

Basic procedures for running the Varian 500 MHz using VNMR-J

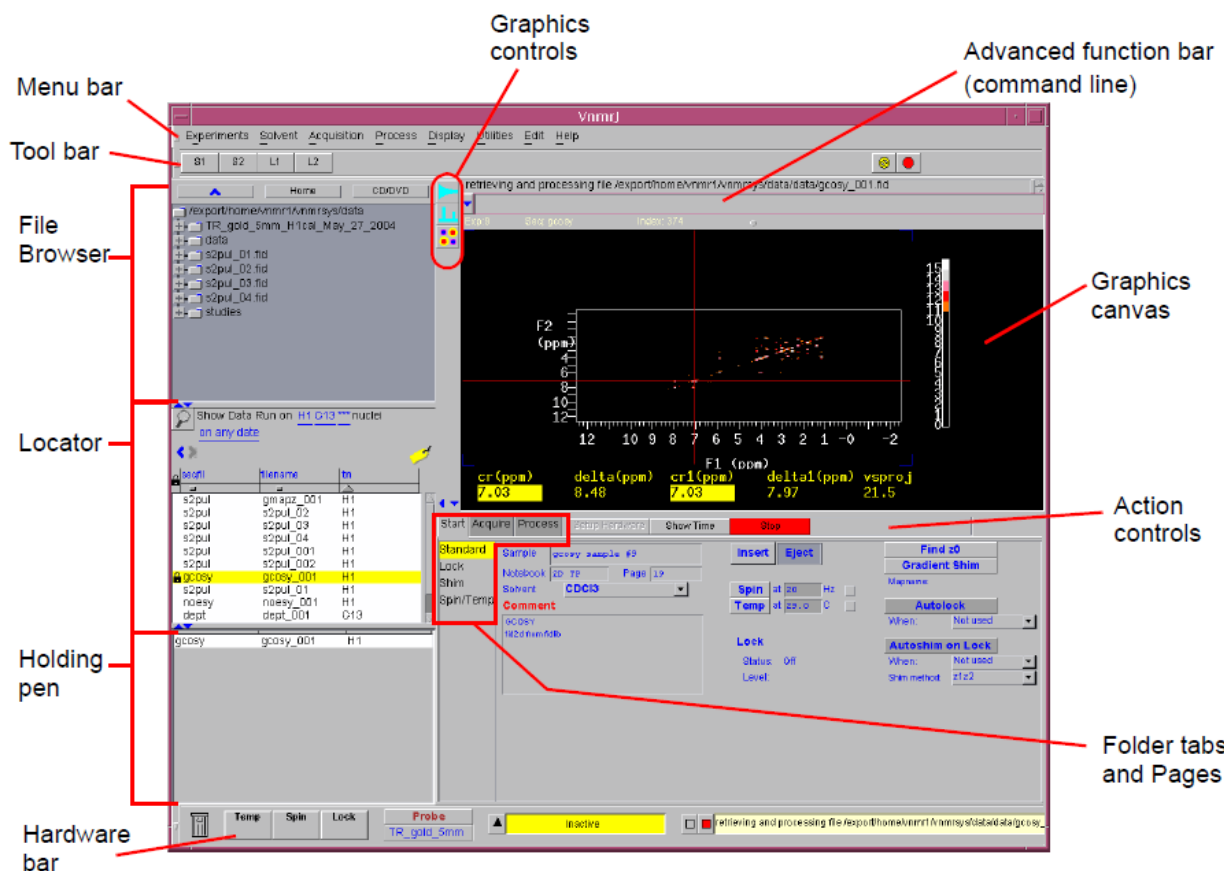
1. Logging in and Start

- type your *username*
- type your *password*.

