VNMRJ on Mercury400 NMR Operating Instructions
Update (February, 2007)

Login
Login to the system by entering your user name and password.

## *Starting vnmrj*

1. Start vnmrj by clicking the vnmrj icon Once at the top of the screen. (*If you click twice, then there will be two vnmrj open, you could close one, otherwise, some operation will not work*).
2. Choose to run a ^{1}H experiment by dragging the proton icon on the left of the screen to the black area.

Inserting your sample
1. Click Lock (on the left menu column); Click Lock off; Click Spin off and then Click *eject* to remove the standard from the magnet.
2. Replace the standard with your sample and click on *insert*.
3. If you wish to use a solvent other than CDCl$_3$, Click the Study tab, then select which solvent you would like to use with the drag-down box on this page.
4. If you would like to print a sample name/ID on your spectrum, insert this text into the comment box here (will print later by the command **pap**).
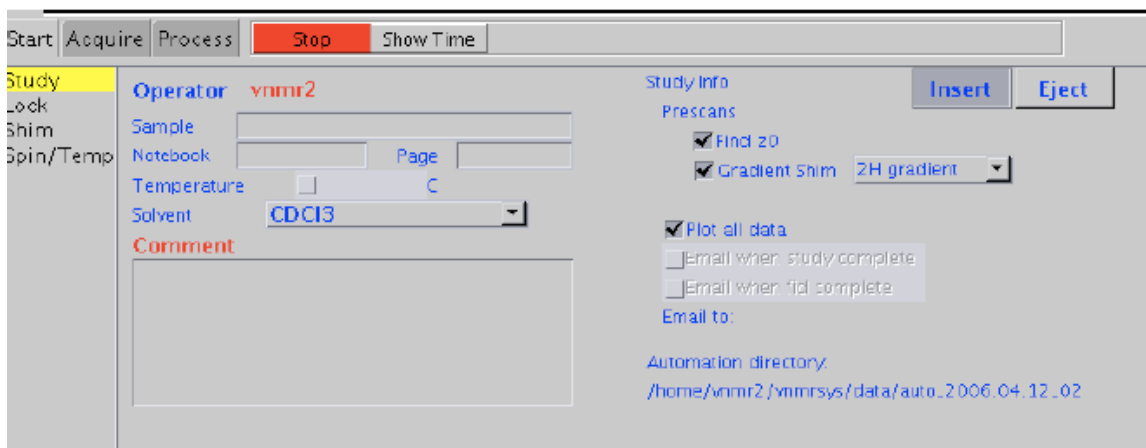
Locking and Shimming your sample

1. If you are using a solvent other than $CDCl_3$, click on the lock tab and change Z0, Power, Gain, and Phase as necessary to lock your sample. The values can be adjusted with the left and right mouse buttons or by typing in the desired value and pressing enter.
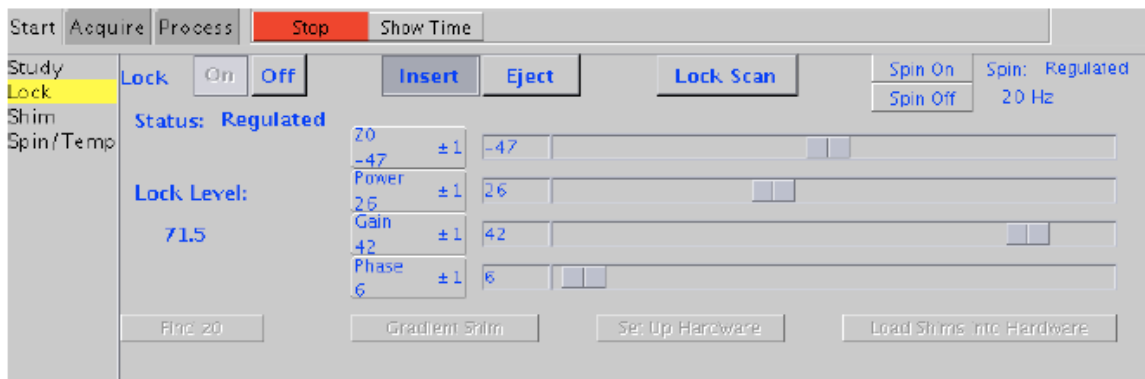
Note: You MUST click lock off before you could adjust the Z0 value. The Z0 value will be changing because of magnet drifting, Liquid Nitrogen refilling and Liquid Helium refilling. I will post a list of Z0 values for different solvents for your convenience. But you should know how to change Z0 to lock on your sample properly.

2. Shim your sample by first clicking on the shim tab, then by adjusting the Z1 and Z2 values, etc. If the shims are far away, you may type "loadshim" to load the standard shims. Make sure the lock power, lock gain and lock phase are correct, these parameters for the same solvent should not be changed too much.

Note: This instrument is capable of automatically finding Z0 for your solvent and setting up for automatically shimming on your sample. Whenever the instrument is stabilized, I will setup for this option.

