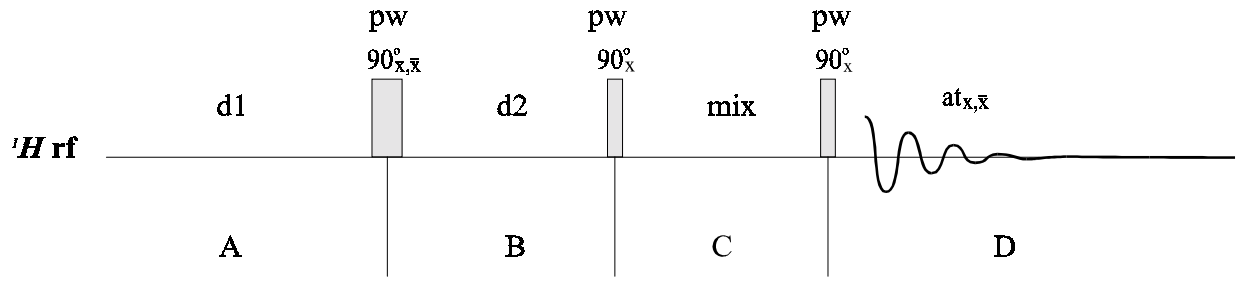
XI. NOESY – 2d NOE and Exchange Spectroscopy

(17-Jul-00)

A. Discussion

- NOESY can be a powerful experiment for the correct types of compounds, and with proper care during acquisition and analysis of the data. **Care is recommended during interpretations!** See Sanders&Hunter Chap. 6 for an excellent introduction to NOE's; see the Bruker User's Guide for more details about NOE experiment particulars.
- Often, combinations of MW, solvent, and temperature conspire to make NOESY crosspeaks non-realizable during nominal mix time in this transient experiment. ROESY will often be better alternatives for small (≤2000) MW.
- Performing at the least an inversion-recovery null-estimate of ^1H T_1 values is highly recommended prior to acquiring NOESY spectra. This method of obtaining ^1H T_1's is quick, and simple to setup. Perform full T_1 analysis for best results (see **dot1** macro).
- Degassing leads to longer T_1 values for smaller MW compounds, and is recommended for 1D variants. The longer T_1 values, which can lead to greatly enhanced NOE build-ups, sometimes cannot be maintained for 2D experiments, due to limitations in time for the experiment.
- NOESY crosspeaks for small MW will be positive (opposite to the inverted diagonals) as will exchange crosspeaks (use temp variation to differentiate exchange from NOE crosspeaks). For larger MW, crosspeaks will be negative (same sign as diagonals).
- Multiple mix times (≤ T_1) will be required for quantitative work in using NOESY build-up curves for obtaining distance information.
- **ulnoesy** is recommended (but currently unavailable) over the vnmr standard sequence **noesy**. For now, use **tnnoesy** if needing presaturation, otherwise **noesy** (or equivalently SETUP SEQUENCES NOESY).

2d NOESY and EXSY Spectroscopy (ulnoesy)



B. Critical Parameters

pw, tpwr = 90° pulse width at power **tpwr**; recalibrate this parameter for noesy experiments
ni = number experiments, or number of points in t_1 ; should be set ok by macro, time allowing; want F1 digital resolution ≤ 6 Hz/pt = sw1/(2ni)
nt = multiple of 8
sspul = 'y' gives homospoil-90-homospoil preceding **d1**
d1 = relaxation delay; set 2-4*T_1 (do not set too small, or will get very bad t_1 noise)
np = number of points in t_2, usually want ≥ 2048 since costs nothing but disk space and gives better resolution in F2
mix = mixing time; often is varied to provide build-up curves. Set close to T_1 should provide maximized crosspeaks; for high MW (>2000) ~0.3s should work.
mixvar = (ulnoesy only) variation in **mix** in percent; 10 works reasonably well to remove cosy/tocsy type crosspeaks; will increase t_1 noise, so not recommended unless know is needed

C. NOESY Acquisition

- set-up similar to DQCOSY
- make certain to optimize the **gain**
- make certain to optimize baseline flatness for the particular value of **sw** you are working at:
  - acquire spectra with **ni = 2**, and phase carefully
  - run **calfa** which uses the **lp** value from the phasing
  - reacquire the **ni** = 2 data and rephase; you should get a good phase with **lp=0**; if you do not, rerun **calfa** and require again
  - if the baseline still has some curvature (either convex or concave), then you might want to:
    - note the values of **rof2** and **alfa**
    - keeping the sum **rof2+alfa** constant, change **rof2** and **alfa**
    - reacquire and note the effect of the change with **rof2** and **alfa**; one direction should move the baseline towards convex, the other towards concave; some combination should give optimal baseline flatness
    - write down the values of **sw rof2** and **alfa**; these should always work for this **sw**
- as a minimum, estimate T_1 using inversion-recovery null method to provide setting for **d1**

D. Calibration

- always recalibrate 90° pulses for **pw,tpwr** with NOESY spectra (see ^1H section for instructions)
- always work from a measured T_1 estimate

E. NOESY Data Workup and Plotting

- same as **dqcosy** (see DQCOSY section for phase-sensitive workup)
- baseline flatten/fitting routines can be particularly useful for NOESY spectra workup; it is recommended that the baseline is made as flat as possible prior to acquisition (see section C), but **wft1da bc('f2') wft2da bc('f1')** can be particularly useful here; **fn1 = fn** is required for the integration regions in F2 to work for the F1 baseline correction