

VNMRJ 4.2 INSTRUCTIONS: QANUC 500 FOR CHEMISTS

April 16, 2018

1. Sample preparation

- a. Tubes of any length can be used
- b. Make your samples 4 cm deep. They will not shim as well if they are shorter; you will waste deuterated solvent if they are longer
- c. Make sure that the sample is clear: filter it if necessary
- d. Use a pen to label the cap or the top of your NMR tube if needed (do not use labels and do not write below the top few cm of the NMR tube—this is to avoid ink getting into the inside of the spinner and from there getting into the NMR instrument)

2. The QANUC 500 spectrometer can only be used for certain experiments, which are listed in the McGill tab after login

The spectrometer is **excellent for experiments where ^1H is detected directly**. “Excellent” means that the shims are very good (and therefore the lineshape is narrow and resolution is very good). It also means that sensitivity is very good.

Experiments you can run include 1D ^1H experiments, ^{13}C HSQC, COSY, ^{13}C HMBC, and ^{13}C H2BC.

You may not run ^{13}C , ^{31}P , or other experiments.

3. Experiment preparation

- a. Log in to the walkup NMR software

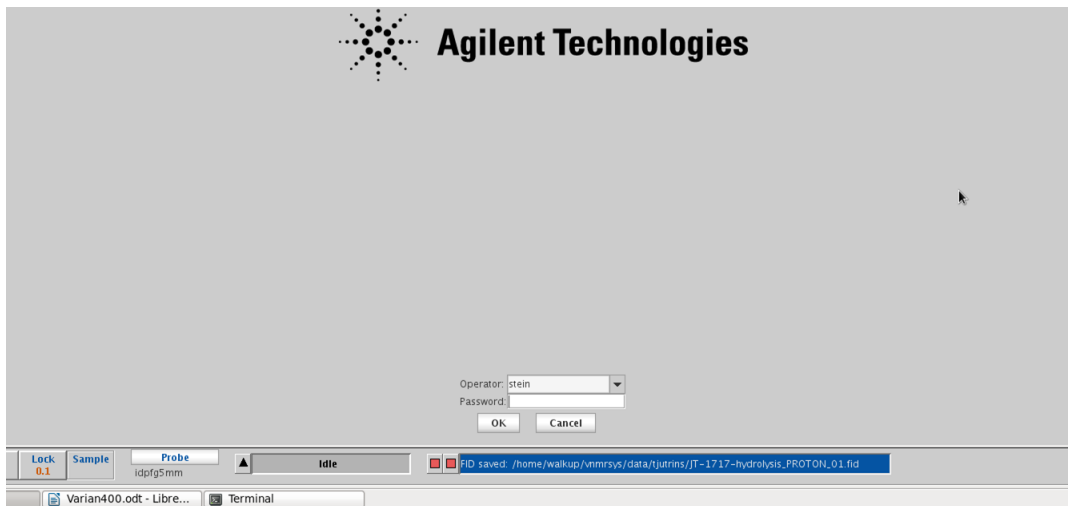


Figure 1: VnmrJ walkup interface login screen. Enter your username and password to log in.

- b. Inserting sample

- i. Place sample into spinner (if no spinner is available, then eject the current sample, step v., before continuing with step i)
- ii. Check that the depth gauge is adjusted to 67 mm
- iii. Adjust sample position with depth gauge (the sample should touch the bottom of depth gauge, unless the sample is very short, in which case it should be raised so that it is centered on the box marked at the back of the sample changer)
- iv. Wipe NMR tube with Kimwipe without affecting its position in the spinner
- v. Click the button marked “eject” or type **e**
- vi. Listen for the jet of air. When you hear it, remove the dummy or previous sample and place your sample on the jet of air
- vii. Click the button marked “insert” or type **i**



Figure 2: Sample adjusted properly in the depth gauge.

4. Queuing experiments

- a. On the bottom left, click on New Study
- b. In the middle of the screen, enter a sample name (usually of the form <initials>_<lab notebook number>; for example, RSS_0216). **Only use letters, numbers, hyphens, and underscores as your sample name.** Do not use spaces or any special characters such as %, #, !, or anything else. Most special characters will prevent email from working and many will prevent the file from being saved properly.
- c. If you want, enter a description of your sample in the “Comments” box. Anything entered here will appear in Mnova or other NMR processing program.
- d. Choose one or more experiments from the list on the upper left of the screen, under the McGill tab. See the end of this handout for a description of the experiments available.