Start | Acquire | Process | Stop | Show Time

Study
Lock
Shim
Spin/Temp

Lock Scan
Fid Scan

z0 -47 ±1
Lk Power 26 ±1
Lk Gain 42 ±1
Lk Phase 6 ±1

71.4

Lock   71.4

Z1 732 ±1
Z2 -203 ±1
Z1C 308 ±1   Z3 22 ±1
Z2C 54 ±1   Z4 84 ±1
               Z5 -225 ±1

Spin On | Spin Regulated | Recr gain
Spin Off | 20 Hz | 20

Acquiring your ^{1}H spectrum

1. Check the current acquisition parameters by clicking on the process tag, and click Text tab. then by typing **dg** in the input box above the black box. All the acquisition parameters will be displayed.
2. To change any parameters, click the acquire tag and make Adjustments as necessary by typing in the input box. Use the format **"parameter = value"** to make changes.
3. Alternatively, you may use the default parameters and simply change the number of scans by typing in the command input box (i.e., type **nt = 16** for 16 scans).
4. Start the experiment by typing **ga**.

Processing your spectrum

1. After acquisition is complete, the spectrum should be displayed on the screen. If you need to save the data, type **svf('my_data')** to save the data. (See more about saving data)

exp1: Experiment started
exp1: Acquisition complete
wft vsadj aph

Exp:1   Seq: Proton   Index: 1
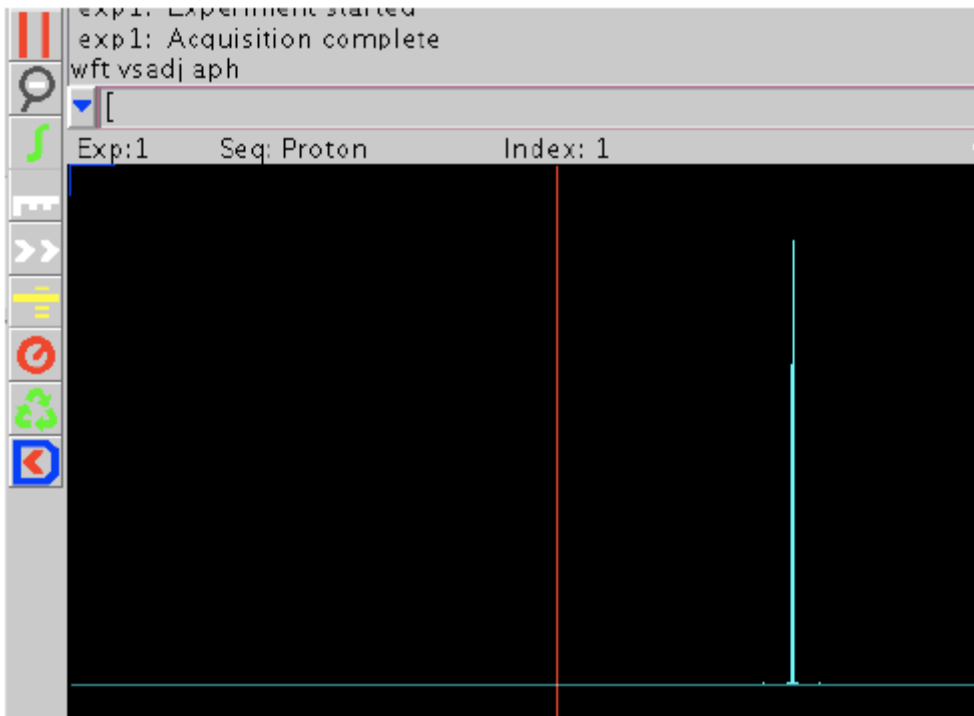
2. Type **wft vsadj aph** in the input box for Fourier Transform (FT), vertical scaling of peaks, and autophasing, respectively).

3. *Displaying the scale*: Note the set of processing icons to the left of the spectrum. Display the scale by clicking on the white ruler icon.

4. *Expanding the spectrum*: To expand the spectrum, select the top icon containing 2 red lines. Place the cursors where desired, then expand the region by choosing the zoom in/out (magnifying glass) icon. Use this icon to expand the spectrum and also to return to normal size.

5. *Setting the solvent reference peak*: Place the red cursor on your solvent peak, then type **rl(x.xxp)** in the input box to place the cursor directly on the peak and set the reference value (i.e., **rl(7.27p)** for CDCl$_3$).

6. *Integration*: Click on the green integral icon, then take note of the two additional integration icons that appear just below it. Click the middle integration icon and use the mouse to cut the line, then return to the spectrum by typing **ds** in the input box. If necessary, you can also click on the top integral button or type **cz** to display the full integral.

7. *Setiing a peak integral reference value*: To set a specific peak integration value, click on the process tab, then the cursors/integration subtab. Place the cursor on the desired peak and

type in the desired number of protons in the *normalization value* box. Save changes by clicking on the *set integral value* icon.

8. **Set a precise spectrum width**: To set a specific numerical range for your spectrum, use the same commands as used previously on other instruments (i.e., typing **cr = 9.5p delta = 10p** then clicking the expand icon will set the window from -0.5 to 9.5 ppm).

