Step-by-Step Instructions – Varian Spectrometers

I. *^1H* 1d acquisition

[updated: 27 May 2010]

1. *Current sample out*: Check the STATUS panel to insure that Status = IDLE , i.e., that it is not ACTIVE, in which case the previous user is still acquiring data.
2. If the previous user does not show up within 5 mins of the start of your time, do an **svf**↵ and save the data to a temporary filename. Then enter **aa**↵ to stop the acquisition.
3. **eject** or **e** or click on EJECT in the *acqi* window
   - you should always have ≥ 0.45ml (0.6ml recommended) solvent; with less (w/o a Shigemi tube) you may not be able to obtain a reasonable shim.
   - for samples with ≤ 0.6ml solvent, be careful to *center* the solvent column in the hatched box—indicating the detect region—in the depth gauge. Failure to do so may lead to poor line shape, even following significant shimming.
4. *Your sample in*: **insert** or **i** or click on INSERT in the *ACQI* window
5. Set the temperature:
   - on the UNITY, use the PC to set the temperature; set **temp** in VNMR correctly, but keep in mind this does not affect the actual temperature
   - on the INOVAs, enter **temp**↵
   - if you are making a change >5°, allow at least 10 mins for the temperature to equilibrate prior to shimming or tuning the probe
6. Read in the facility shim file:
   - **rts**↵ ***probename*.shim**↵ **su**↵ or
   - click on MAIN MENU → UWMACROS → LOADSHIMS and type in shim filename
7. *Read in experimental parameters*: click on MAIN MENU → SETUP → NUC,SOLV
8. *Locking*: 2 methods can be used:

   <u>*Automatic locking*</u> (recommended; as of June 2009)
   - enter **findlock**↵ and wait for the macro to finish
   - in the *ACQI LOCK* panel: turn the lock on
     - → if the signal does not lock, try increasing the LOCKPOWER and LOCKGAIN
     - → adjust the LOCKPHASE so the lock signal goes positive;
     - → the spectrometer should now lock; re-read in shim file; find help if still no lock

   <u>*Manual locking*</u> (the automatic *findlock* method above is recommended)

   In the *ACQI LOCK* panel:
   - turn the LOCK off
   - turn LOCKGAIN and LOCKPOWER both to 60

- adjust Z0 to remove all oscillations in the lock signal (lock to on-resonance); use only the slider bar and/or the ±64 button for Z0 adjustments
- reduce the LOCKPOWER slowly (−4 at a time) to get a good lock level (which is any lock signal > 30 and < 100)
- the lock signal should go positive; adjust LOCKPHASE if not
- click LOCK when on-resonance and a positive lock signal

9. Reduce the LOCKPOWER to the recommended value for the solvent, but still keeping the lock level > 20. You may have to adjust the Z1 shim prior to getting the LOCKPOWER to the recommended value.

10. *Shimming:* Go into the *ACQI SHIM* window, and stay there unless LOCK is lost (the last panel in the *SHIM* window has LOCKPOWER, LOCKPHASE, etc.)

    - ***Always*** "shim" LOCKPHASE **prior** to careful shimming on Z1 and Z2.
    - Shim Z1, then Z2, then Z1; repeat until no improvements are obtained. Re-"shim" LOCKPHASE following any large changes in shims (e.g., following a change in Z2 ≥ 128 units).

11. ***DISCONNECT from the ACQI* window prior to doing anything in the main VNMR window; failure to do so may lock-up the Sun workstation.**

12. *Tune the ^{1}H channel on the probe*: this must always be done for the first sample.

    For *^{1}H* 1d and COSY experiments, the probe need not be retuned for following samples at the same temperature and similar solvent conditions.

    For *^{13}C*, *^{31}P*, and other *X* experiments (where high-power *^{1}H* decoupling is used), and for more advanced experiments—NOESY1D, TOCSY1D, ROESY1D, DQCOSY, HSQC, HMBC, etc— the probe should be retuned on ***every*** sample.

13. *Check cabling (UNITY only)*: check that the *^{1}H* cable to the probe is plugged into the 1H OBSERVE port on the magnet leg preamp box.

14. *Adjust receiver gain*: enter **nt=1 ss=0 gain=60 ga↵**

    - Look and listen carefully following the **ga**: two "beeps" and a message "ADC Overflow" or "Receiver Overflow" means **gain** is too high. Reduce the **gain** by 20 until you get only one "beep" and neither message above.
    - (UNITY only): If you still get two "beeps" and one of the messages above when **gain=0** , then you must insert an external attenuator(s) into the labeled "Attenuator" position on the magnet leg preamp box.

15. *Check your shims*: Expand the spectrum about the TMS (or other solvent) peak; typically one scan (**nt=1**) is sufficient to check shims, but take more scans if necessary. Type **nl dres↵** to get an estimate of the linewidth. For small organic compounds, 1Hz or lower is typical. For peptides and proteins, ≤2Hz may be acceptable.

16. Set **ss=2** and **nt=*i*×8** (ie., a multiple of 8) and acquire the FID.

17. Save the data using **svf↵** ; use a filename ***without*** spaces or other special characters (underscore _ plus + period . and hypen - are ok to use in a filename).

18. Prior to leaving the spectrometer, put the standard CDCl3 sample into the spectrometer, and establish lock on this sample (a higher than normal LOCKPOWER of ≤ 40 is OK for this sample; ie., don't spend time shimming this sample).
19. Useful commands for processing and plotting the data are:

    ff dsx aph dc va

    pl pscale(0) pap page