from which you will transfer your data. It might take a minute for the connection to be made.

3. Note the format of the window which appears: the middle window shows the local location where your files will be transferred, the left half of the window (Local System) is where the transferred files will show up and the right window (Remote System) is the NMR computer set to the directory containing your data (e.g. /home/woodward/ochemII). Your NMR data is in one of the **folders** in the right window.

   Make sure that you are in the Temp folder on drive C in the Local window. You need to go up to the highest level on drive C and then choose the Temp folder by double-clicking.

4. In the right, Remote System, window, double click on the folder corresponding to your lab section (i.e. monpm for Monday afternoon), followed by **unknowns.**

5. Clicking on your folder should have caused a list of files to appear in the right window. Select the folder corresponding to your unknown (unk.1.fid for unknown 1).

   Note that files with these same names may also appear in the left window (usually in the aaanmr subdirectory). These are the files of the last person who transferred data, since everyone is transferring files with the same four names. It is extremely important to ensure that these files are overwritten and replaced by your files. You can check this by reading the comments in the scroll screen after downloading your files. Also, it is important to do this data transfer before going into MestRe-C to work up the NMR data, or you will be processing someone else's data!

6. After selecting your files, transfer your data to the PC by clicking the download button (to the right of the magnifying glass) on the toolbar. You want to click on the button that shows an arrow pointing down toward a hard drive icon. Your files are now copied to the ***C: /Temp/aaanmr*** folder. **If files with the same names were already on the Local System folder, make sure they have been overwritten with your data.**

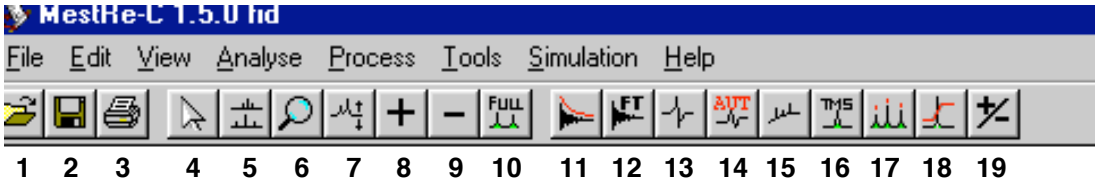
7. Close the connection to the NMR computer by quitting the FTP program. Just click the "**X**" box in the upper right corner of the window, as with any Windows program.

Starting MestRe-C

1. Choose the **Mestrec1.exe** icon under **Programs**.
2. Since the data you transferred is not MestRe-C data, you will need to *Import* your data to the MestRe-C program: **FILE/IMPORT** (i.e. select **IMPORT** under the **FILE** menu).
3. A dialog box will appear asking you what type of data you want to import. Select the *Varian VRX/Unity* file type; click **OK**.
4. A dialog box appears that you will use to select a file to be opened. Go to the ***C:/Temr*** folder where you saved your data. Inside the folder you downloaded are four files: *fid, log, procpar, text*. Double click on the *fid* file (or highlight it and click the Open button). Your FID will now appear in the window.

Processing Your FID

Most of the commands you need to process routine data can be accessed through the buttons on the toolbar that runs across the top of the spectrum/FID window in MestRe-C. The toolbar is shown below, with numbers assigned to each button for easy reference. When a button is referred to in this document, its number is always given in bold in parentheses.



Step-by-Step Data Processing

1. First you must perform the Fourier Transformation (FT) calculation to convert the FID into the spectrum. Click the **FT** button (**12**) and click OK in the dialog box that appears.
2. Your spectrum is likely to appear with very small peaks. Click on the **+** and **-** buttons (**8** and **9**) to increase and decrease the vertical scale.
3. You will need to *phase* the spectrum if the baseline around the peaks is not flat. You can let the program autophase your spectrum with the **Autophase** button (**14**). It is likely that you will still need to do a little manual phasing to get a good baseline. To do so, select the **phase correction** button (**13**). A dialog box appears with slide bars to adjust the zero-order and first-order phase corrections. A red cursor line (pivot) also appears on the tallest peak in your spectrum. If it is not already located at one end of your spectrum, move this pivot to any tall peak near one side of the spectrum or the other. Use the **Displace Pivot Point** slide bar to make this movement. Use the **order-0** phase correction bar to flatten the baseline around the peak with the pivot. Don't worry about the phasing of the other peaks at this point. You may find it useful to greatly increase the vertical scale of the peaks (**+** and **-** buttons; **8** and **9**) to get a good look at the baseline. When you are satisfied with the phasing of the peak with the pivot on it, use the **order-1**