9. **Setting the peak threshold**: To choose the level of peaks for peak picking, press the icon labeled by the yellow threshold line and adjust the line accordingly.

Plotting and printing the spectrum
To plot and print, type any combination of the following commands into the input box.
- pl : print spectrum
- pir: print integrals
- pscale: print scale
- pltext: print text label (from the comment box)
- pap: print acquisition parameters
- pll: print peak locations (ppm and Hertz) and peak height
- ppf: print peak frequencies above each peak
- page: eject page from the printer

Saving the data
1. If you do not yet have a directory in your home folder, make a new directory by typing the command **mkdir('directory_name')**. Alternately, you can open your home folder (top left of the desktop) and create a new directory by selecting *File→ Create folder*.
2. To save a file in an existing directory, first choose the directory by typing **cd('your_directory')**. Next, save the file by typing **svf('file_name')**. To check the current directory by typing **pwd**
3. To retrieve your data later, find your file in your home folder on the desktop and drag it to the black processing window. Process as described previously.

Finishing the experiment
1. Click spin off, click lock off and click Eject to eject your sample and replace with the standard sample.
2. If you used a solvent other than $CDCl_3$, adjust the lock parameters back to the required ones listed under the monitor.

3. Click on the *loadshim* icon at the top of the page to load the standard shim file.
4. Manually shim the sample to reach the required lock level.
5. Exit the system by choosing *File*→*Exit vnmrj* or by typing **exit** into the input box.
6. Logout by choosing *Actions*→ *logout*, then click *OK*.

If you could remember the following commands, it will help you to work around the instrument and data more efficiently.

dg svf('**') cd   nt ga nl rl pl pscale pir pap page pwd loadshim cd('***') text('***') dtext cz ppf   cr= ##p delta=##p   ds full   cd   time gain? Or gain=10 wft dscale   aph   vsadj**

VNMRJ ^{13}C Operating Instructions

Follow the VNMRJ ^{1}H instructions to login to vnmrj and insert, shim, and lock your sample.

Acquiring your ^{13}C spectrum

1. Choose to run a ^{13}C experiment by dragging the carbon icon on the left of the screen to the black area.
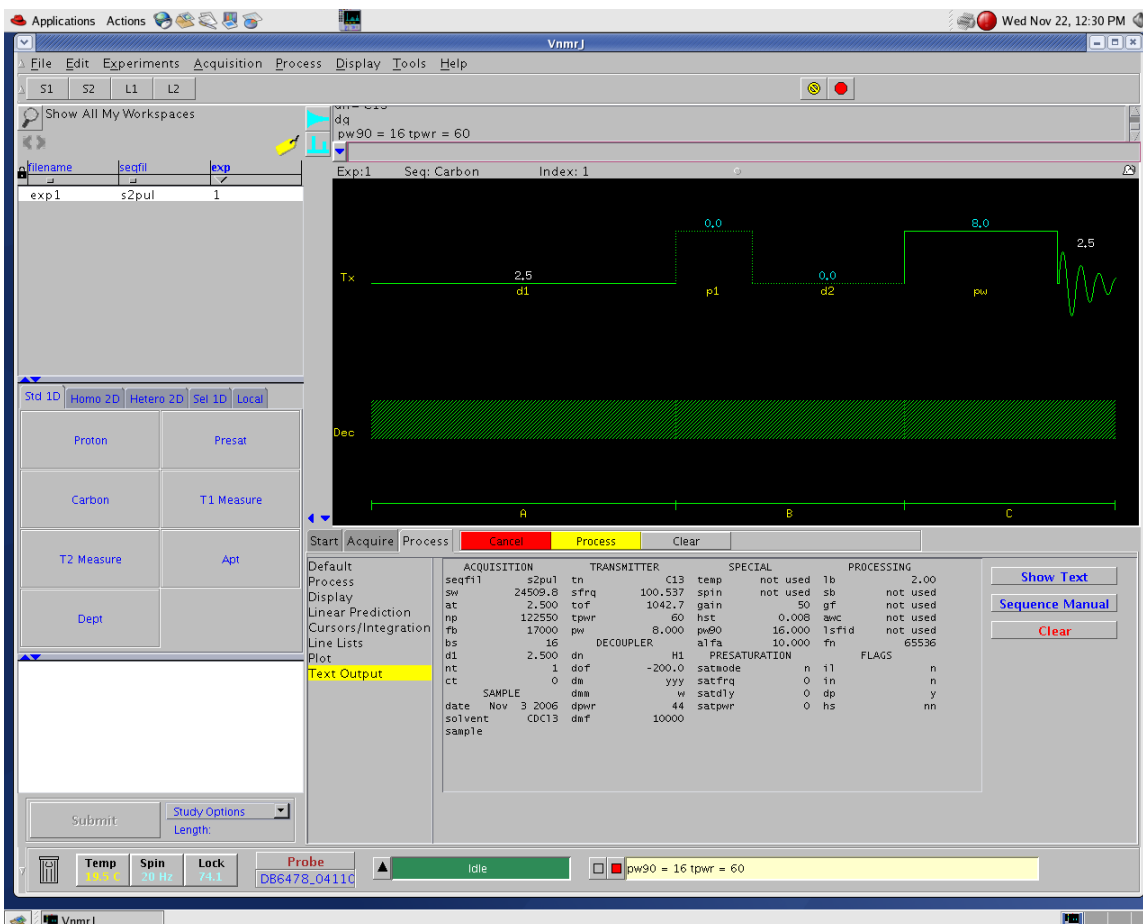
Note: This experiment required two channels: one is observe for C13 and the other is for decoupling for H1. The decoupling is controlled by dof (decoupling center frequency) dpwr (decoupling power), dm (for C13, it is set to 'yyy') dmm (it is set to 'w'), and dmf (it has been calibrated, do not change it)

