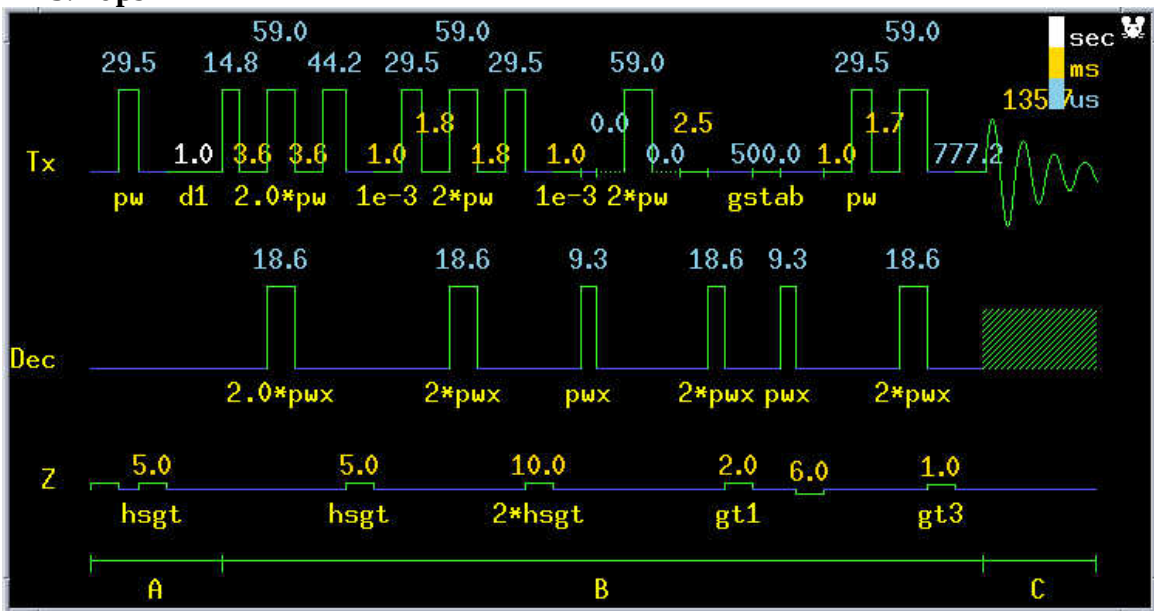
gHSQC experiment

1. Run a 1D spectrum for 1H and also 13C and array **pw90** at the desired **tpwr**. You need to know **T1** and **T2** of this molecule.
2. Set cursors ~0.5 ppm beyond last proton resonance on both sides of spectrum, type command: **movesw**.
3. **ga mf(1,3) jexp3**.
4. **gSHQC**
5. **dps**

