TOCSY 2D spectroscopy

90

mix

t1

MLEV-17

Z

vectors 'locked' along B1-axis

X

spin-lock$_y$ Y   B1

Introduction

TOCSY is the abbreviation for TOtal Correlation SpectroscopY. It is also called HOHAHA (HOmonuclear HArtmann HAhn). During this pulse sequence, after the evolution period t1, the magnetization is spin-locked (for example by a series of 180` pulses) for a period of time on the order of 20-100 ms. During this mixing time the magnetization exchange through scalar coupling. During this spin-lock period, the magnetization behaves as a strongly coupled spin system and evolves under the influence of a "collective spin-mode". In that collective mode, coherence transfer is possible between all coupled nuclei in a spin system, (even if the are not directly coupled). One of the most popular mixing schemes in TOCSY is the MLEV-17.

For small mixing period, (e.g. 20-30 msec) Relay1-COSY type of data can be observed. As the mixing period gets longer, correlation with more distant protons can be observed (e.g. mix=80-100 msec can correlate H1 to H6 in carbohydrate). The extent of correlation depends mainly on the length of the mixing period.

This sequence is much more useful than the Relay-COSY as it very efficiently transfer magnetization before relaxation takes place.

TOCSY experiment

1. Acquire a 1D 1H NMR first, calibrate **pw90** at **tpwr=57**.
2. Set cursors ~0.5 ppm beyond the peaks on both sides.
3. **movesw↵**
4. re-acquire a 1D NMR.

5. **mf(1,3) jexp3↵**
6. **TOCSY↵**
7. **dg↵**

You should see that **tpwr=57** with **pw=23.25**. the pw was calibrated previously at tpwr=57. the spin lock power, *slpwr*, is usually lower(46-49 dB). This experiment, it is set to 48 which is 9dB smaller than *tpwr*. So *slpw* should be increased in this way. **23.25*2*1.414=65.188.** The math is like that, if you lower 1dB, you need to increase

pulse width by $\sqrt[6]{2} = 1.12$ times. So you have 9 dB decrease, then you need to

increase pulse width by $\left(\sqrt[6]{2} \right)^9 = 2 * \sqrt{2} = 2 * 1.414$.

8. **dps↵**

```
   ACQUISITION            TRANSMITTER             FLAGS              PRESATURATION
seqfil        TOCSY   tn              H1   hs              n   satmode            n
sw           3870.0   sfrq       399.729   sspul           y   satfrq             0
at            0.265   tof         -403.2   PFGflg          y   satdly             0
np             2048   tpwr            57   hsglvl       4854   satpwr             0
ss               64   pw          23.250   hsgt      0.002000       PROCESSING
d1            0.600        DECOUPLER              SAMPLE        gf             0.061
nt                2   dn              H1   date    Mar  3 2004  gfs         not used
ct                2   dm             nnn   solvent      CDC13   fn             4096
  2D ACQUISITION            SPIN LOCK           sample   undefined   2D PROCESSING
sw1          3870.0   slpwr           48          SPECIAL       gf1            0.048
ni              200   slpw        65.188   temp     not used   gfs1        not used
phase        arrayed  mix          0.080   spin     not used   proc1             lp
                      trim        0.0020   gain           24   fn1            2048
```