2. Check the current acquisition parameters by clicking on the process tag, then by typing **dg** in the input box above the black box.

3. To change any parameters, click the <u>acquire</u> tag and make adjustments as necessary by typing in the input box. Use the format **"parameter = value"** to make changes.
4. Alternatively, you may use the default parameters and simply change the number of scans by typing in the command input box (i.e., type **nt = 16** for 16 scans). Tip: You could use big nt number, whenever it reaches enough signal, you could stop it by type aa or click red dot icon.
5. After typing in the number of scans, type **time** to find out how long the experiment will take. If you run an excess number of scans and would like to stop early, type **aa** to abort the experiment.
6. Start the experiment by typing **ga**.

Processing your spectrum

1. Apply the same commands as used on the other instruments to process the spectrum (i.e., type **wft aph** in the input box for Fourier transform and autophasing, respectively).
2. *Displaying the scale*: As in the case for a ^{1}H experiment, note the set of processing icons to the left of the spectrum. Display the scale by clicking on the white ruler icon.
3. *Expanding the spectrum*: To expand the spectrum, select the top icon containing 2 red lines. Place the cursors where desired, then expand the region by choosing the zoom in/out (magnifying glass) icon. Use this icon to expand the spectrum and also to return to normal size.
4. *Setting the solvent reference peak*: As in the case with the other instruments, place the red cursor on your solvent peak, then type **rl(xx.xxp)** in the input box to place the cursor directly on the peak and set the reference value (i.e., **rl(77.23p)** for $CDCl_3$).
5. *Manual phase the spectrum*: Clcik the manual phase icon on the left of the graphic window, select right side of the spectrum and phase the spectrum, and then select left side of the spectrum, and phase the spectrum. Repeat until all peaks in the phase, click the recycle icon to exit manual phase routine.

Follow the VNMRJ ^{1}H instructions to plot and print the spectrum, save the data, and replace your sample with the standard sample. **Before exiting the system, make sure to drag the proton icon to the black window to turn of the decoupler.**

I will work on the instructions for APT, DEPT, Solvent suppression, COSY, HETCOR and 1D NOE experiments. Any of you need special experiments, please let me know, I could arrange the setup and training. If you have any questions about the hardware, software, please feel free to contact me at 404-727-6621 or email <u>swu@emory.edu</u>. If there is a emergency, you could call me at 678-481-8331 (cell), 770-734-0147 (home).
Do not try to fix the instrument yourself (rebooting, probe tuning, adjust air pressure, these will cause other problems you may do not know, it will make me more difficulty to find the original problem). If you feel the instrument is not functional, call me first or leave me a message and then leave a note on the console. I will come to check as soon as I could.

VNMRJ COSY Operating Instructions

Preparing for the COSY experiment

1. Use the VNMR ^{1}H instructions to shim and lock your sample. Change solvent settings as necessary. Drag the proton icon to the black window and acquire the proton spectrum using standard parameters.

2. Process the ^{1}H spectrum as listed in the instructions. Set the solvent reference peak, and save the file for later reference.

3. Type **gain?** in the command input box to determine the value to use for gain in the COSY experiment.

4. Use the ^{1}H spectrum to determine the spectrum width of the COSY. Do this by placing the red cursor on both sides of the spectrum and typing **movesw**.

Acquiring the COSY spectrum

1. Click on the Homo 2D tab on the left of the screen.

2. Drag the COSY icon to the black window.

3. Type **dg** to view the current parameters. The parameters will be displayed after choosing the process tab (horizontal menu) and text output tab (vertical menu).

4. Change the gain value to the number used for the ^{1}H experiment in the previous section. This can be done by typing **gain = x** in the command input box.

5. Change the number of scans by typing **nt = x** (where x = 16, 32, etc.).

Note that the minimum is 4 scans, and a larger number of scans will lead to a better spectrum.

6. Change the number of FID's by typing **ni = x**. Typically the desired values for x are 128, 256, 512, etc. It is best to start with at least ni = 256, and a larger value is better.

7. Check the parameters again as explained in step 3. Type **time** to see the length of the experiment. Adjust nt and ni to fit your allotted time.

8. Turn off the spin (under the start and lock tabs), and type **ga** to begin the experiment. Make sure fn = fn1 (1024, or 2048) and sw = sw1.

