

Set up 2D/3D using Xwinnmr

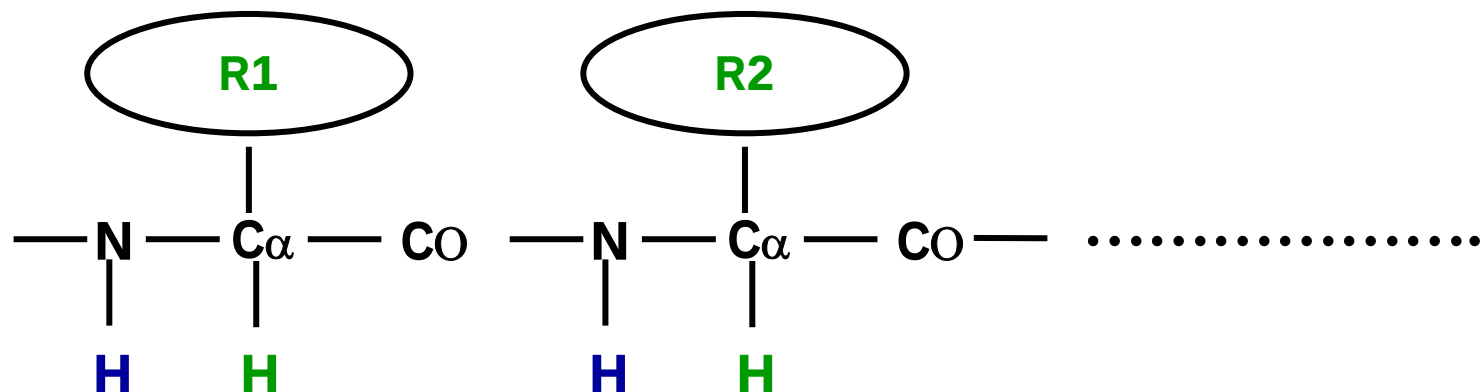
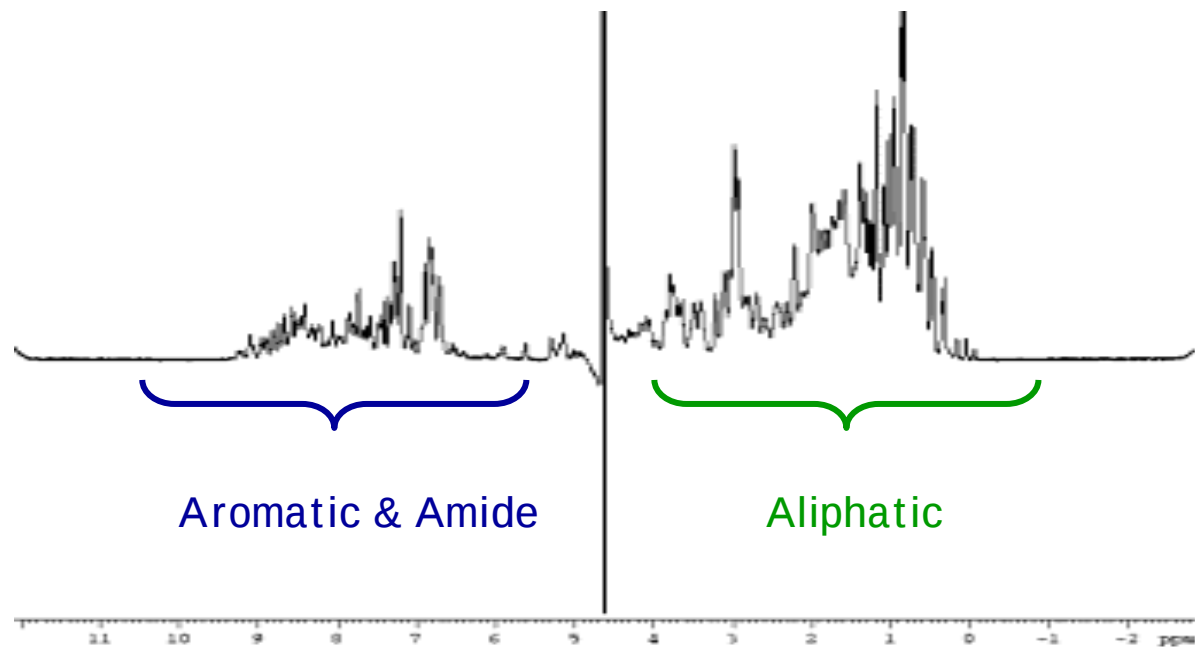
Chi-Fon Chang
March 30, 2005