Start VNMR-J from the CDE graphical interface ("Common Desktop Environment", CDE):



The VNMR-J window should now pop up. Some functions of the graphical interface are:

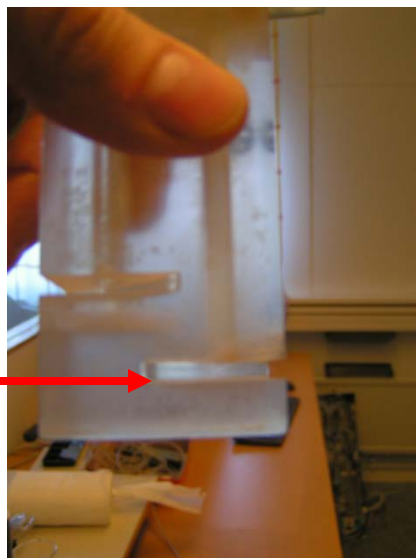


When the setup of the graphical interface is finished, type **reset** in the command line (this macro loads standard parameters for a standard 1D ^1H in CDCl_3 , and standard shims for the standard probe).

2. Sample Positioning Using the Depth Gauge

Set the sample position using the sample depth Gauge provided, shown below

1. Insert the turbine into the top of the sample depth Gauge.
2. Insert the NMR sample tube into the turbine. Gently push the sample tube down until it touches the moveable bottom of the sample depth Gauge. Pull it back a bit so that the round bottom of the tube becomes just 'invisible'.



Would you put the sample at incorrect position it will be impossible to shim it properly and you may destroy the temperature sensor as well as break your sample inside the magnet. Would you observe that the tube easily slides in the turbine and there is a risk that it does not remain in the correct position when put into the magnet please use another turbine.

Would your sample have broken in the magnet contact Anna-Carin (037-703 8215) or Göran (031-786 9022) or Mate (031-786 9033) immediately and do not allow anyone else to try to use the instrument. With a broken sample inside their sample would break anyway as soon as they put it into the magnet...

3. Load a sample

Use the manual eject/inject button on the 'magnet leg' to turn on the air flow to insert a sample. Wait a few seconds to ensure that there is no sample inside the magnet. Put the turbine with your sample on the airflow. Push the eject/inject button again to load your sample.

You should always eject first (even if no sample is in the magnet) to start airflow to carry the sample. The eject air is turned on and the sample, if present, rises back to the top of the upper barrel. If no sample present you will hear the airflow and can simply place your sample on the top of the barrel.

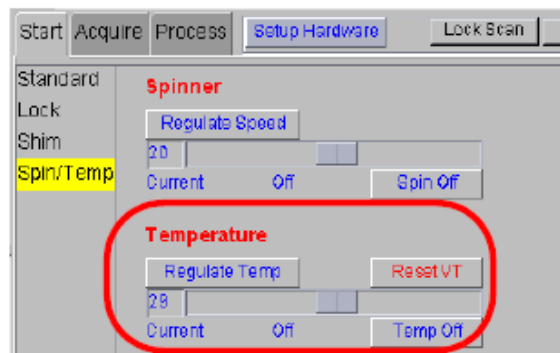
4. Set up standard parameters

Standard shims for the standard Inverse Detection Pulse Field Gradient Probe (IDPFG) are stored as 'CDC13' and 'CD3OD'. Reload them at any time with the commands `rts('CDC13') load='y' su`. These shim sets are only useful for the IDPFG probe.

5. Set the temperature

To set the temperature to 25 °C please type `temp=25 su`. Alternatively you can regulate the temperature from the Spin/temp page (folder tabs and pages). You can alter the temperature without any hardware changes between ambient temperature and 80 °C. For lower or higher temperatures consult Anna-Carin (anca@chem.gu.se).

To turn the temperature control off type `temp='n' su`.



