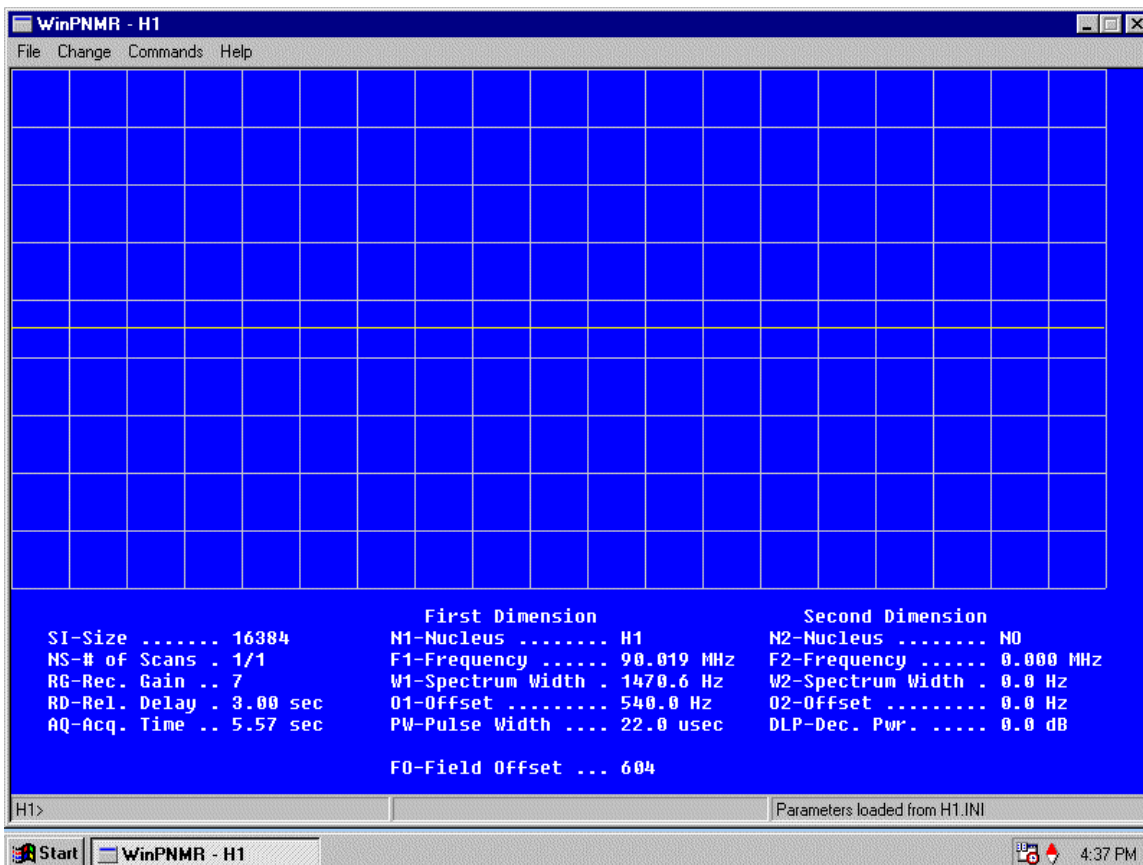


ACQUIRING AND PROCESSING DATA ON THE EFT-90 SPECTROMETER

The *PNMR* Program

The NMR computer uses separate programs for *acquiring* and *processing* data. The program used for acquiring data is *PNMR*; a screenshot of this program is shown below.



Here is a brief explanation of the various parameters:

1. SI represents the size of the dataset; this is the amount of computer memory used to store each file. Typical values are 8192, 16384, or 32768.
2. NS represents the number of scans. Typically 4 or 8 scans are used for an H-1 spectrum; up to several thousand scans may be required to obtain a suitable C-13 spectrum.
3. RG is the receiver gain, or the amount by which the signal obtained from the receiver is amplified. This value ranges from 1 to 100. The more dilute the sample, the greater the receiver gain will have to be in order to obtain a suitable spectrum.
4. RD is the relaxation delay; this is the time the computer waits between the end of data collection and the application of the next pulse.