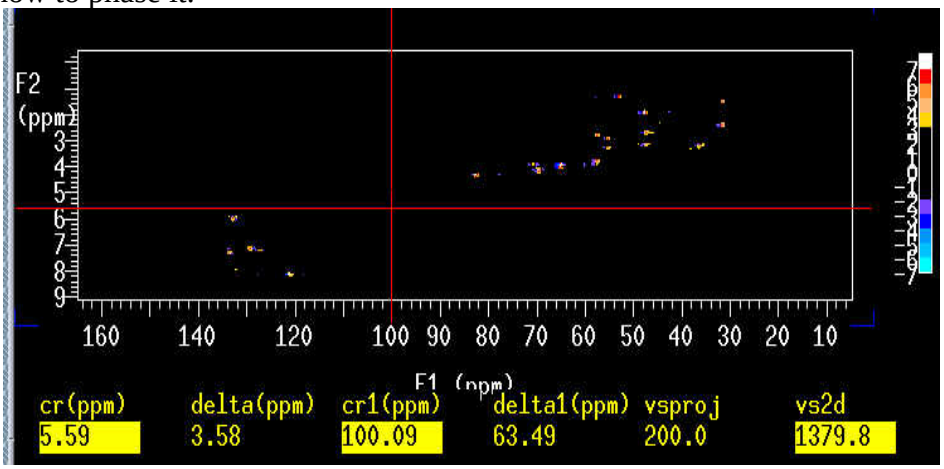


6. **dg you will see the following parameters.**

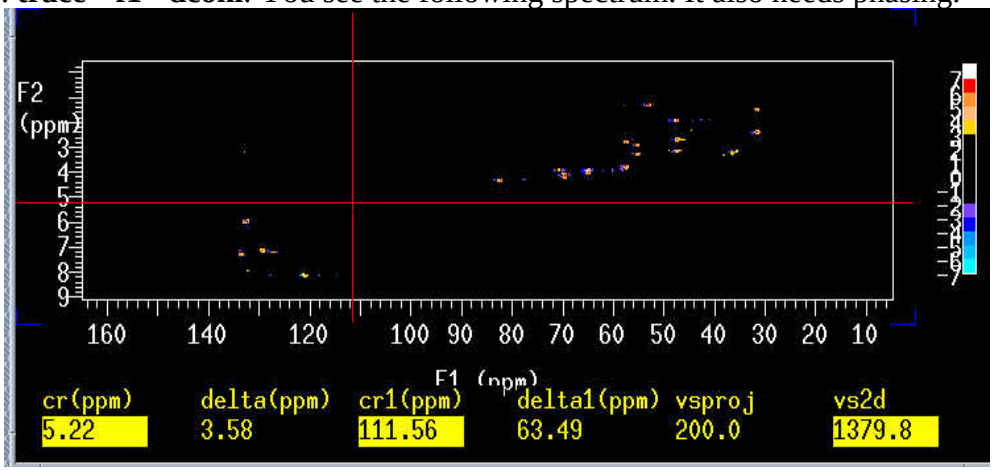
ACQUISITION		TRANSMITTER		HSQC		PROCESSING	
sw	3773.6	tn	H1	j1xh	140.0	gf	0.063
at	0.136	sfrq	399.729	nullflg	y	gfs	not used
np	1024	tof	-308.2	GRADIENTS		fn	1024
ss	8	tpwr	57	gzlvl1	4854	2D PROCESSING	
d1	1.000	pw	29.500	gt1	0.002000	gf1	0.034
nt	2	DECOUPLER		gzlvl3	2427	gfs1	not used
ct	2	dn	C13	gt3	0.001000	proc1	lp
2D ACQUISITION		dof	-1470.2	gstab	0.000500	fn1	2048
sw1	16080.4	pwxlvl	57	hsglvl	4854	pmode	full
ni	128	pwx	9.300	hsgt	0.005000	SAMPLE	
phase	arrayed	dm	nny	SPECIAL		date	Apr 7 2004
		dmm	ccw	temp	not used	solvent	CDCl3
		dmf	18519	spin	not used	sample	undefined
		dpwr	41	gain	30		
				sspul	y		
				pw90	29.500		

7. **nt=2**. (**nt** might need to be increased for sufficient signal-to-noise, generally nt = 2 is sufficient if decent 1H spectrum can be acquired in 32 scans)
8. **ni=128**, the number of t1 increments which should be multiple of 2.
9. **np=2048** or 1024, number of points in t2 dimension, must be even number. This will also set up **at**. **at=np/(2*sw). [note, at1=ni/(2*sw1)].**

10. **d1=1**, relaxation delay which is **5T1**(T1 for strychnine is 200 ms).It is normally should be slightly longer than in gCOSY.
11. turn off spin. open Acqi window and manually turn it off.
12. **sw1** is set to the default value which is the spectral width in 13C dimension(160 *100= 16000 at 400 MHz machine).
13. **dof** is the center of the carbon spectrum which is like **tof** in 1H spectrum. Use the default.
14. **j1xh=140** a default value. Aromatic 1-bond 1H/13C coupling constants are 170-250 Hz, so j1xh = 180.
15. **go**. At the end of the experiment. Save the fid by typing **svf('cungen').** This is really important. You can re-load you original data at any time if you want to start from beginning.
16. if you do not do any weighting function, you can type pmode='full' wft2da and you see the following spectrum. It needs to be phased on F1. You will see later how to phase it.