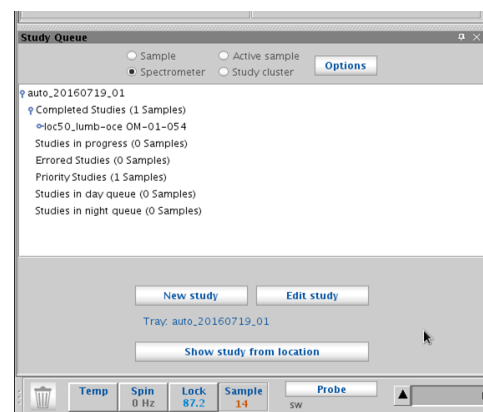


Figure 3: Starting a new study.

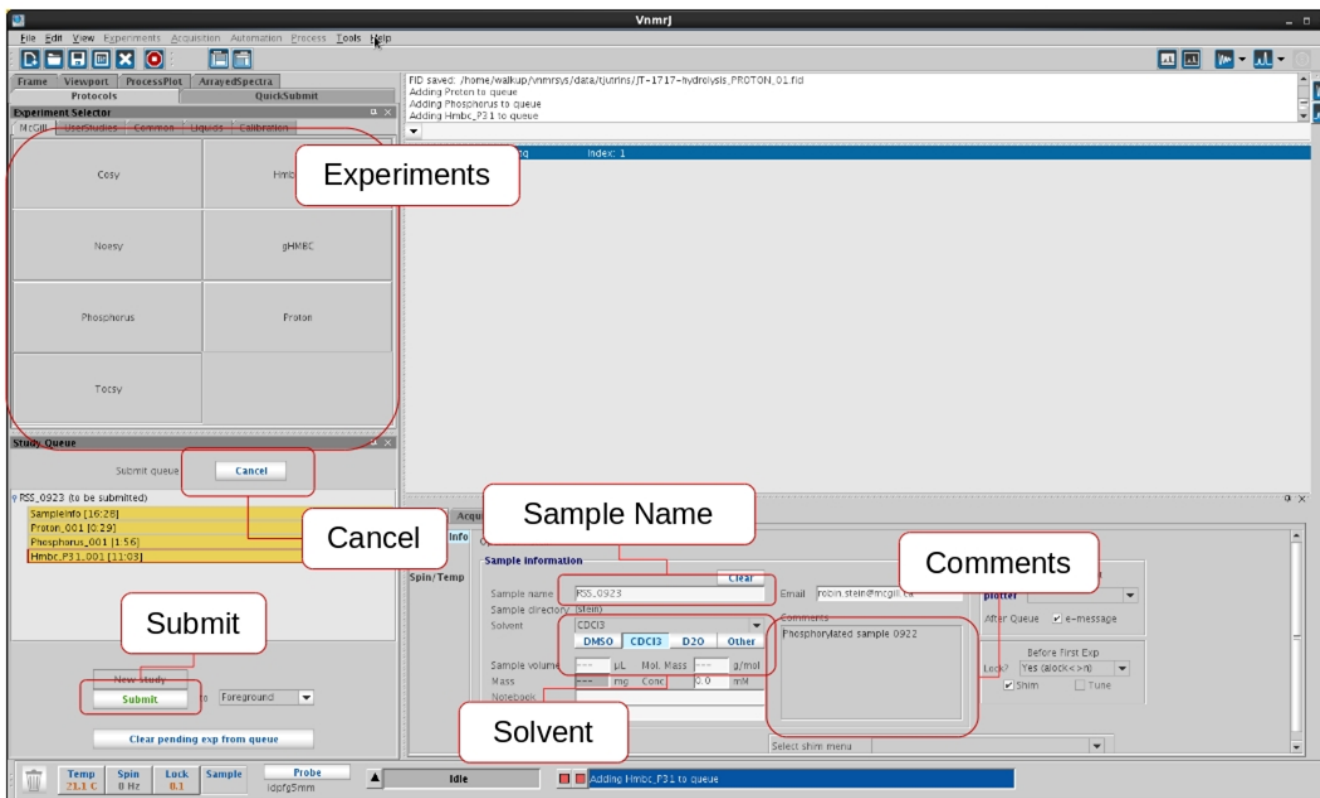


Figure 4: Submitting experiments to the sample queue. At a minimum, choose an experiment, enter the sample name, choose the solvent, choose a sample position, and push submit.

e. To **remove an experiment from the list** (for example, if you change your mind about running it), click on it and drag it to the trash can below the list

f. To **modify an experiment** (for example, to add more scans), double-click on it in the list. Then change one or more of the options listed on the right. If you do not see the option you want listed, click on Acquisition (under the Acquire tab, just to the left of the options shown). After customizing your experiment, click on Save (or click on Quit-No save to cancel any changes you have made). Use the “Show time” button to see how long an experiment will take.