9. **np=2048**, number of points in t2, it must be even number. A number to the power of 2 is required. This will also give a value to **at**. Bigger **np** gives better resolution in F2 dimension.
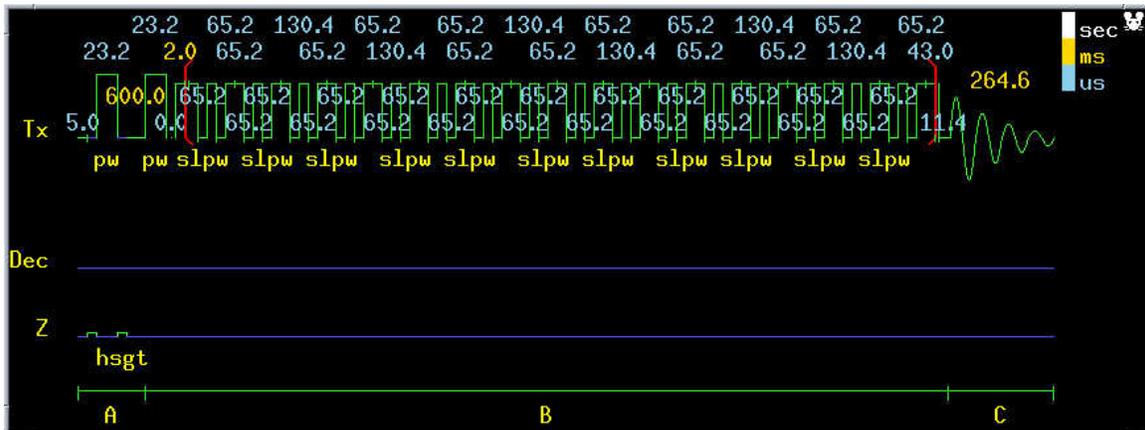

10. **phase =1,2** which is *arrayed* as is seen in dg window. Type **da** to see the details.