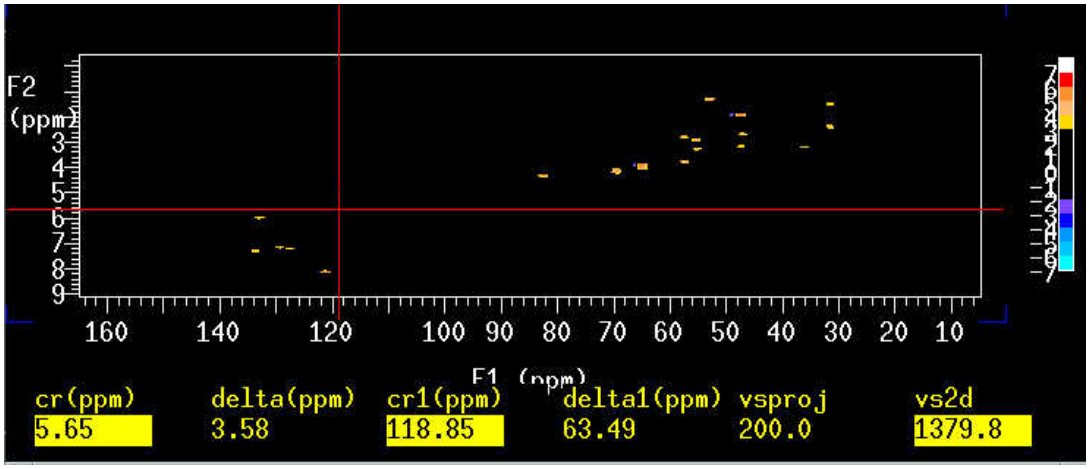


17. If you want to set appropriate weighting functions and linear prediction parameters, you need to re-load the file and you can type commands: **setLP1 sqcosine wft2da**
18. **f full dconi.**
19. **trace='f1' dconi**. You see the following spectrum. It also needs phasing.

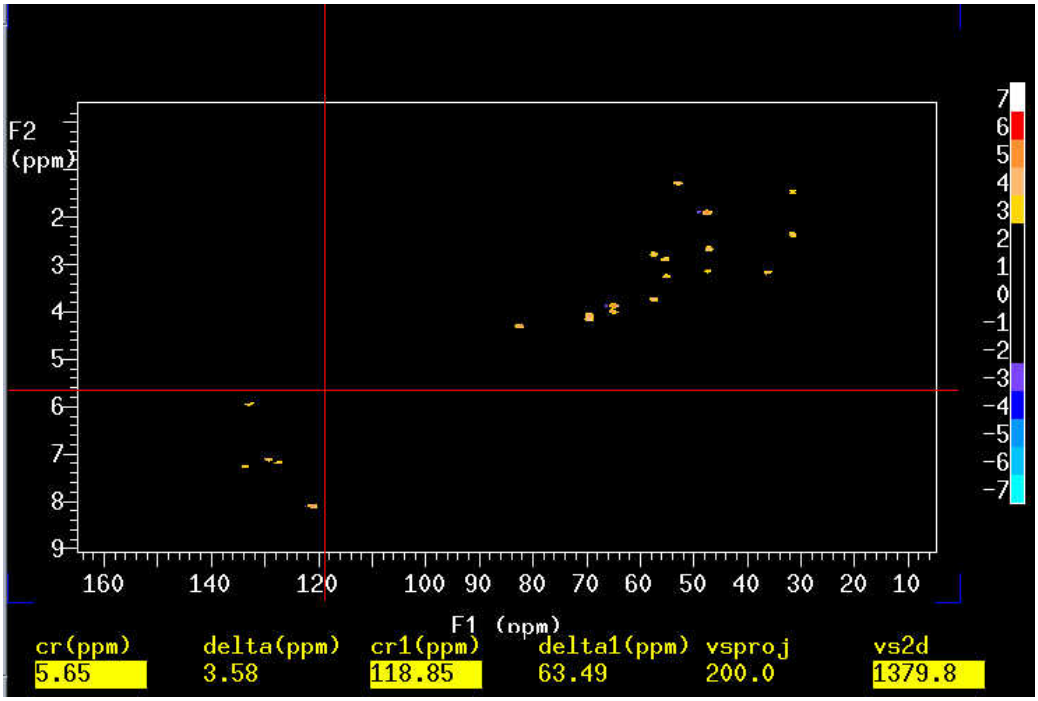


20. Reload the the FID and type the following commands:

21. **pmode='full'**, If **fn** parameter now equals 4096, processing will be slow, then type: **fn=np fn1=fn**.
22. **setLP1**.
23. **sb=-at sbs=sb sb1=-ni/sw1 sbs1=sb1**.
24. **wft2da**
25. **f full**
26. **trace='f1' dconi**.