5. AQ is the acquisition time; this is the length of time the computer collects data after each pulse. This cannot be set directly, but is a function of SI. The larger the size of the dataset, the larger will be the acquisition time. Feel free to experiment with changing SI to see how this influences the acquisition time.
6. N1 represents the nucleus studied; the two most common are H1 and C13.
7. F1 is the transmitter frequency; for an H1 nucleus this has a value of 90.019 MHz. This number will be different if N1 is set to C13 because C13 nuclei come into resonance at a different frequency.
8. W1 represents the spectral width; the default value is 1470.6 Hz. On a 90 MHz spectrometer, 1 ppm = 90 MHz and so this value is equivalent to a spectral width of $1470.6 / 90 = 16.34$ ppm. This value can be changed, depending upon the desired spectral width.
9. OF represents the offset frequency; the amount by which the transmitter is moved from the base frequency.
10. PW represents the pulse width; the length of time the radio-frequency pulse is applied. Note that this is given in *microseconds*; hence the pulse is applied for a very short time.

The lowest portion of the PNMR screen is divided into three windows; the leftmost window is used to enter commands. To change a parameter, type the parameter, followed by a space and the desired value. For example, to change SI from 16386 to 8192 you would type `SI 8192` followed by the return key.

Acquiring a Spectrum

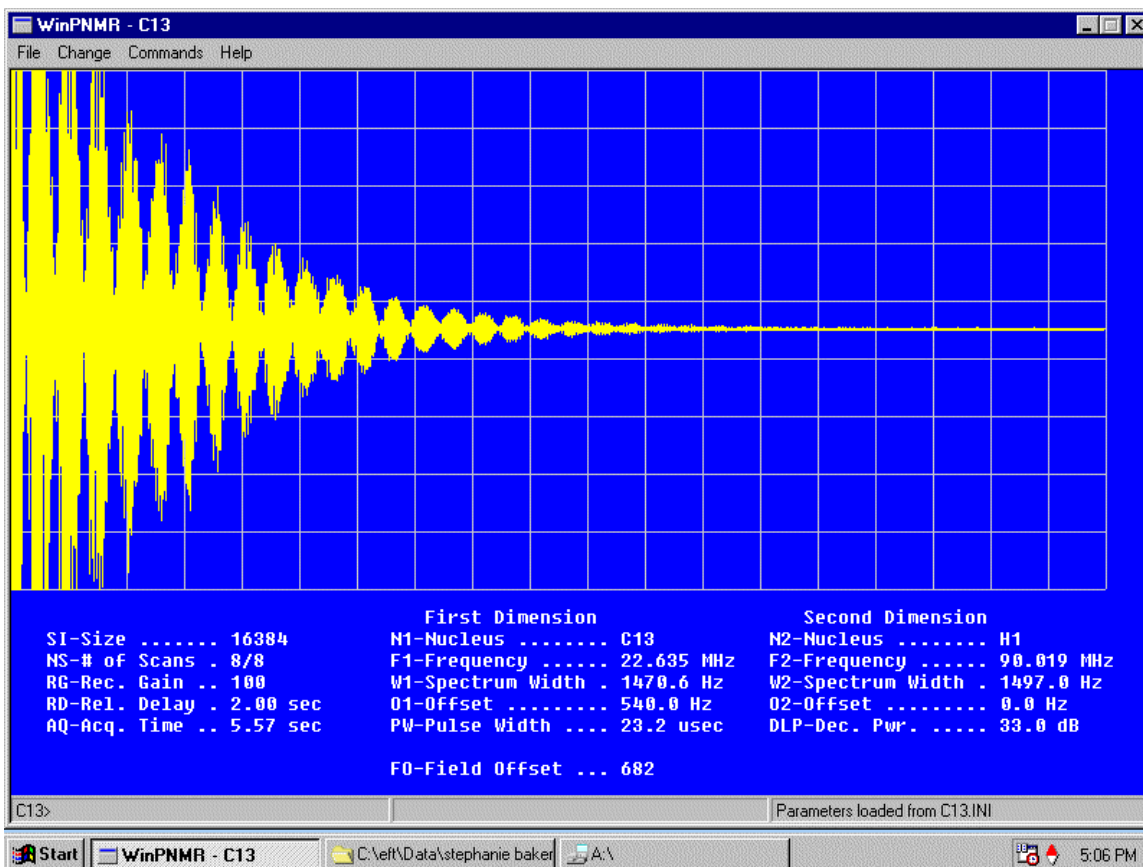
To acquire a spectrum using PNMR, you follow the following steps.