Temperature controls

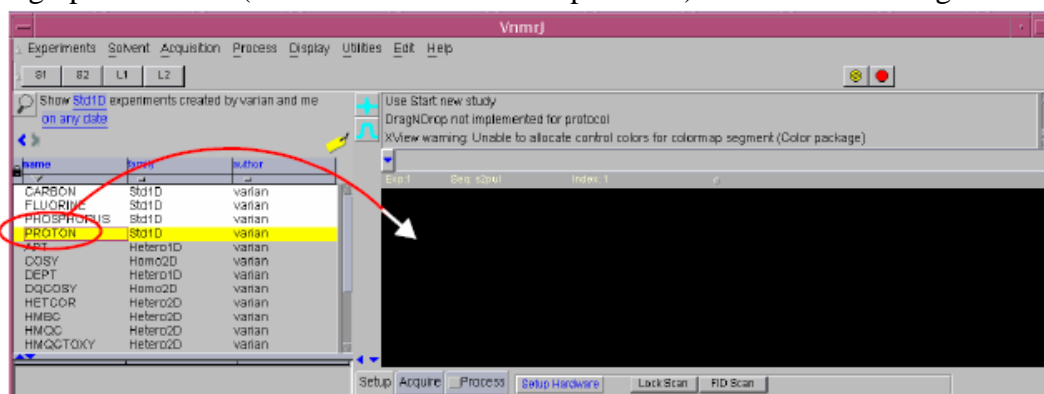
6. Selecting an Experiment

VnmrJ provides you with several ways to choose and load an experiment. This section describes three of these, from the menu bar or from the command line and from the locator. After an experiment is selected, VnmrJ reads and loads the standard parameters and then reads the probe file and loads the probe calibrations.

To select from the menu bar, please push '**Experiments**' on the menu bar and select the experiment you wish to use.

To select an experiment from the **command line** type **PROTON**, **CARBON**, **gCOSY**, **gNOESY**, **gTOCSY**, **gHSQC**, **gHMBC**, etc. Note: use capital letters if you wish get calibrated acquisition parameters.

To select an experiment in the 'Locator' click on the desired experiment in the Locator and drag it into the graphics canvas (or double-click on the experiment) as shown on the figure below.



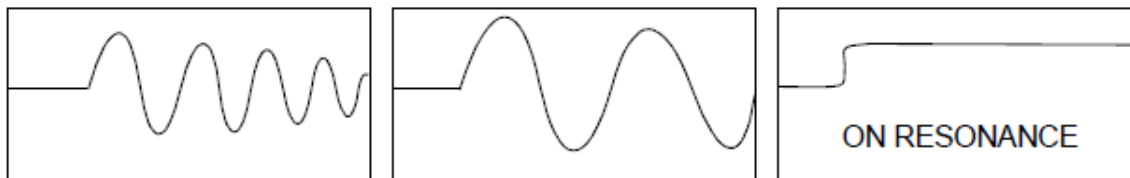
7. Lock

On the action controls below the Setup button choose Lock. Unmark “Activate Lock”. Push ‘Lock Scan’ on the action controls top see the lock signal. Adjust the lock level: find Z0 by clicking on and dragging the Z0 slider bar until lock signal is on resonance.

Approximate Z0 values for some common solvents are:

CDCl₃: 1750; **CD₃OD**: -3500; **CD₂Cl₂**: -1107; **CD₃CN**: -4975; **Acetone-d₆**: 1971.

Changes from poor match to good match

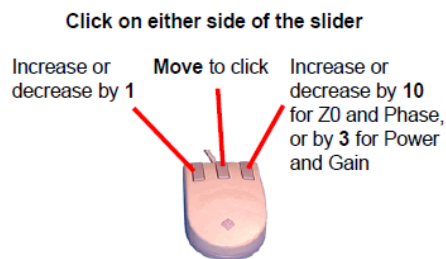
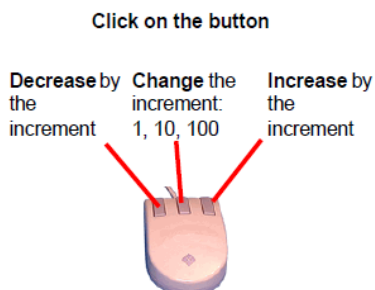
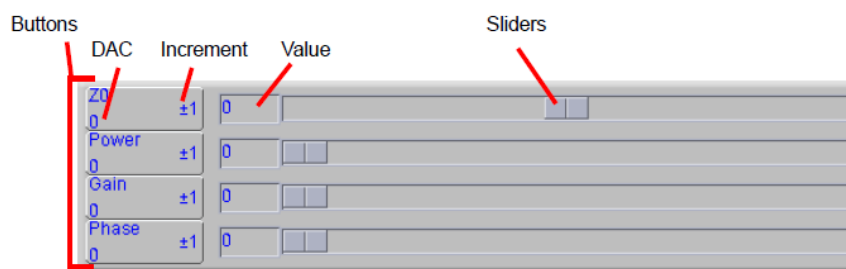