General Concept for 2D/3D experiment

(I) Why bother multi-dimension?

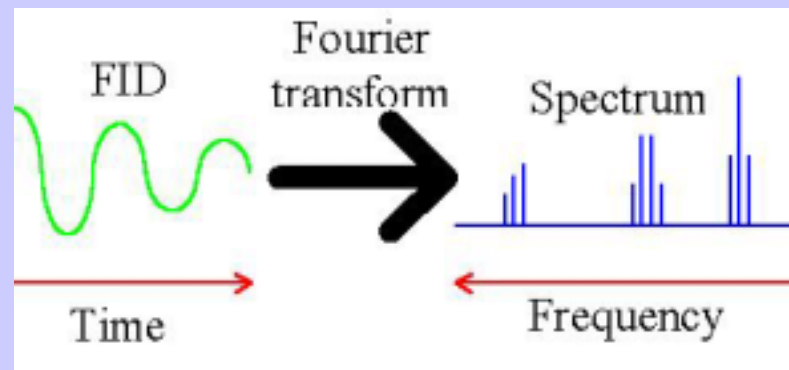
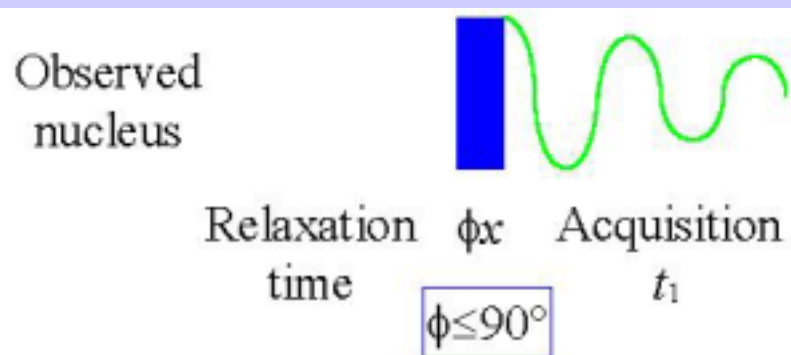
Homo Nuclear 1D NMR