Processing the spectrum

1. If you would like to use the default processing/plotting method, you

can print now by typing **pcon page** in the command input box. If you would like to manually process the spectrum, continue below.

2. *Peak shape adjustment (F1 and F2):*

- After selecting the process tab on the horizontal menu, choose the weighting tab on the vertical menu. To adjust F2, first click on *FT 1D - 1st increment*. If the full spectrum is not shown, type **full** followed by **f** to show the whole spectrum.
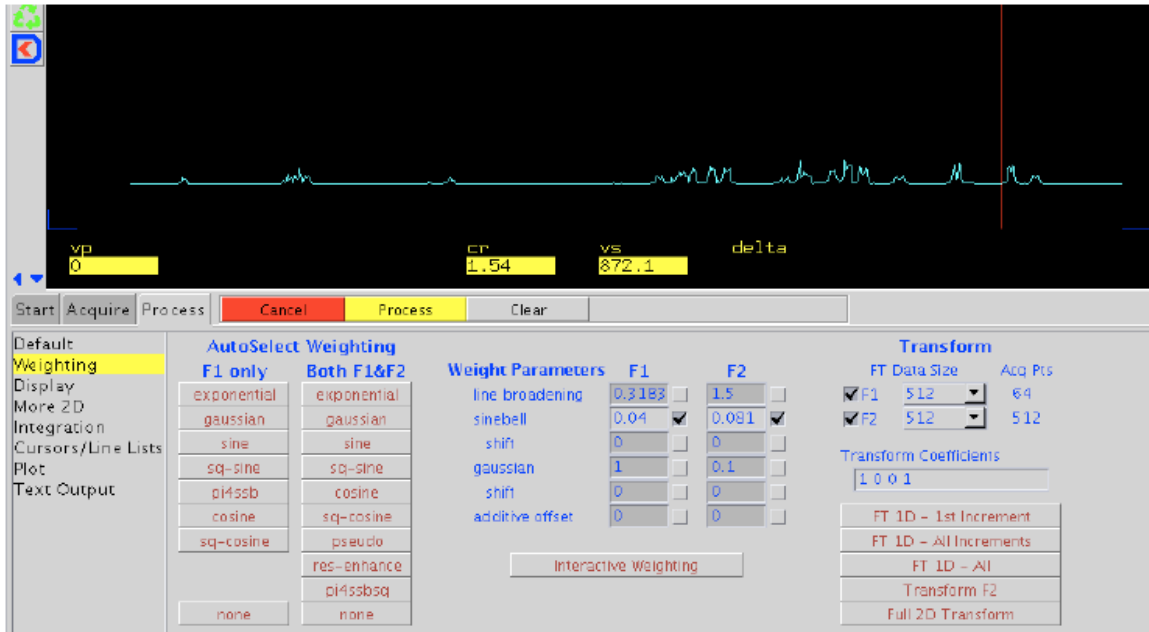
- There are two options for adjusting the values for F2. You can simply input new values for F2 by entering numbers in the boxes under the weight parameters heading and F2 subheading, and then clicking the *Transform F2* icon.

- Alternately, you can manually adjust F2 by clicking on *interactive weighting* and adjusting the Gaussian curve with the left mouse button until the desired peak shape is obtained. When adjustment is finished, click the *transform F2* icon.
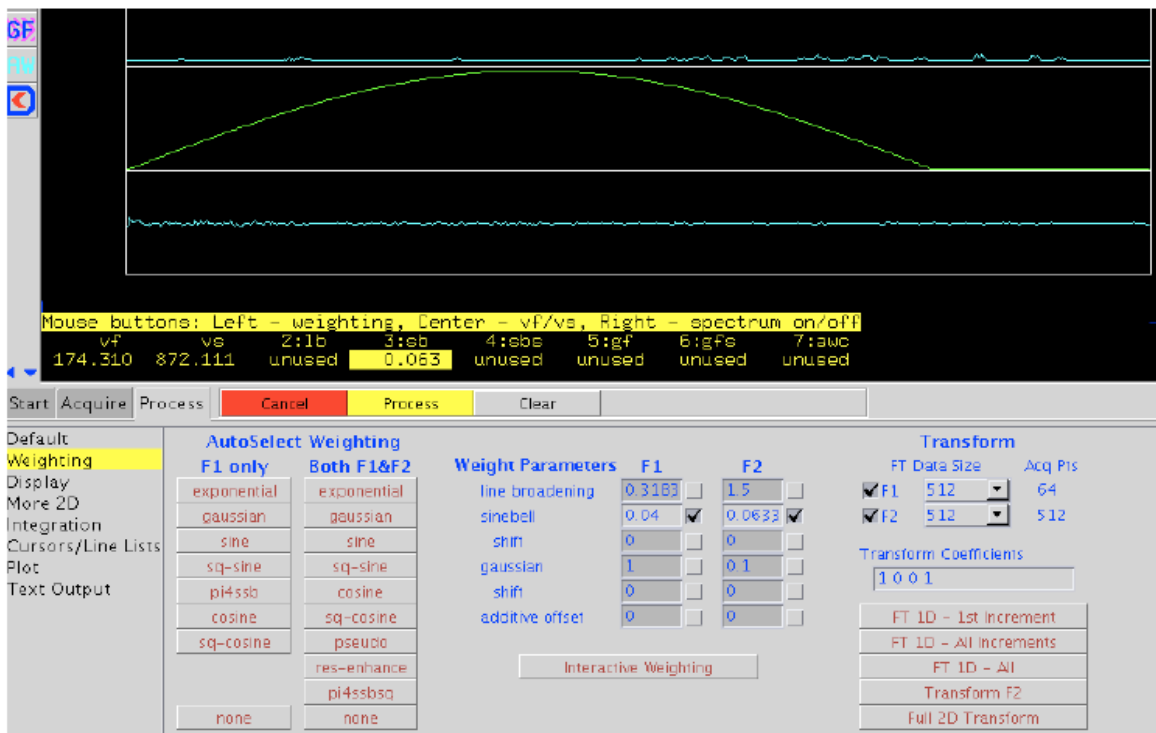
- To adjust F1, choose any single line of the spectrum using the red cursor lines, then click on *interactive weighting* and manually adjust the curve as described in the last step. When adjustment is finished, click the *full 2D transform* icon.