phase correction to adjust the phasing of other peaks of your spectrum (do NOT move the pivot). Usually the peaks farthest away from the peak with the pivot will be most in need of the first-order phase correction. When you are satisfied, click the Apply button to apply the phasing corrections you made to the spectrum. If the spectrum looks good, exit the phasing routine by clicking the OK button. If not, continue the phasing process until you are satisfied.

4. This program prints exactly what you see on the screen, so you will probably want to move the spectrum down in the window, so that it sits about an inch above the ppm scale on the bottom of the window. To do this, click the **Vertical Shift** button (**7**). In this mode, the baseline of the spectrum moves to the point where you place the cursor and click the left mouse button. You can also hold down the left mouse button and move the black line that appears to where you would like the baseline of the spectrum to be located. At this point release the mouse button. Leave room for the integral values to be printed below the baseline. To fully exit this vertical shift mode, click the **Default Mode** button (**4**). The program stays in the vertical shift mode until you explicitly exit this mode (button **4**). **It is not a bad idea to click this button (4) after each major manipulation, just so that you exit fully the previous mode in which you were operating.**

5. To expand around a region of interest in your spectrum use the Zoom button (**6**). In this mode, you can use the left mouse button to drag a box around the region of interest. Releasing the mouse button causes the expansion to occur. Exit the Zoom mode by clicking the Default Mode button (**4**).

6. To set the ppm scale, expand (step 5) around the TMS peak (or any other reference peak). Select the Spectrum Reference button (16 - labeled with TMS). In this mode, the arrow cursor becomes a cross hatch. Place the cross hatch on the reference peak and click left mouse button. A dialog box appears. Enter the desired ppm for this peak in the "New Reference" entry (0 ppm for TMS, 7.24ppm if you are using the residual chloroform peak) and click **OK**.

7. To get back to the full spectrum click the Full button (**10**).

8. Now expand around the peaks you wish to plot.

9. Before putting the integral lines on the spectrum you will want to correct the integral baseline. If you don't do this you get poorly phased integrals with poor baselines (and bad integral values!). So, to correct this before you display the integral, go to the **Tools** menu. In the **Integral Options** entry, select **Baseline Correction**. Your integral lines should appear nicely phased when you display them in the next step.

10. To enter the integration mode click the Integrate button (**18**).

11. To integrate individual peaks, drag a box (left mouse button) around each peak, one at a time. After you release the mouse button after making a box around a peak, the integral line will appear and an integer value for this resonance appears under the peak. Do this for each peak of interest. Note that the program sets an integral value of 1 to the first peak you integrate; the integral value of all other peaks are set relative to this first peak. So, if you suspect that one of your peaks is due to a single proton, integrate it first. You can always start over by selecting the **Delete Integrals** entry under the **Tools** menu.

11. To set the vertical offset of the integral segments (i.e. where you want the integral baseline to be), use the scrollbar on the right side of the spectrum window.

12. You can control the vertical scale of the integral lines with the **arrow up** and **arrow down** keys on your keyboard. Set the largest integral line to a size that fills most of the page.

13. When the integrals are displayed as you would like them to be, exit the Integrate mode by clicking the Default Mode button (**4**).

14. To add a title, go to the **Edit** menu and select **Inset Comments**. In the dialog box that appears, enter your title or any other comments you wish to make. Click **OK** and your comments/title will appear on the left side of the spectrum.

15. To Print your spectrum, select the print button (**3**). **Before** clicking **OK**, you must set the paper orientation to **Landscape**. To do this, click on the **Properties** button in the upper right corner of the Print dialog box. Click on the orientation box and select the **Landscape** orientation and click **OK**. Now, click **OK** in the Print dialog box to print your spectrum.

18. When you are done, quit the application. You will be asked if you want to save the spectrum; usually just click the **NO** button. If you want to save your work, save it on a floppy...but there is no need to do this; the data remain on the NMR computer and can always be re-transferred to the PC's.