1D one pulse 1H

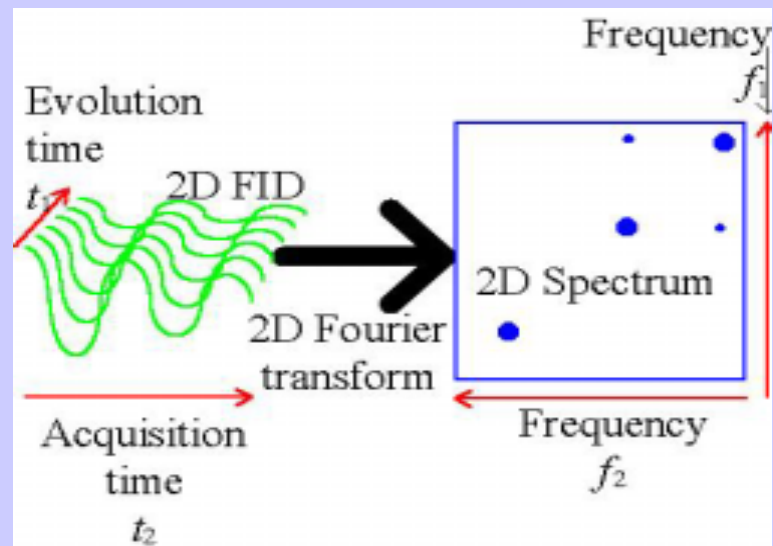
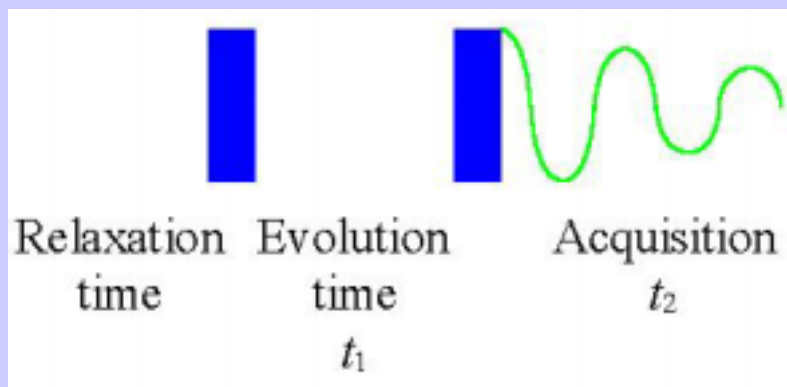