If you see no sine wave (perhaps just noise), click on the button for Z0 (+/-, 100) until some discernible wave appears. If you know the concentration of the lock solvent is high, say greater than 50%, turn down the lock power. If the lock power is too high, the deuterium nuclei become saturated the signal oscillates (goes down and then back up), and it is difficult to establish lock. The correct amount of lock power is difficult to determine, but it is helpful to remember that acetone is more easily saturated than most solvents.

Adjust Z0 until the signal changes from a sine wave to an essentially flat line. If the solvent is concentrated, the line may start to move up on the screen as the lock condition is approached.

Always use lower lock power than lock gain and preferably a lock power below 30. If your lock signal is jumpy you very probably have a much too high lock power (or extremely bad shims)

When you found the Z1 that gives a maximum lock level click in ‘Activate Lock’. Adjust lock phase. The lock and shim buttons provide on-the-fly configuration; the slider values can be moved with the mouse or entered directly:



8. Shimming

For automatic gradient shimming recable the lock cable from the lock filter at the back of the magnet leg to the X-channel observation (J5311). Push the button “**Gradient Autoshim on Z**”. This should give a good shim within a minute in case you selected a good shimmap. When the program shows “Gradient Autoshimming on Z done!” recable the lock from the X-channel (J5311) to the lock channel on the back of the magnet leg.

Ensure that you have proper cabling for 1H detection. Decrease lock power and lock gain until the lock level is between 20-80. For standard 1D experiments the lock level should be between 20-50, for gradient 2D experiments 40-80.



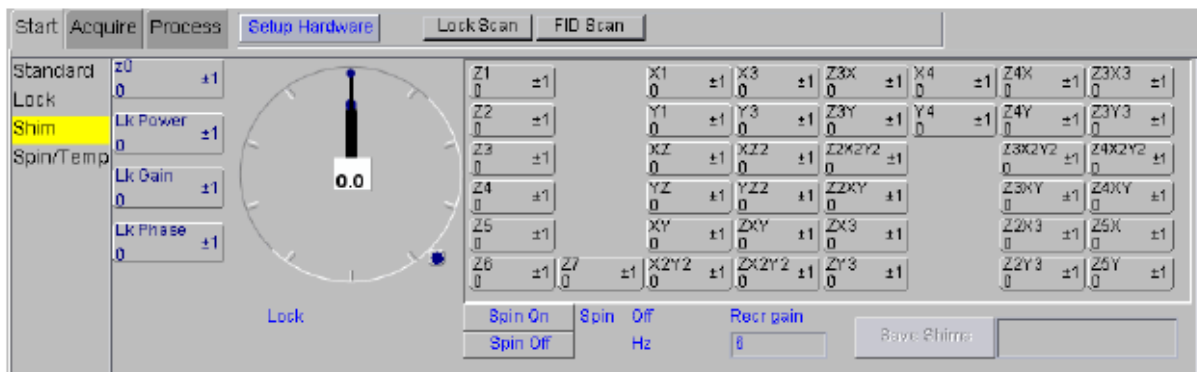
This is the J5311 connection, where 1H should be connected for 1H detection

This is the J5311 connection, where 13C should be connected for 13C detection or the lock for gradient shimming.

This is the back side of the magnet leg, standing close to the magnet. This is the connection of the lock to the lock channel on the probe. This cable you should move to the X-channel for automatic gradient shimming.



If necessary adjust the **shims manually**, by iteratively changing Z1, Z2, Z3, Z4 and lock phase.