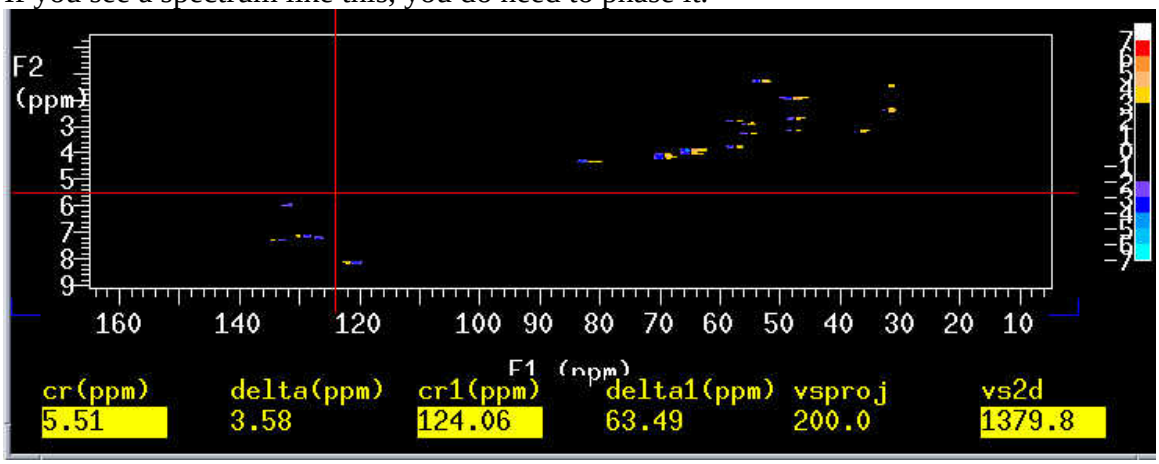
27. To lengthen the F2 axis, use the mouse to pull down the window, then type **dconi**.



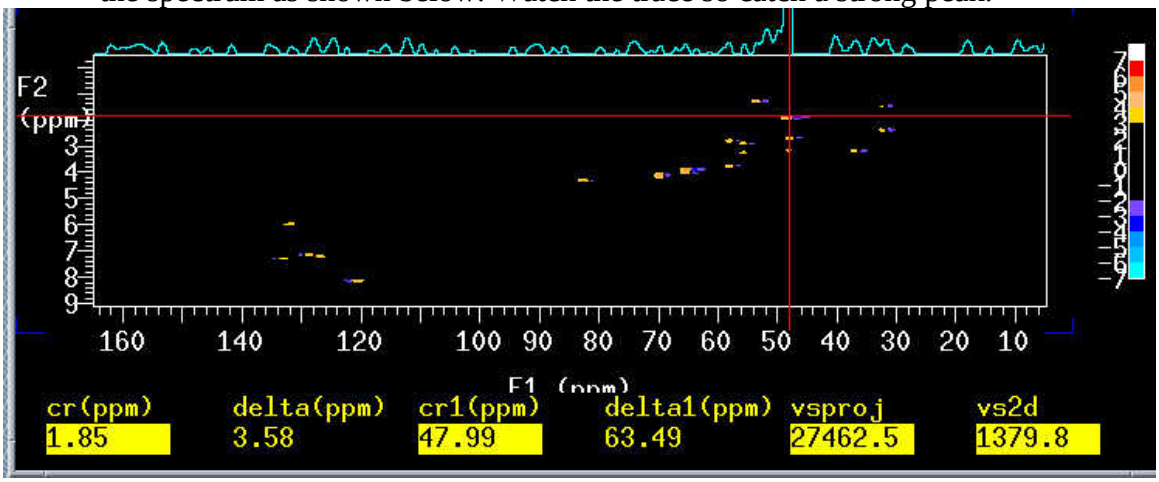
Now you got the decent spectrum. You may have trouble in some other experiments. So you need to learn how to phase 2D.

Phase a 2D spectrum

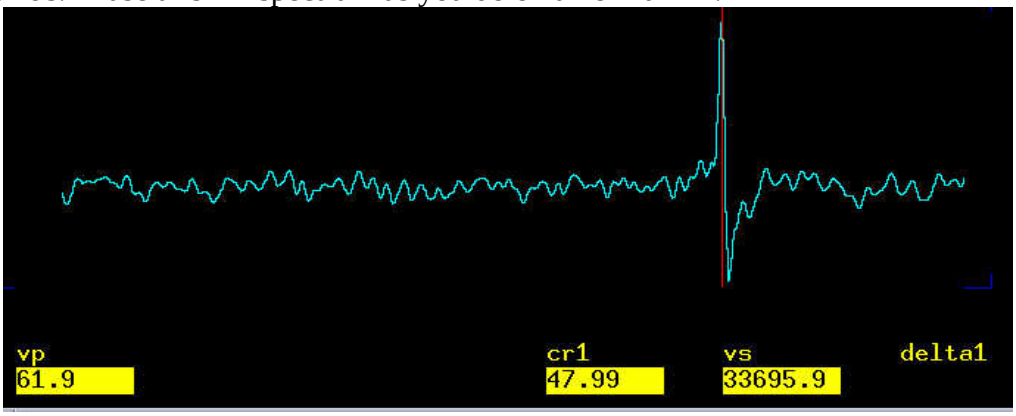
If you see a spectrum like this, you do need to phase it.