Homo/Hetro Nuclear 2D NMR

Basic 1D Experiment

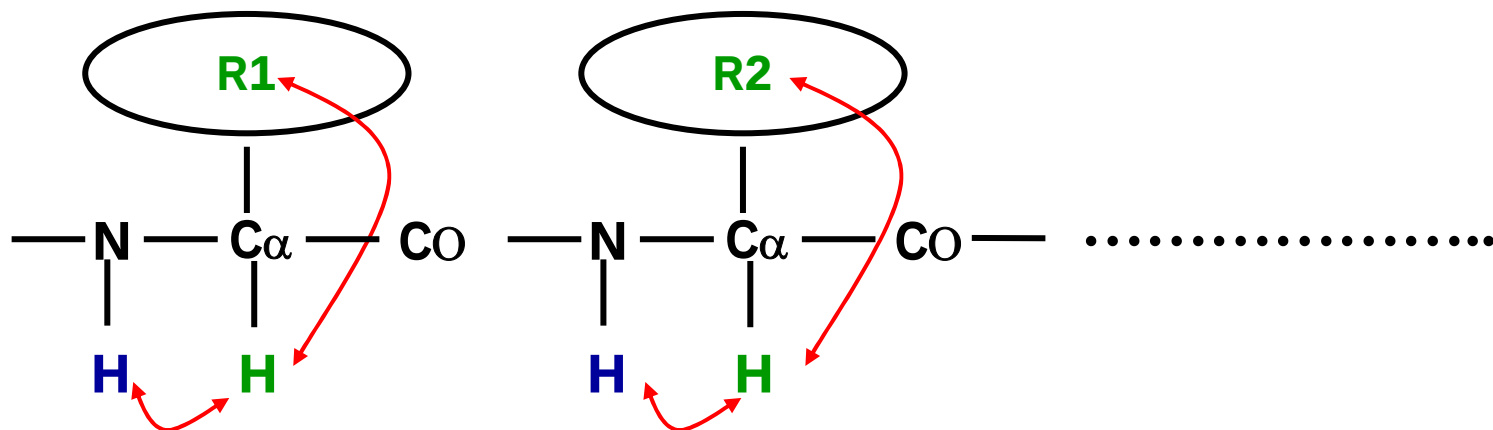