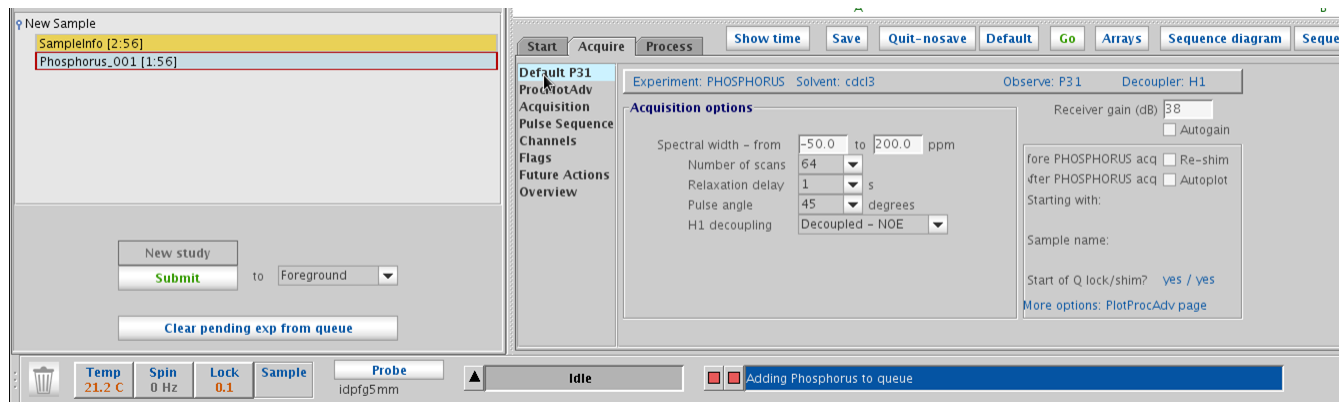


Figure 5: Customizing an experiment. If the parameter you want to change is not displayed, click on “Acquisition” on the left menubar, under “Acquire”.

g. Click on “Submit” to submit experiments to the queue, or click on “Cancel” (above the study

queue list of experiments to submit) to cancel submitting experiments.

5. Processing data at the spectrometer

a. If your data is listed in the Study Queue on the left of the screen, you can double-click on it to open and auto process it.

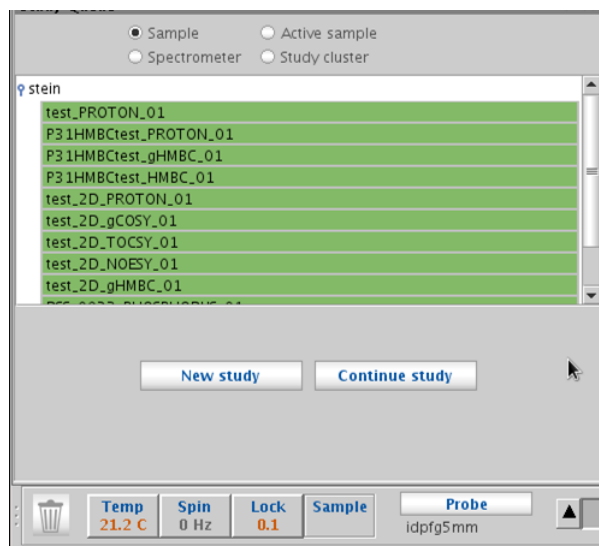


Figure 6: Experiments that have run already (green) and can be viewed by double-clicking on them. Experiments in blue (currently running) can be viewed if the number of completed scans (**ct**) is greater than the number of scans in the block scan (**bs**).

b. You can also go to the File menu and choose Open. Your data will be in `/home/walkup/vnmrsys/data/<username>`.