1. You have already typed **pmode='full' before wft2da**.
2. Click on ***trace*** button on top and then move the cursor to the right-upper peak in the spectrum as shown below. Watch the trace so catch a strong peak.

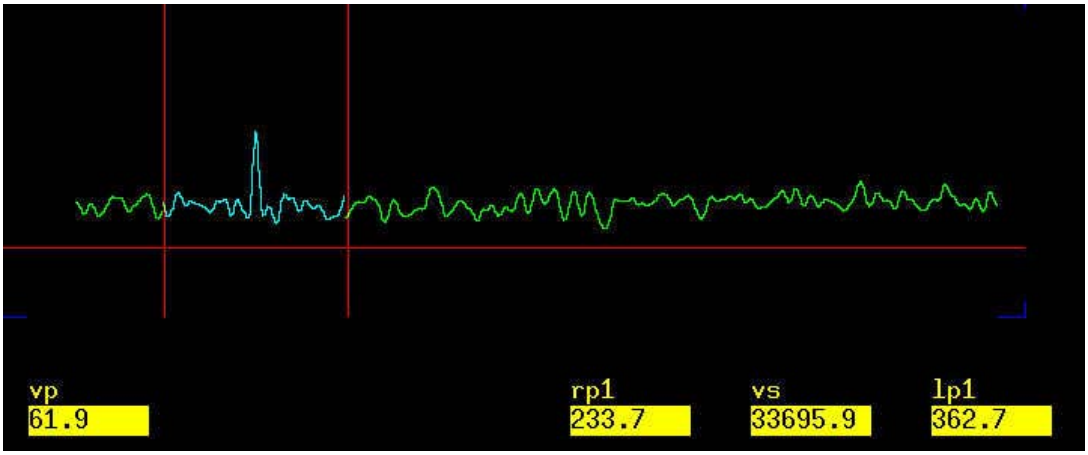


3. **ds**. Phase this 1D spectrum as you do on a normal 1D.