- Clicking the left mouse button on the middle button decrements the value by the amount of the increment displayed on the right side of the button.
- Clicking the right mouse button increments the value.
- Clicking the middle mouse button toggles the amount of the increment between one of three values.
- Holding down the **Shift** key and clicking the left mouse button enables you to type in a new current value. Press **Return** to make the value take effect.

A good enough shimming is indicated by a half line width of < 1 Hz on a singlet such as the chloroform residual signal (put the cursor on the signal and type **nl res**). Acquiring a badly shimmed spectrum is just for fools, not for a scientist, since a badly shimmed spectrum has a significantly lower signal pro noise (for a ^{13}C spectrum it may cause several hours of difference in the necessary total acquisition time), distorted information content (couplings) and is not publishable. Generally you don't need to touch the higher order shims to get a well-shimmed spectrum. Would the signal still look bad following a thorough shimming of Z1-Z4 try to adjust the shims Z5-Z7 and X1, Y1, X3 and Y3 and then readjust Z1-Z4.

Are you lost you can try the **best** or **first** buttons to go back to previous shim values. Or read the system shims again (**rts('CDC13') load='y' su**)

To save a good shim file for reuse later on type **svs('filename')**. To retrieve a shim file type **rts('filename') load='y' su**.

Hint: if the shims are very bad possible causes may be:

- The liquid N_2 Dewar is standing much too close to the magnet. Move it a bit further away.
- The sample contains less than 0.6 mL solution. Add more solvent.
- There are particles happily floating around in your solution. They are your enemies. Filter the solution through a cotton plug.
- If you made the classical mistake of putting your NMR tube in the oven to dry it the tube may have changed its shape. Never do that again, but dry with vacuum instead. In this case transfer your solution into another NMR tube and try again..
- Your solution contains paramagnetic rests. You have to live with the bad shims...
- You are working with a solvent mixture – they are tough to shim and you need to book the instrument for an evening or a weekend to practice shimming.

9. Running spectra

Read in the pulse sequence you would like to use – see “6. Selecting an experiment” above.

Adjust number of transients (**nt** – number of transients. To check the quality of your shimming choose **nt**=1, to run standard experiment at least **nt**=4. Adjust **d1** (**d1**=0.7 for 1H, **d1**=3 for 13C, **d1**=2.5 for NOESY, **d1**=1.5 for other standard 2D spectra). Adjust any other parameters you wish... or just run the spectrum with the standard parameter set. To display the set of parameters type **dg**. (**np**=number of points (32768))

Start a 1D experiment with the command **ga** or **go** (**ga** includes an automatic Fourier transformation when the number of transients set by **nt** has been collected, using the **go** command there is no automatic Fourier transformation).

To process type **ft** or **wft** (weighting function is used). To adjust weighting functions type **wti** and change the value of **lb** (**lb**=0.2 for better signal pro noise for 1H, **lb**=2 for 13C spectra) or **gf** and **gfs** (for good resolution **gf**=0.5 **gfs**=0.8 may be a good enough starting value) or **sb** and **sbs** according to your needs. To leave the apodization mode and return to the standard mode type **wft**.

For automatic phasing type **aph**. To change the phase by 180 degree type **phase(180)** To phase manually click the ‘red watch’ on the left from the spectrum. Adjust the phase with the left and right mouse buttons (left for frequency independent, right for frequency dependent phase correction). The intensity of the spectrum is adjusted with the middle mouse button.

If the phasing is messed up completely type **lp=0 rp=0** to reset the left and right phase to their starting values zero and try again to adjust the phases.

Would you observe a rolling baseline –sometimes seen for 13C spectra on this instrument- type **dc** (drift correction). If this would not solve the problem make a ‘left shift’ of your FID to eliminate the distorted points of your FID by typing **lsfid=2 wft aph**.

11. Saving spectra

To name your file type:

```
text('useful_name_that_provides_info_for_identification_of_your  
_spectrum_even_after_several_years_following_acquisition')
```

This text may be identical to the file name.

Save your file by the command **svf('filename')**.