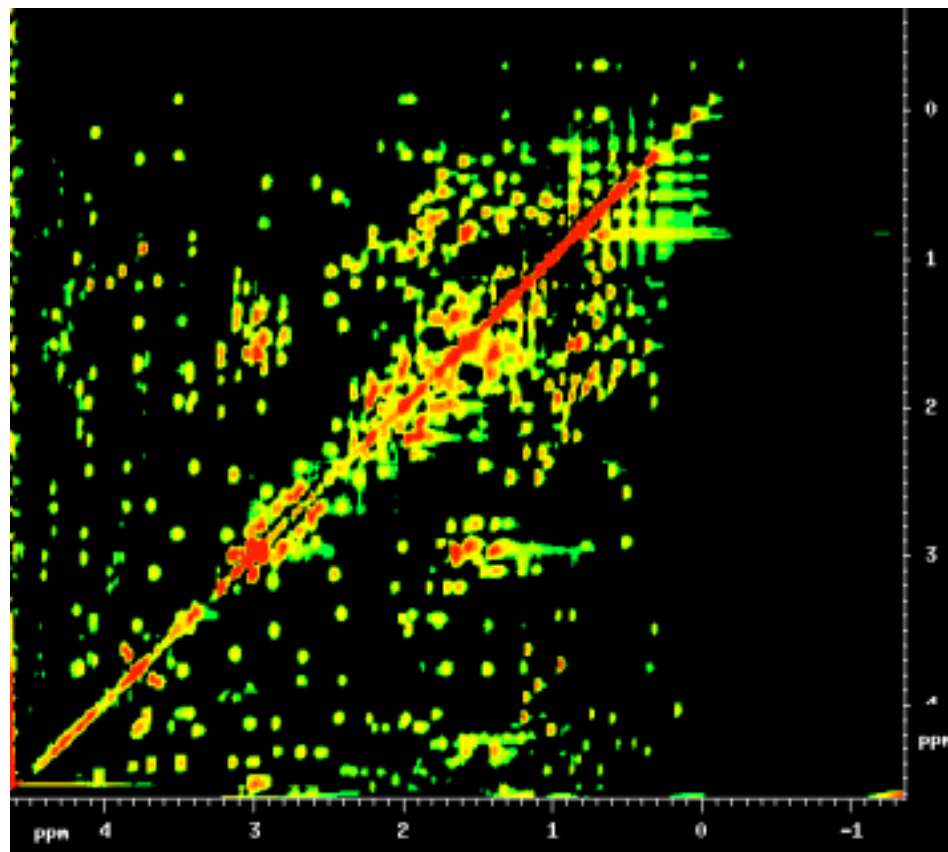
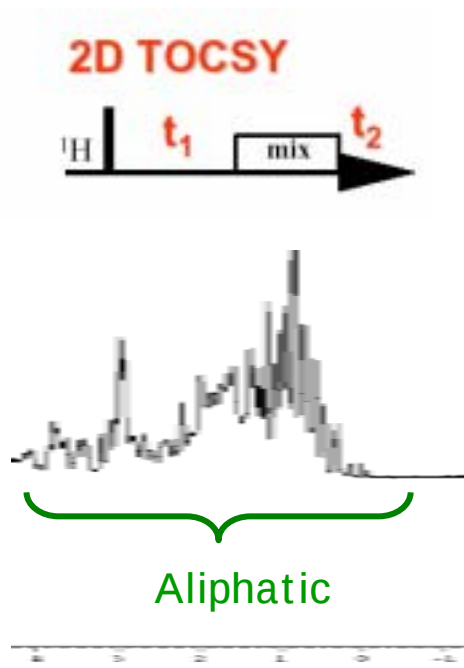