10. **at=np/sw=2048/3870**(*which has been already calculated by the computer*)
11. **slpw** is the spin lock pulse width which must be a 90 degree pulse width.
12. **slpwr** is the power of the spin lock, the max is 49. It must be correlated with **slpw**.
13. **mix** is the mixing time; the larger the mixing time the more transfers will occur(*the more protons will be correlated*). It is normally 0.080-0.100 s, which is suitable for 5-8 transfers. Mix is
14. **d1** can be longer than gCOSY. {**T1=200ms** for strychonine, so **d1=0.6** is OK. Normally you should set **d1+at ≥ 3T1. Here 0.6+0.265 = 0.865** which is more than 3*T1(=0.6)}.
15. **ni=200** or shorter if you do not have enough time. To have a reasonable value of ni, please remember this: $ni = sw1/12 \sim sw1/24$. In this experiment, it is between 161~322. So 200 is a suitable value. **{sw1/(2*ni) is the resolution in F1, an idea value is 6Hz/pt if time allows. But it should be smaller than 12Hz/pt}.**
16. turn **spin** off, set **gain** to the highest value which does not cause overflow. **go↵** *if ADC overflow,* **aa↵** *lower gain,* **go↵.{** you can type **set2dgain** or type **ni=1 nt=1 ss=1 phase=1 gain='n' ga** then after the experiment finishes, type **gain='y'}**
17. **svf↵ TOCSY_strychonine(or whatever you like)↵**
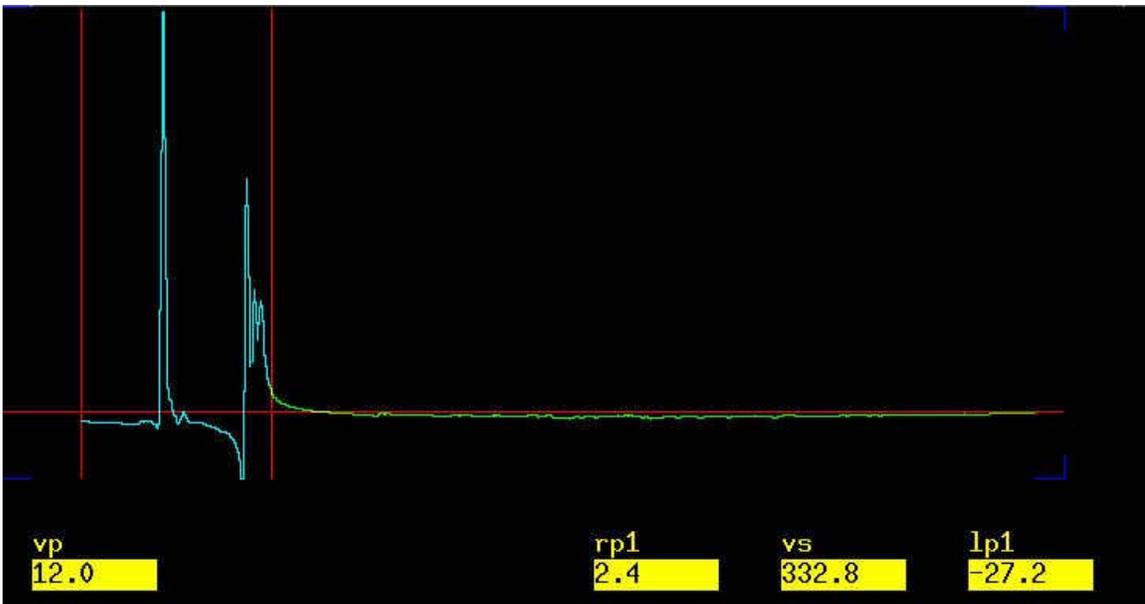18. **setLP1 sqcosine fn=np fn1=fn wft2da↵**
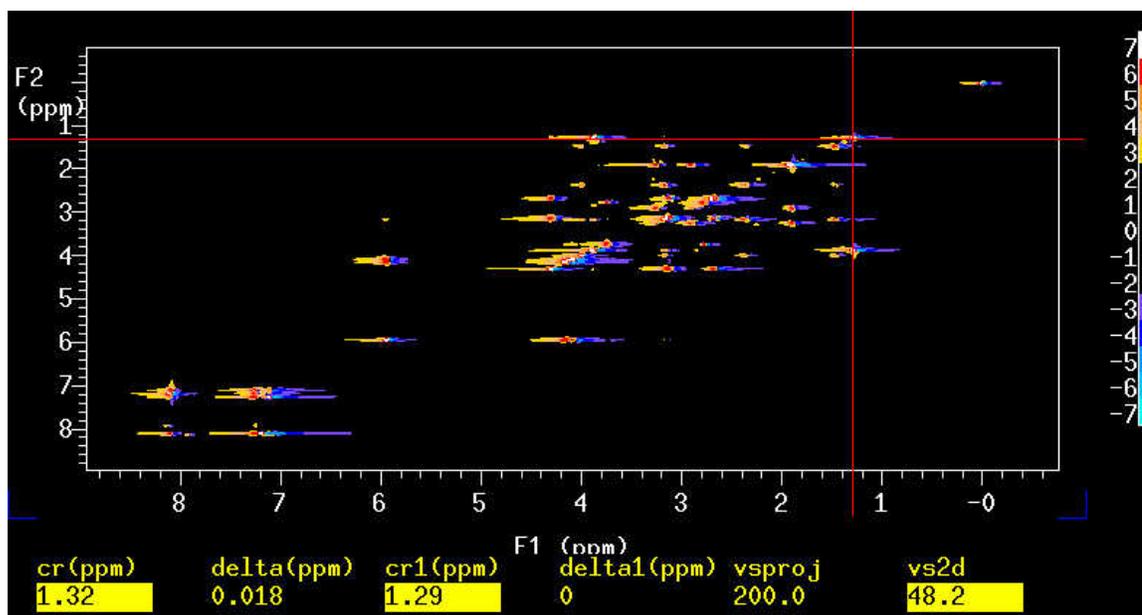
You see the above spectrum. Apparently you need to phase the 2D spectrum.