c. The basic processing command for 1D data is `wft`; for 2D data, it is `wft2da`. (All typed commands are entered in the command bar at the top of the screen.) Or, you can go to the Process tab and click on the Auto process button. If you try to process a 2D dataset and you get an error message about `strflt`, go to More 2D (under Process) and deselect F1 under Linear Prediction.

d. For phasing, type `aph` or click on Autophase in the Process tab. For manual phasing, click on the Phase mode icon to the right of the spectrum. Left click on the right side of the spectrum, then click and drag with the left mouse button (coarse control) or the right mouse button (fine control) for 0 order phasing (which affects all peaks across the spectrum). Make sure that the peaks at the right side of the spectrum are phased correctly. Then, left click on the right side of the spectrum. Click and drag with the left mouse button (coarse control) or the right mouse button (fine control) for 1st order phasing (which affects the peaks on the left more than on the right). Make sure that the peaks on the left side of the spectrum are phased correctly. Repeat 0 and the 1st order phasing as many times as needed. Then click on the Redraw icon to quit the phasing mode.

e. To adjust the vertical scale, type `vsadj` or use the mouse wheel. If integrals are displayed, then this command will adjust the integrals, so remove the integrals by clicking twice on the Integrals icon to the right of the spectrum. To move the baseline of the spectrum, try typing `vp=10` or click and drag the far left side of the spectrum up by treating the mouse wheel as a button.

f. To peak pick, go to the Process / Default panel and click on Find Peaks. Then click on Peak Threshold to show the threshold. Use the left mouse button to position the threshold appropriately (click or

click and drag).

g. To integrate, click on the Process / Integration panel and click on the Partial radio button. Click on Clear Integrals and then on Interactive Resets. Left click to start and to stop every integral (ie left click on the left and left click on the right of each peak) and then click on Scale display to fit. If necessarily, type the command `dc` for a baseline correction (which should level the integrals). To normalize the integrals, place the cursor on an integral with known proton number, enter that number in Integral Area, and click on Set integral value (below it).

6. Plotting and generating PDF of data at the spectrometer

- Zoom in on the region you are interested in and adjust the vertical scale.
- Go to the Process / Plot panel. Select the items you want to plot (eg Integral lines (Partial), Integral values (Horiz), Peak labels (ppm, top in the drop-down menu), Axis (ppm), etc.
- To generate a PDF for saving or emailing, click on Preview
- To email yourself a copy, save the Preview to the documents folder, then start Firefox and email it to yourself

7. Processing data offline

Your data will be emailed to you as a ZIP file. Once you unzip it, you can drag and drop it into Mnova. Or, you can use the `vconv` command in TopSpin to open it there. See the TopSpin notes for more information about reading Varian data in TopSpin (1D data is straightforward, but some 2D data is problematic.)

8. Finishing and logging out of walkup interface to VnmrJ

Use **e** (eject) or the Eject button and **i** (insert) or the insert button to retrieve your sample and insert a dummy sample into the spectrometer.

Click on Logout (right-middle of the screen, with Start selected) to log out of VnmrJ. Note: do not close the VnmrJ program or log out of Linux. After you log out, you should see the login screen (Figure 7) displayed.

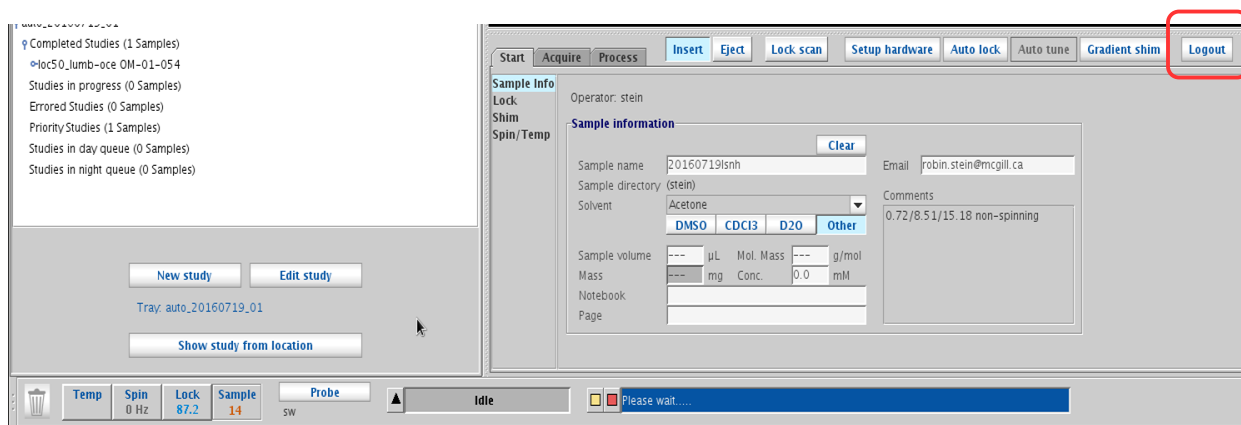


Figure 7: Log out of VnmrJ by selecting the "Logout" button. Do not quit VnmrJ and do not log out of Linux.