To stop an ongoing measurement type **aa** (abort acquisition). If you would like to stop your acquisition for a short time and then continue type **sa** (stop acquisition) and the **ra** (retrieve acquisition). Of course this will not work with **aa**. If you would like to know how many scans are completed type **ct=?**

12. Displaying spectra

ds	display spectrum
n	display next spectrum if you ran an array
p	display previous spectrum if you ran an array
dssa	display all spectra in an array with vertical stacking
dssh	display all spectra in an array with horizontal stacking
ft	Fourier transform (go from time to frequency, i.e. from FID to spectrum)
wft	weight Fourier transform (go from time to frequency, i.e. from FID to spectrum)
delta	if you would like to check the difference of the frequency of two peaks, for example look for coupling constants, put the two cursors on the spectral lines and type delta?
dconi	display contours interactively
dpcon	display plotted contours: dpcon(10,1.2)
dpf	display peak frequencies
dpir	display integral regions
dps	display the pulse sequence
dscale	display scale
da	display array parameters
f	display full spectrum
full	display full spectrum in full window

In case your spectral window is too small or too large you can adjust it by typing **sw=1.5*sw** to increase the spectral window without altering the transmitter offset (**tof**). Rerun the spectrum with **nt=1**, **ga**, type **f** and adjust the spectral window to the wished size by setting the cursors on the two endpoints of your wish and typing **movesw** (move spectral window). To move the tof (the middle point of the spectral window) set the cursor on the frequency you wish to have it at and type **movetof**.

If you would like to set reference select the signal you know the chemical shift of and type **nl** (nearest line) and then **r1(***p)**, i.e. **r1(7.26p)** for the solvent residual signal in CDCl₃. Solvent shifts and boiling points can be displayed by typing **shifts**.

13. Printing spectra

pl	plot spectrum
pli	plot tabulated integral values
pll	print line list
ppa	plot parameters
ppf	plot peak frequencies
pscale	plot ppm scale
page	send to printer
pir	print integrals

A standard series of printing command is: **pl pscale ppa ppf pir page**

14. Moving data between experiments

mp(1,4)	copy parameters from exp1 to exp4
mf(1,4)	copy fid
md(1,4)	copy display parameters
mt(1,4)	copy text
mz	copy integral regions (reset points)

15. Run carbon spectra, APT, DEPT, etc

Basically there is no difference in running these spectra as compared to running standard ^1H . If you wish to run a standard broadband decoupled ^{13}C spectrum, type CARBON into the command line to read in the standard parameters for it. Adjust the parameters you wish to adjust. Some advice here: A **d1=2** is the minimum for getting an okay quality ^{13}C . Do you have slowly relaxing carbons (for example carbonyls) you may want to increase d1 up to 3-5 seconds to observe them.

Run concentrated samples if you want a spectrum in a half an hour. Do you have a few mg's only – run your spectra over night.

To check for different type of carbons (C, CH, CH₂, CH₃) use APT or DEPT. APT has a 0.7 relative sensitivity as compared to standard broadband decoupled ^{13}C . DEPT has higher sensitivity, on the other hand you may need to run a series of DEPT (45,90,135) to get all type of carbons.

16. Run 2D spectra

The very first time you want to run 2D spectra using gradients please type **newuser** in the command line. This will activate the necessary settings enabling you to use pulse field gradients.

Read in any pulse sequence you wish to use by typing the name of the pulse program in the command line. Remember that capital letters provide you calibrated parameters, so use gCOSY, gNOESY, gROESY, gHSQC, gHMBC, gHMQC, etc. For ^1H homonuclear experiments the labeling is identical to that necessary for a simple ^1H spectrum. For inverse detected heteronuclear correlation experiments (gHSQC, gHMBC, gHMQC) you should move the ^{13}C channel to decoupling from observation (see figure on the magnet leg).

The command **setup2D** will fix most standard parameters in standard 2D experiments. Using this will save you time and understanding; however, you may still need to adjust the number of transients (**nt=16**) so it fits the concentration of your solution, adjust the solvent (**solvent='CD3OD'**) and a few pulse program selective parameters, such as mixing time for a NOESY.