1. Change N1 to the nucleus to be studied; type `N1 H1` or `N1 C13` and press return. If the nucleus is already set to the one you wish to study, you can skip this step.
2. Perform a shim; type `shim` and press return. The purpose of shimming a sample is to obtain a completely homogenous magnetic field; shimming adjusts the current in small coils that augment the magnetic field. Shimming varies from sample to sample, depending upon such parameters as the amount of sample in a tube, or small imperfections in the tube itself. Therefore, it is a good idea to perform a shim every time a new sample is inserted in the spectrometer. Poor shimming usually results in broad or asymmetric lines.
3. Set the receiver gain. The shim routine automatically adjusts the receiver gain, and if you are obtaining an H-1 spectrum this value is usually acceptable. If you are

obtaining a C-13 spectrum, you will most likely need to set the gain as high as possible; in this case enter RG 100 and press return.

4. Set the number of scans. If you are obtaining an H-1 spectrum, usually 4 or 8 scans is sufficient. If you are obtaining a C-13 spectrum, you will need more scans, depending upon the concentration of the sample.
5. Type `zg` and press return. You will be prompted to enter a filename for your dataset. The software is configured to look for data in the data directory; so your filename should be entered in the form `data\filename`. It is a good idea to get into the habit of using descriptive filenames for your datasets. For example, if you use filenames such as `mydata1` and `mydata2` you will quickly lose track of your files. Better names would include the sample and type of experiment performed, such as `sarah_unknown1_H1` or `sarah_unknown1_C13`.

In the Fourier transform experiment data is collected as a function of *time* rather than frequency. The resulting data is referred to as a FID or *free induction decay*. A typical FID is shown below. If the FID is shown in red the receiver gain is too high; you should re-acquire the data with a lower receiver gain.



You are now ready to process your data.

Processing Data

Processing data is performed using the NUTS program. If this program is not already loaded, load it now.

1. Performing the Fourier Transform

The NUTS program has a series of macros for processing various types of data. A macro is simply a text file that contains a series of commands; the same effect can be achieved by entering the commands manually, but it is easier to use the macro. From the file menu at the top of the screen, choose `run macro`. You will be taken to a dialog box containing a list of macros. If you have acquired a H-1 spectrum, choose `H1.mac`; if you have acquired a C-13 spectrum choose `C13.mac`. After choosing the proper macro, you will be taken to a second dialog box asking for the name of the file to be processed.

For processing C-13 spectra, you will be asked to enter a value for the line broadening (LB). This is a function that is applied to the FID prior to processing. Larger values give a better signal-to-noise ratio at the expense of resolution. A good general-purpose value is 1.0. If spectrum contains a number of closely spaced lines; you may wish to enter a smaller value.

At this point you will see a dialog box that lists the parameters used to acquire the spectrum. If you wish, you may enter a comment (such as the compound name) or a user name. When finished, click the OK button on this box.

WinNuts - C:\left\Data\2-bromobutane\2-BROMOBUTANE_H1

File Edit View Process 2D Process Tools Help

Comment

User Date 12/04/06 (11:57)

pulse EXperiment zg.ppg

Number of Acquisitions 8 Pulse Width 22.0 [usec] Recycle Delay 3.0 sec

Direct Dimension		Decoupler		
Frequency [MHz]	90.019463	F1	0.000000	F2
Sweep Width	1470.6	W1	0.0	W2
Dwell Time [usec]	680.0		500000.0	
Acquisition Time [sec]	5.571		0.500	
Offset Frequency	547.7	O1	0.0	O2
Number of Points	8192		1	
Domain	Frequency		Time	
Type	Complex		Real	