9. Some notes on experiments

- General notes on parameters

Some parameter to consider changing are:

nt, the number of scans; the more scans you acquire, the better will be your signal-to-noise ratio.

d1, the relaxation delay (the time between the end of one acquisition and the next excitation pulse); if this is too short, then spectra may not be quantitative (integrals may not appear correct). Increase it to 5 or even to 10 s for more quantitative spectra.

sw, the spectral width; if this is too small, then you will not see highly shielded or deshielded protons. Check that the spectral width covered is sufficient for your expected sample.

b. Proton

The basic proton experiment has 4 scans and takes 15 s (plus sample change, locking, and shimming). The main parameter to change is nt, or number of scans: consider using 1 or 2 scans if your sample is reasonably concentrated, or 16, 32, or even 64 to increase your signal-to-noise ratio. For better integration, you might increase the relaxation delay by a few seconds. If you expect proton signals to show up with very low chemical shift values (for example, hydrides), then be sure that your spectral width is wide enough.

c. Hsqc

This experiment correlates protons with directly bonded carbons. CH and CH₃ carbons have the same sign (generally positive) while CH₂ carbons have the opposite sign (generally negative). Pay attention to the spectral width of ¹³C (under the Defaults tab) – if you don't have any aldehydes, you can reduce it to improve your resolution, but if you do have aldehydes, you may want to increase it to be sure you can see them.

d. Hmbc

This experiment correlates protons with carbons that are two and three bonds away. It is somewhat sensitive to the average J coupling chosen under the Defaults tab. It is less sensitive than the HSQC experiment. Pay attention to the spectral width of ¹³C (under the Defaults tab) – if you expect ¹³C with a very large chemical shift, make sure your range extends out far enough.

e. Cosy

This experiment is a 2D experiment in which the presence of a cross-peak (a non-diagonal peak) indicates that two protons are J-coupled to each other. You might need to increase the number of scans above 1.

f. Noesy

This experiment is a 2D experiment in which the presence of a cross-peak (a non-diagonal peak) indicates that two protons are near each other in space. For small and medium sized molecules, the NOESY effect will be greater at lower field than at higher field. Parameters to consider are: the number of scans, the number of increments (128 may suffice, but will give poorer resolution in the vertical or indirect dimension), and the NOE mixing time (the longer the mixing time, the further the correlations that can be seen, though with too long a mixing time, relay transfers become important). Note that chemical exchange between two sites appears in NOESY spectra in the form of peaks that are the same sign as the diagonal peak and the opposite sign as the off-diagonal peak.

g. Tocsy

This experiment is a 2D experiment that correlates all protons that are in the same spin system. So, if

proton A is J-coupled to proton B, and proton B is J-coupled to proton C, protons A, B, and C will show up in the same row and same column, even if A is not J-coupled to C. The TOCSY mixing time (“Spinlock duration”, under Defaults) can be increased to see longer range J-coupling or decreased to see shorter range J-coupling.

10. Troubleshooting

a. *To start VnmrJ if it is not running*

Click on the VnmrJ icon on the desktop to start VnmrJ, then log in when prompted.



Figure 8: VnmrJ icon on desktop.

b. *If a sample breaks outside the magnet*

Please clean it up using the broom and dustpan near the sink in room 35 and the glass disposal box in room 37.

c. *If a sample breaks inside the magnet*

Report this to Robin or Tara as soon as possible in person, by email (robin.stein@mcgill.ca), or by phone (x6219). Tell her what kind of sample it was and whether any special safety precautions are necessary. Use FACES to block out some time and put a note near the spectrometer that the spectrometer is unusable

d. *If you want to stop a running experiment*

Go to the Automation window, go to Acquisition / Background Acquisition / Stop/Save/Resume.