The use for gradient experiments will save you time. However, don't forget to think - there may be reasons why you prefer to use a slower non-gradient version of a pulse program!

To process a 2D spectrum type **wft2da**. To learn more about weighting functions, phasing, linear prediction, etc read the manual, attend an NMR course or ask more experienced users.

2D spectra are saved in the very similar way to 1D spectra with the command **svf**.

17. Finishing your NMR session and logoff:

1. Type exit.
2. Press the exit button on the CDE toolbar (or logoff entry on mouse menu).
3. Confirm that you really want to log off by clicking OK on the window popping up.

Before you leave: Do not keep spectra which are no longer needed in your experiments. A 2D spectrum can easily occupy 20-30MByte on the disk, whereas a stored FID only takes 1-2 MByte. Therefore clean up your experiment library. Type **cleanup** and the data will be erased from all experiments except **expl**.

18. When the computer hangs

Situation: Keyboard and mouse work, the computer respond, but VNMR-J does not let you run spectra just gives error messages or simply ignores your commands

In case VNMR-J is not willing to communicate with the console anymore, please open a unix window – right click above the desktop, choose tools and then Terminal – and type **su acqproc**. This should stop all acquisition related processes. After ca 10-15 seconds type **su acqproc** again, this should now reinitialize the connection and VNMR-J should run nicely again.

Situation: Screen and mouse are frozen, the keyboard does not respond.

Avoid rebooting the computer! Usually only a few processes have run out of control, but the system as such is still working. Rebooting is unnecessary, takes a long time, and may have unpleasant side effects, leading to corrupt data and eventually system crashes later on. Proceed in one of the following ways:

1. Call the system responsible, Ulrika Brath: 031-786 9112
2. In case she is unavailable and there is no risk for a larger damage of the system send an e-mail to the following addresses: brath@chem.gu.se, hilmers@chem.gu.se, mate@chem.gu.se and one of us will come and fix the system as soon as possible. Leave a note at the spectrometer saying that the system crashed, with your name and phone number.
3. If you can't reach Ulrika and there is a risk that the system damages itself call Göran (031-786 9022) or Mate (031-786 9033).

19. Information about specific commands:

- **man(`ds`)** displays information about the command **ds**
- **man(`gHSQC`)** displays information about the pulse sequence **gHSQC**

- Check the manual entitled 'VNMR Command and Parameter Reference' manual.
- Several **printed manuals** are available on the shelves above the workstations
- Download the vnmr-j manual from the website of Varian at www.varianinc.com, by logging in with the account 'usergroup' and pwd 'vnmrj4me'.

20. Some useful commands:

VNMR commands

unlock(1, 'force')	- if your experiment window is locked. (1 indicates exp1 here)
jexp1	- join experiment 1
cexp(1)	- to create a new experiment window
delexp(5)	- to remove experiment 5
axis	- scale units: axis='h' for Hz or axis='p' for ppm
bs	- block size. Set it to 4 to not lose time
ct	- completed transients
su	-setup hardware parameters

Unix commands

cd	change to "home" directory
cd ..	one level up
cd spectra	change to your "spectra" directory
pwd	display name of current working directory
ls	list all files and subdirectories in the current directory (so-called "working directory")
ll	longer list, including file permissions and creation (or modification) dates (alphabetical order)
ll -t	list with newest files first
ll -a	list including "hidden" files ("dot-files")
mkdir project1	create a directory named "project1"
rm	delete a file
rm -r	delete a directory and all its files and sub-directories
mv	move or rename, e.g. mv lepidin.fid lepidin_H1.fid to change a filename mv lepidin.fid /home/adolfs/spectra/protons to move the file lepidin.fid into the specified directory
cp	copy file (e.g. cp letter1 letter.old)
cp -r	copy a whole directory (including files and subdirectories) Note: this would be the command to copy a FID file, since all FID files actually are directories.
more letter	display contents of the file " letter " (only for SCII files!)