3. **Eliminating peak noise:** To remove excess unsymmetrical noise, type

foldt in the command input box.

4. **Setting the solvent reference peak:** Note that this step is unnecessary if you have already set the reference peak in your $_1$H NMR experiment at the beginning of these instructions. However, if necessary, you may set this value by using the cursor and zoom (magnifying glass) icons to expand the area containing the solvent peak. Place the cursor on the desired peak and type in the command **rl(x.xxp)** to set the reference value.

5. **Decreasing static noise:** Click on the display tab on the vertical menu.
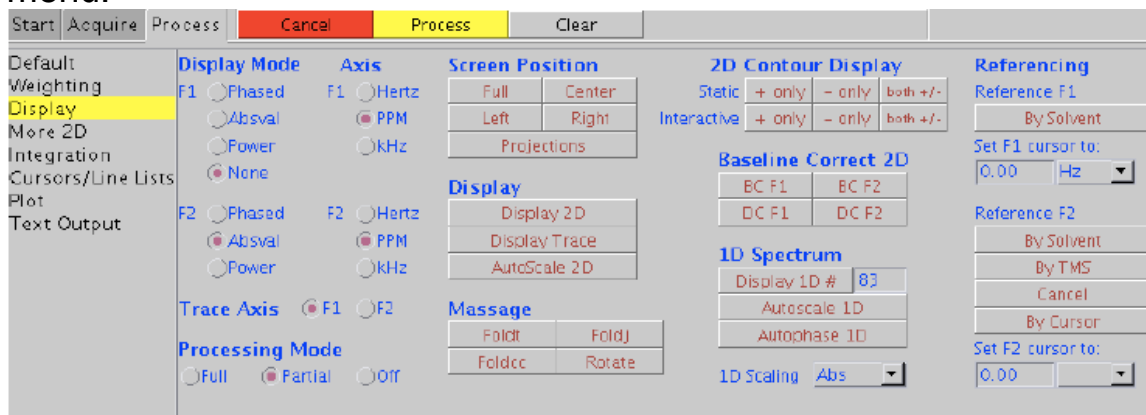
Check to make sure that "none" is selected under Display Mode, then look for the 2D Contour Display heading. Select the static *both +/-* icon to decrease static noise.

6. **Changing the screen position of the spectrum:** look for the Screen Position heading. You may choose from the *full*, *center, left*, and *right* icons, depending on how you would like to plot your spectrum. Typically it is best to choose the *center* option since it will allow for easier plotting of the peaks and spectrum later.

7. **Increasing/decreasing intensity of peaks:** Adjust intensity of peaks by using the yellow bulb icons on the toolbar.
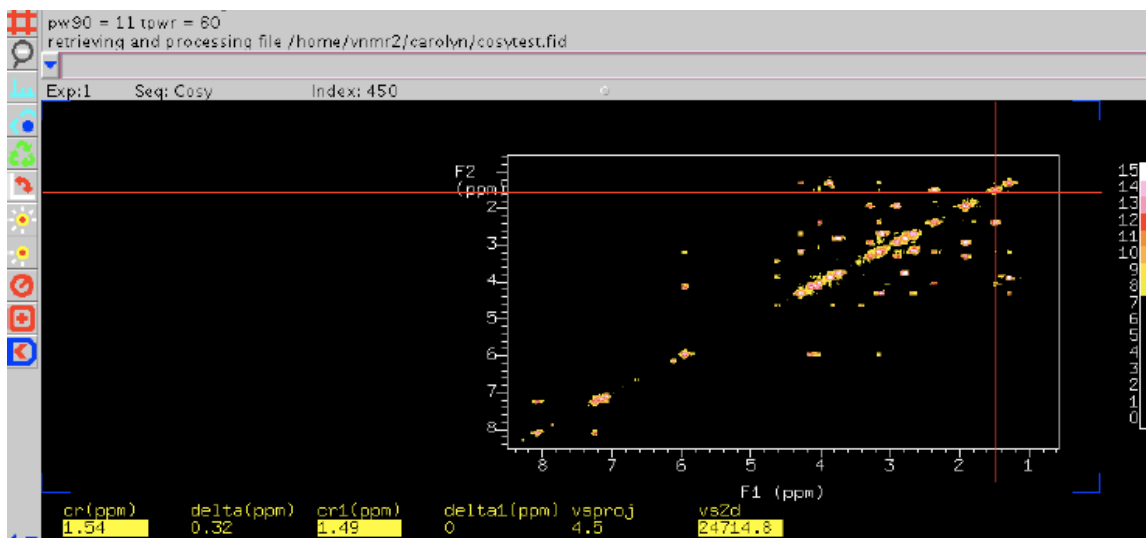
8. Expanding regions of the spectrum: Choose the red cursor icon on the toolbar. Use the left and right mouse buttons to select the area to be expanded and click on the zoom icon to expand. Also use this icon to return to normal size.