Basic 2D Experiment



2D Homo Nuclear

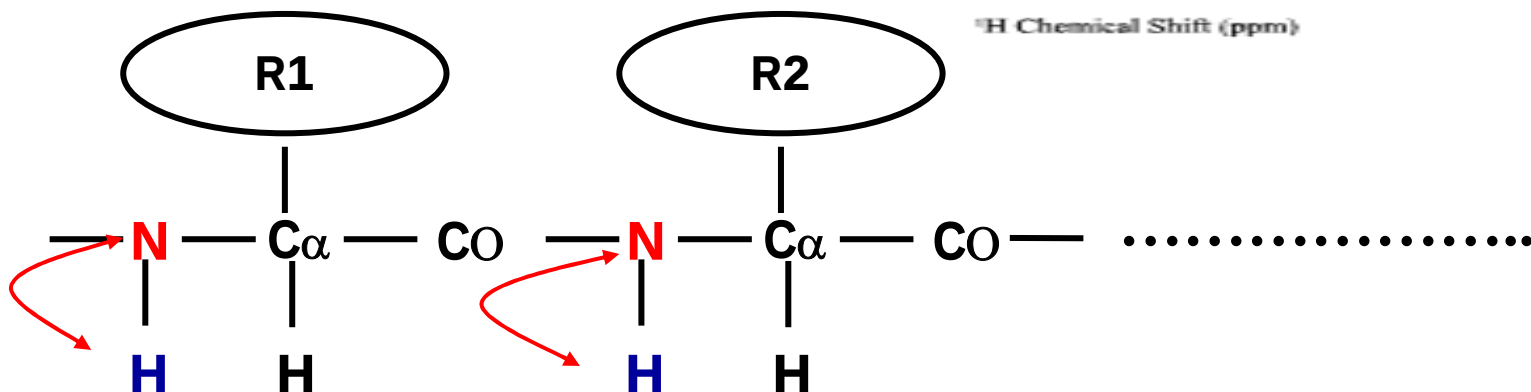
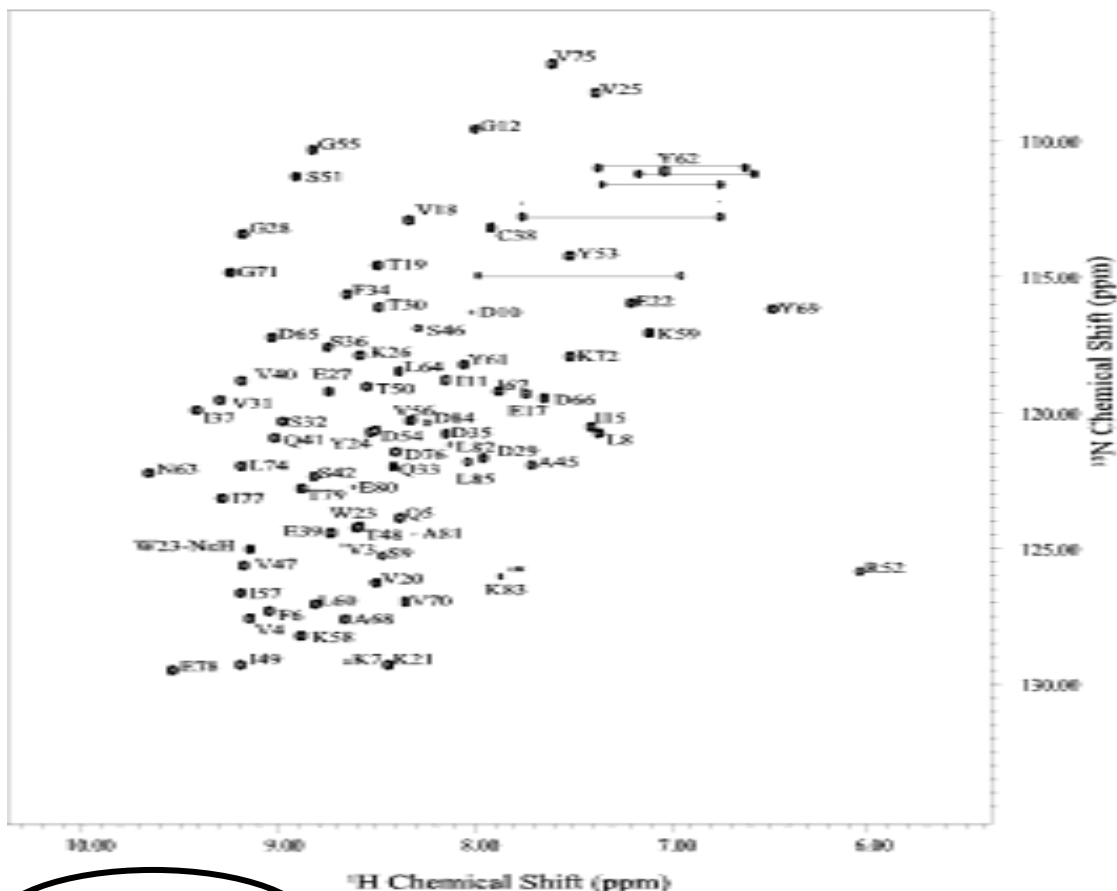
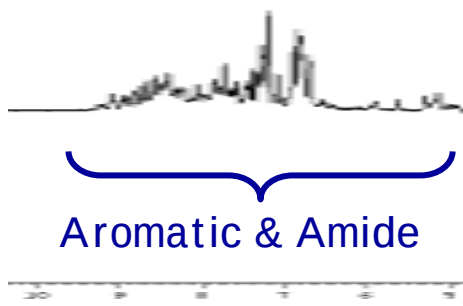
^1H - ^1H