Current Slice number 1

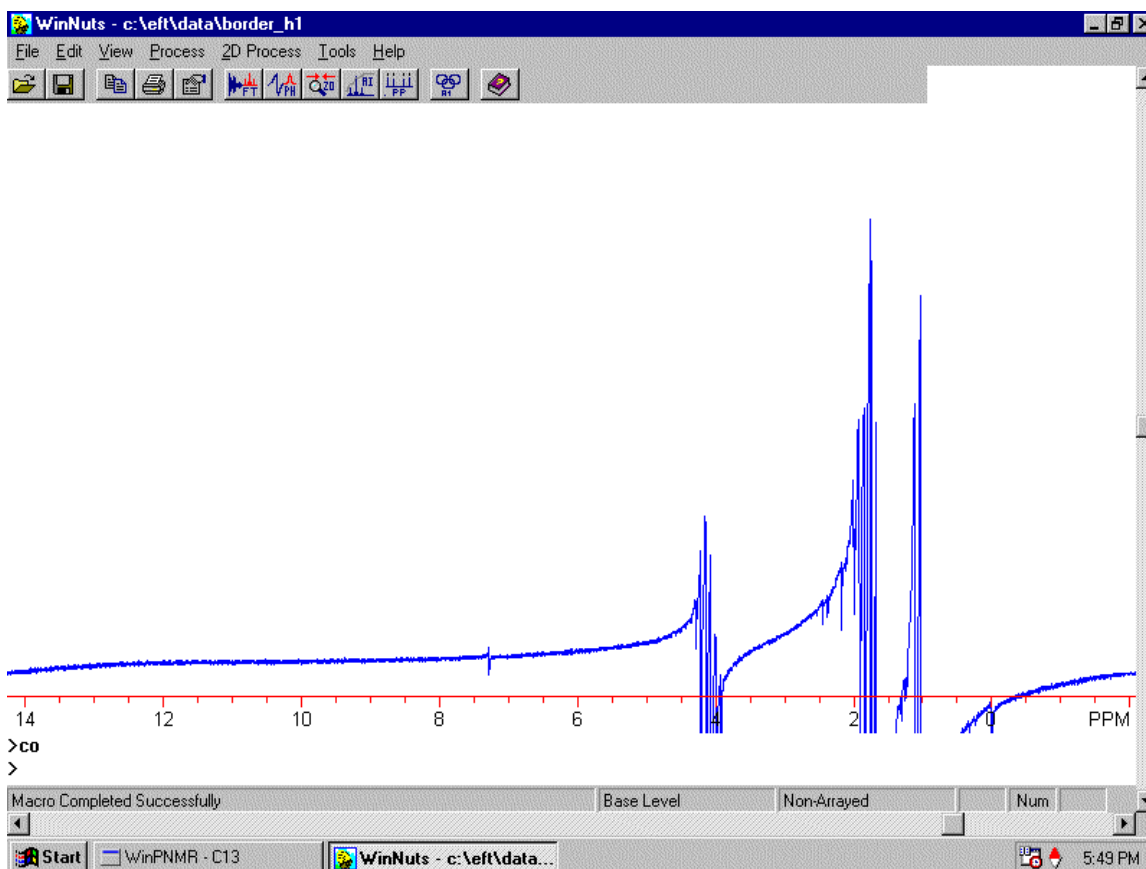
Cancel OK UH and Exit

14 >ac >co >co Base Level Non-Arrayed Num PPM

Start WinPNMR - C13 WinNuts - C:\left\Dat... 5:04 PM

2. Phasing

Once you close the dialog box, you may see something like the screen below. The spectrum looks odd because it has not been properly phased. The next step is to *phase* the spectrum. From the Process menu select Phasing, and then Phase Correct. If this does not give a satisfactory spectrum, select Auto Phase from the same menu. This gives the properly a properly phased spectrum.



3. Choosing a Region of Interest

By default, the entire spectral width is shown when a spectrum is first displayed. However, you may wish to zoom in on a region of interest. To do this, enter the zoom routine by typing `zo` (note that you do not have to press the return key to enter this command); you should see the cursor change to a crosshair. To select a region of the spectrum, hold down the right mouse button and drag the cursor over the region of interest. You will see the spectrum highlighted in red. Then click the left mouse button; you should see your region of interest expand to fill the entire screen. When finished, you must exit the zoom routine by going to the file menu and choosing `exit zo` or by simply pressing the return key.

4. Setting the Chemical Shift Standard

Sometimes the magnetic field of an instrument can vary, and peaks do not always appear in the proper place. This is the purpose of including a chemical shift standard such as TMS (tetramethylsilane) in a sample. The TMS should appear as a singlet at a chemical shift of 0.0 ppm. If it occurs in a different position, you will have to adjust the offset so that it does appear at 0.0 ppm.

Suppose, for example, that TMS signal appears at 0.50 ppm instead of 0.00 ppm. To change the offset, go to the view window and choose `spectral parameters`. On a 90 MHz spectrometer $1 \text{ ppm} = 90 \text{ Hz}$, and therefore $0.50 \text{ ppm} = 45 \text{ Hz}$. Therefore we need to subtract 45 from the offset. If the offset reads 530, for example, enter a value of 485. This will change the scale on the spectrum so that the TMS signal has a value of 0.0 ppm.