Plotting and printing the COSY spectrum

1. Select the Plot tab on the vertical menu.
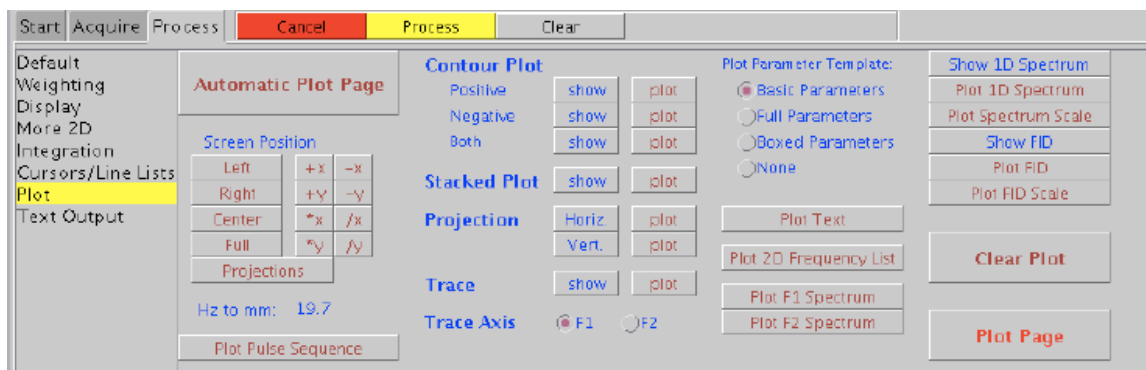
2. Look for the Contour Plot heading, then click on the *show* icon next to the *both* subheading. Click on the *plot* icon just to the right to mount the vertical and horizontal contour.

3. To add your proton peaks to the spectrum, look for the Projection heading, then click on *Horizontal* and *plot* (icon directly to the right) to add the horizontal peaks. Add the vertical peaks in the same manner, by clicking on *vertical* and then *plot*.

4. To print, click on the *plot page* icon.

Finishing the Experiment

1. Remove your sample and replace with the standard sample, then turn on the spin and shim to the required lock level.
2. Under the Standard 1D tab, drag the proton icon to the black window to load standard parameters.
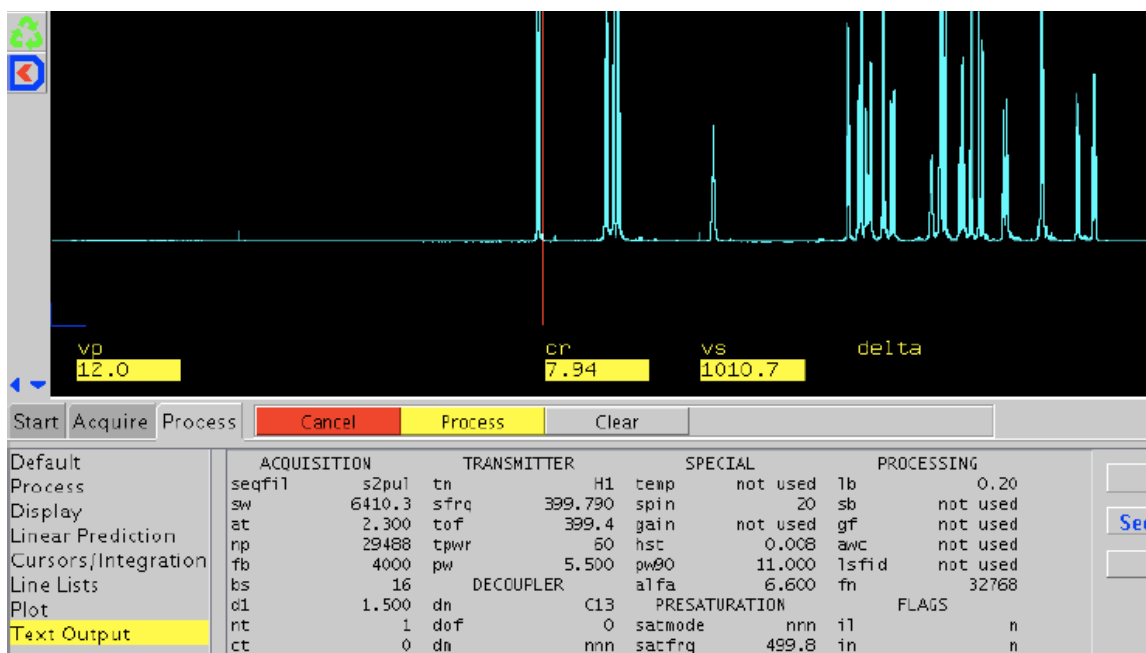3. Exit vnmrj and log out.