2D Hetero Nuclear

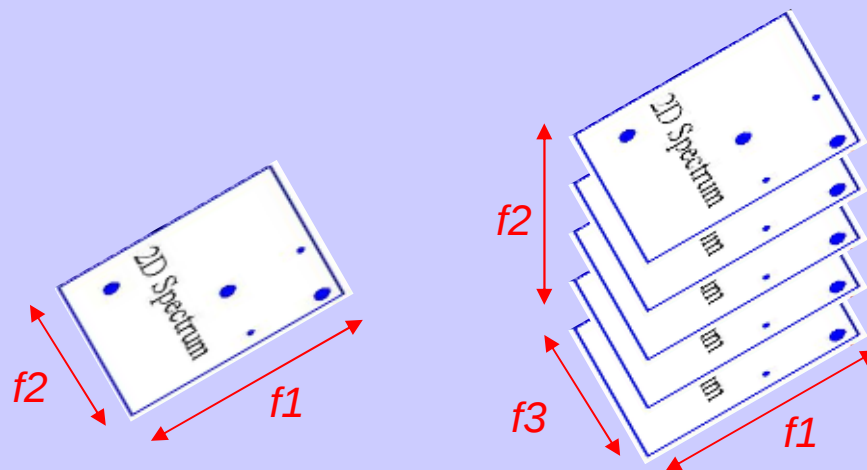
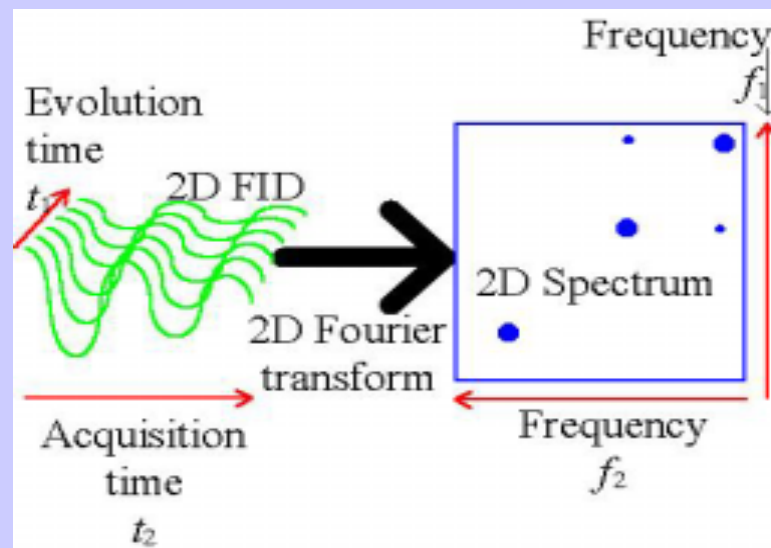
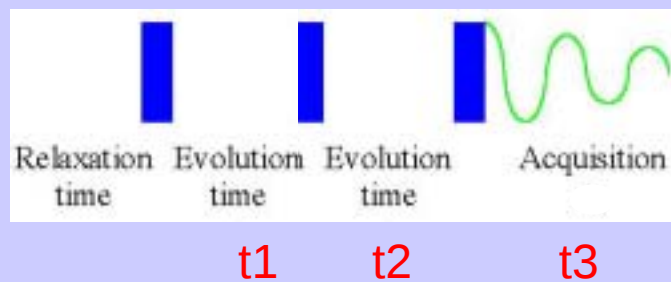
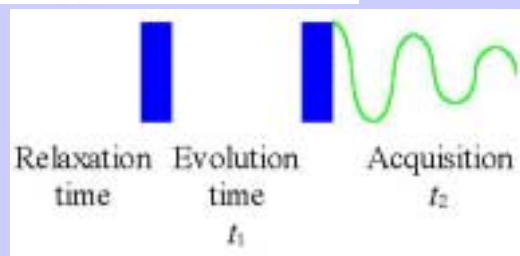
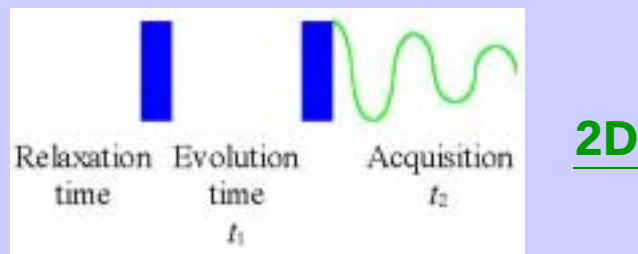
1H-15N