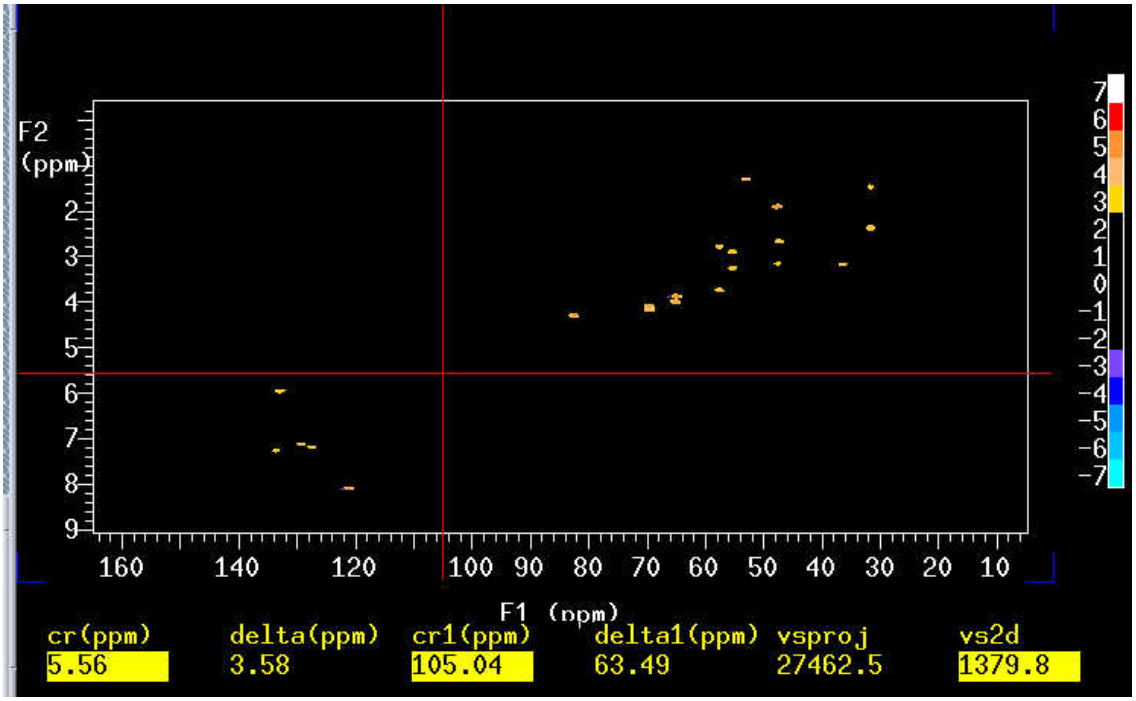


4. dconi. You will see that the streaks are gone on the right part.
5. Click trace button again and move the cursor to the lower left peak.

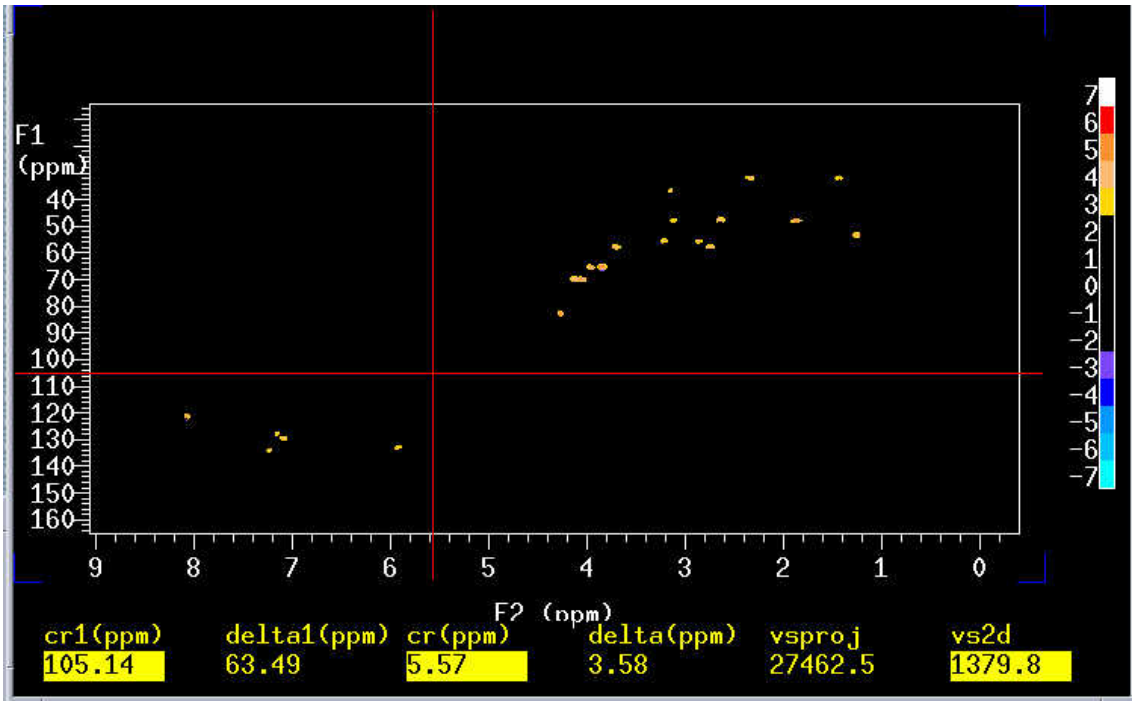
6. ds. Now click on the right of the spectrum without adjusting to keep the right phase value you got. This is zero-order phase. Move the mouse to the peak and adjust the base-line. This is the first-order phase.



7. **dconi**.



8. to change the spectrum axis, **trace='f2' dconi**.



Sometimes, you might need to phase the spectrum on F2. Do it as you did on F1.

9. Spectrum should be appropriately referenced already, but you should confirm this; if necessary re-reference by putting cursor on appropriate diagonal peak and type (assuming CDCl3): **rl(7.26p) rl1(77d).**
10. Adjust vertical scale with ***vs +20%*** and ***vs -20%*** menu buttons or with middle mouse button or manually changing the parameter, vs2d, so vs2d = 100 (the lower the number, the less noise displayed).
11. **svf('I_Love_HSQC')**.

Note: g13C-HSQC is not so common as g15N-HSQC. You are supposed to practice on gCHSQC before doing gNHSQC for protein NMR.

For small organic molecules, HMQC is widely used.