VNMRJ CycleNOE Operating Instructions

Preparing for the NOE experiment

1. Use the VNMR ₁H instructions to shim and lock your sample. Change solvent settings as necessary. Drag the proton icon to the black window and acquire a proton spectrum using standard parameters (nt= 1 or 2 is adequate).

2. Type **wft vsadj aph** to process the spectrum.

3. Click on the Acquire and Default H1 tabs. Make sure the range of your spectrum from step 1 fits in the range selected under the *spectra width [ppm]* subheading. 14 to -2 ppm is the default range. If you need to change this range, make sure the new values are used throughout the experiment (starting over will revert the range to the default value).
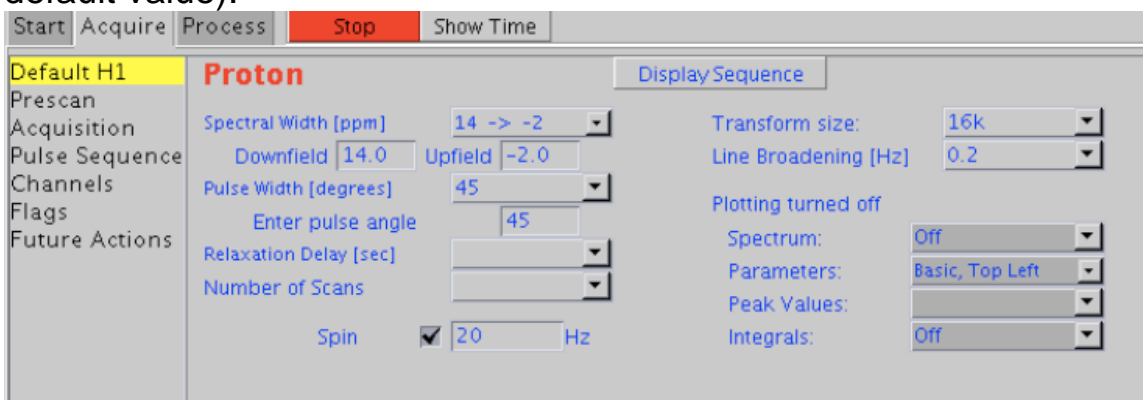
4. Expand the peak you would like to irradiate, and place the red cursor in the middle of the peak. Type **movetof** in the command input box.

5. Click on the Process and Text output tabs. Type **dg** to update the value for tof. Record the resulting tof value for use later in the experiment.

6. To obtain the background tof value, place the red cursor just next to the peak you previously irradiated, but only on the background where the spectrum contains no peak. Type **movetof**, then return. Once again type **dg** and press return. Record the resulting tof value for use in the next step.

vp   cr   vs   delta
12.0   7.94   1010.7

Start Acquire Process   Cancel   Process   Clear

	ACQUISITION		TRANSMITTER		SPECIAL		PROCESSING	
Default	seqfil	s2pul	tn	H1	temp	not used	lb	0.20
Process	sw	6410.3	sfrq	399.790	spin	20	sb	not used
Display	at	2.300	tof	399.4	gain	not used	gf	not used
Linear Prediction	np	29488	tpwr	60	hst	0.008	awc	not used
Cursors/Integration	fb	4000	pw	5.500	pw90	11.000	lsfid	not used
Line Lists	bs	16		DECOUPLER	alfa	6.600	fn	32768
Plot	d1	1.500	dn	C13	PRESATURATION		FLAGS	
Text Output	nt	1	dof	0	satmode	nnn	il	n
	ct	0	dm	nnn	satfrq	499.8	in	n

Acquiring the NOE spectrum

1. Type **cyclenoe** in the command input box. Under the Process and Text output tabs, enter the value for control (background) by typing **control = x** and then pressing return. The number for x is the background tof value determined previously in step 6.

2. Enter the *satfrq* value by typing **satfrq = x**. This value is the number obtained previously in step 5. Type **dg** again and check to make sure the parameters match the modified values.

3. type **nt = x** to input the number of scans for the experiment.

4. Turn off the spin and type **ga** to start the experiment.

5. When the experiment is complete, type **wft aph** to process the spectrum.

6. If you wish to increase the broadness of the lines, type **lb = x** (default value is 0.2).

7. Print the spectrum using the same commands as for a normal proton spectrum.

Finishing the Experiment

1. Remove your sample and replace with the standard sample, then turn on the spin and shim to the required lock level.

2. Drag the proton icon to the black window to load standard parameters.

3. Exit vnmrj and log out.