19. **pmode? pmode='full'** ↵ if it was **'partial'**.
20. **trace='f1'** if it was **'f2'**.
21. point the mouse at the peak of the molecule(do not point at solvent peak)
22. **ds↵**
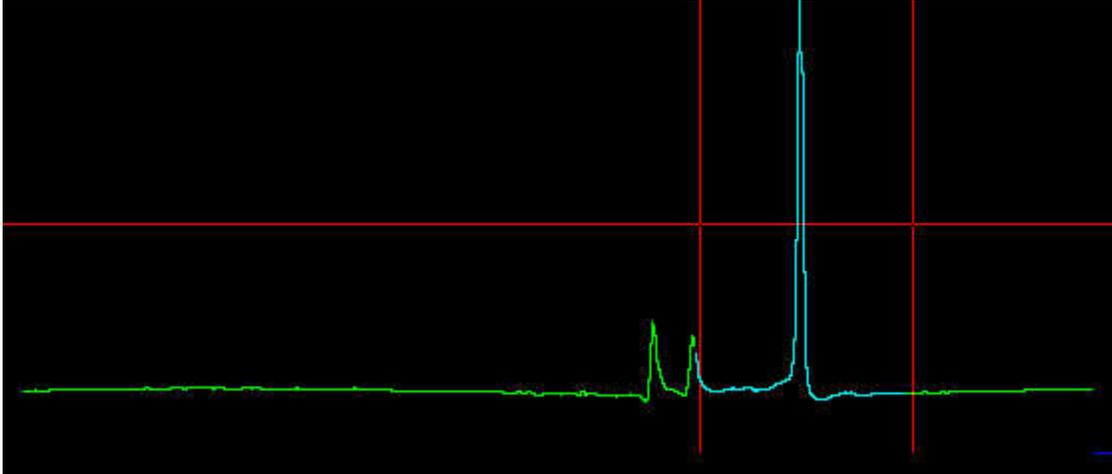
23. click phase button and phase the spectrum as you do on a normal 1D. This time you are adjusting rp1 value, the right phase on f1.

24. **dconi↵** point the mouse at the left peak **ds↵**

vp		rp1	vs	lp1
12.0		2.4	332.8	-27.2

26. click the phase button, then click on the right part of this spectrum, do not adjust the phase at this point. A click at this part sets up the rp1 from the previous spectrum.

25. adjust the phase of the right side peak as shown above.

26. **dconi ↵**

27. **trace='f2',** do the same thing as you did on f2 dimension.

28. **dconi↵** adjust **vs .vs+20%; vs-20%.**
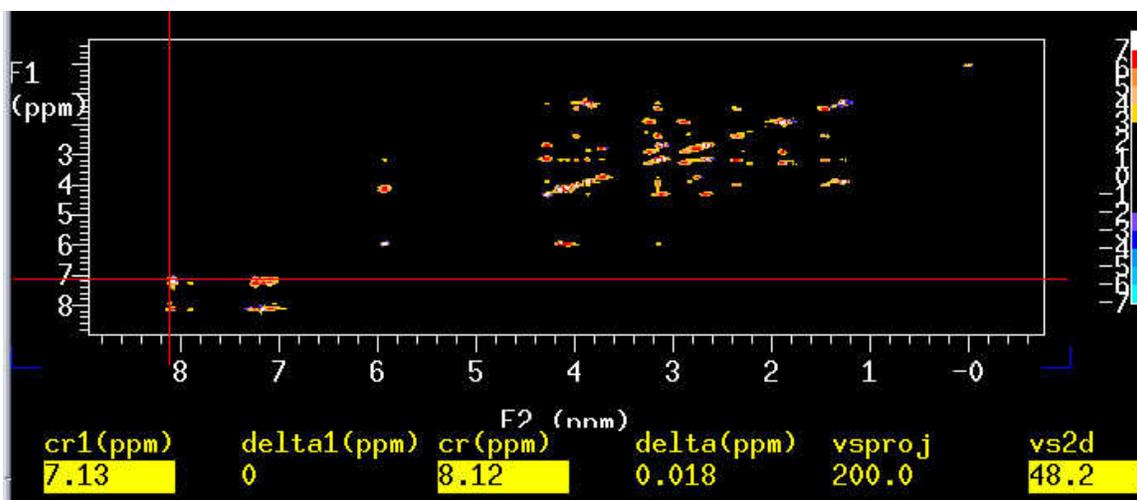
You should see the 2D like this.

29. ***Re-referencing the spectrum***. Point the mouse at the CDCl3 diagonal peak and type **rl(7.27p) rl1(7.27p).**
30. **pcon(10,1.2) page↵**
31. Print with 1D on side with 1D in experiment 1: **plcosy(10,1.2,1).**