2D HSQC



Multi-Dimensional NMR

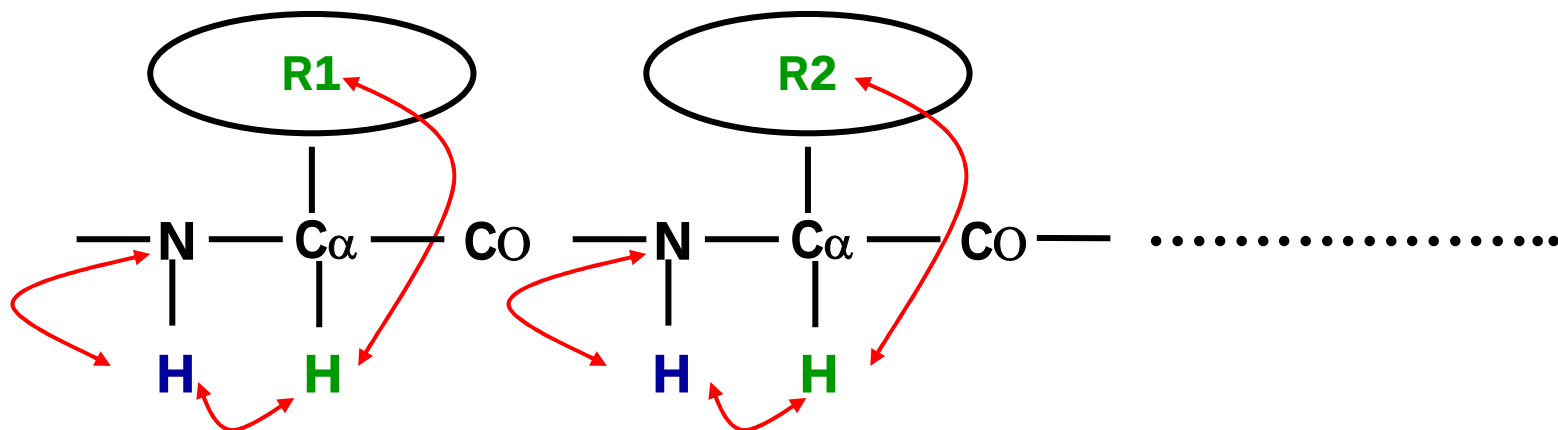
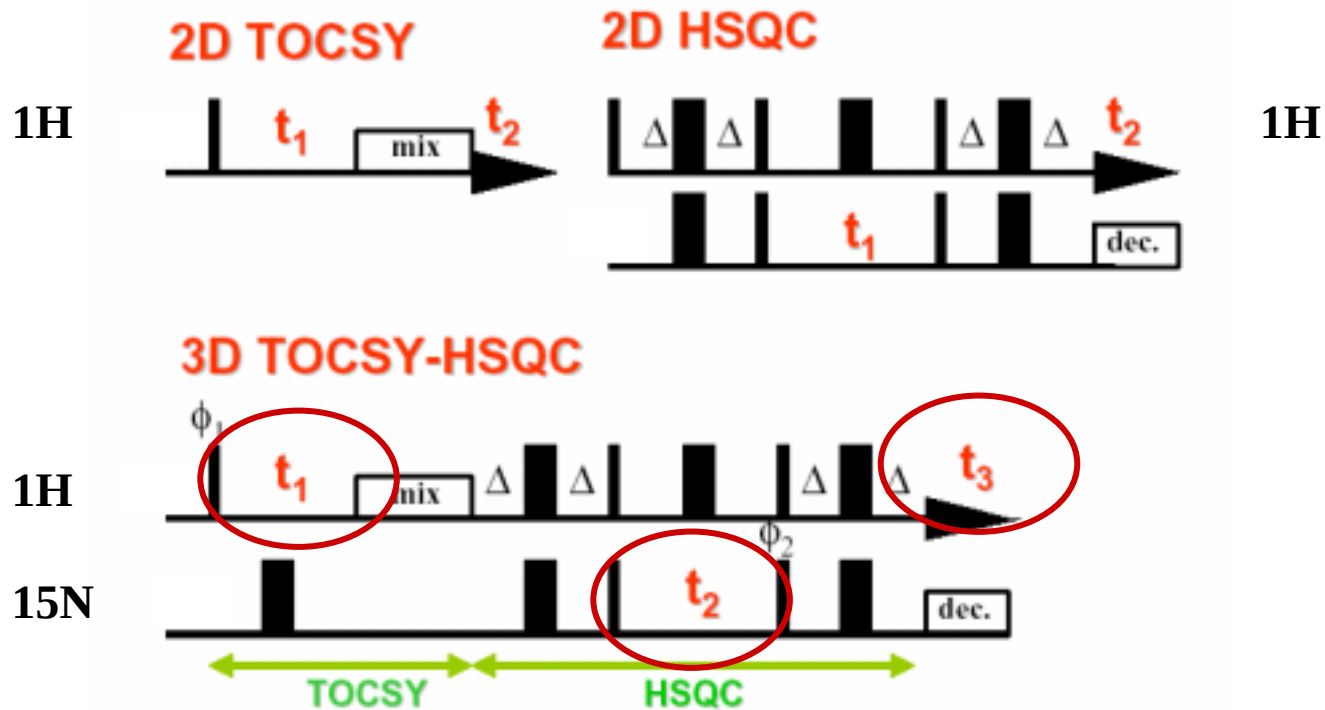
Basic 3D Experiment



3D Hetero Nuclear

^1H - ^{15}N - ^1H

Fig. 1. The 3D pulse sequence.

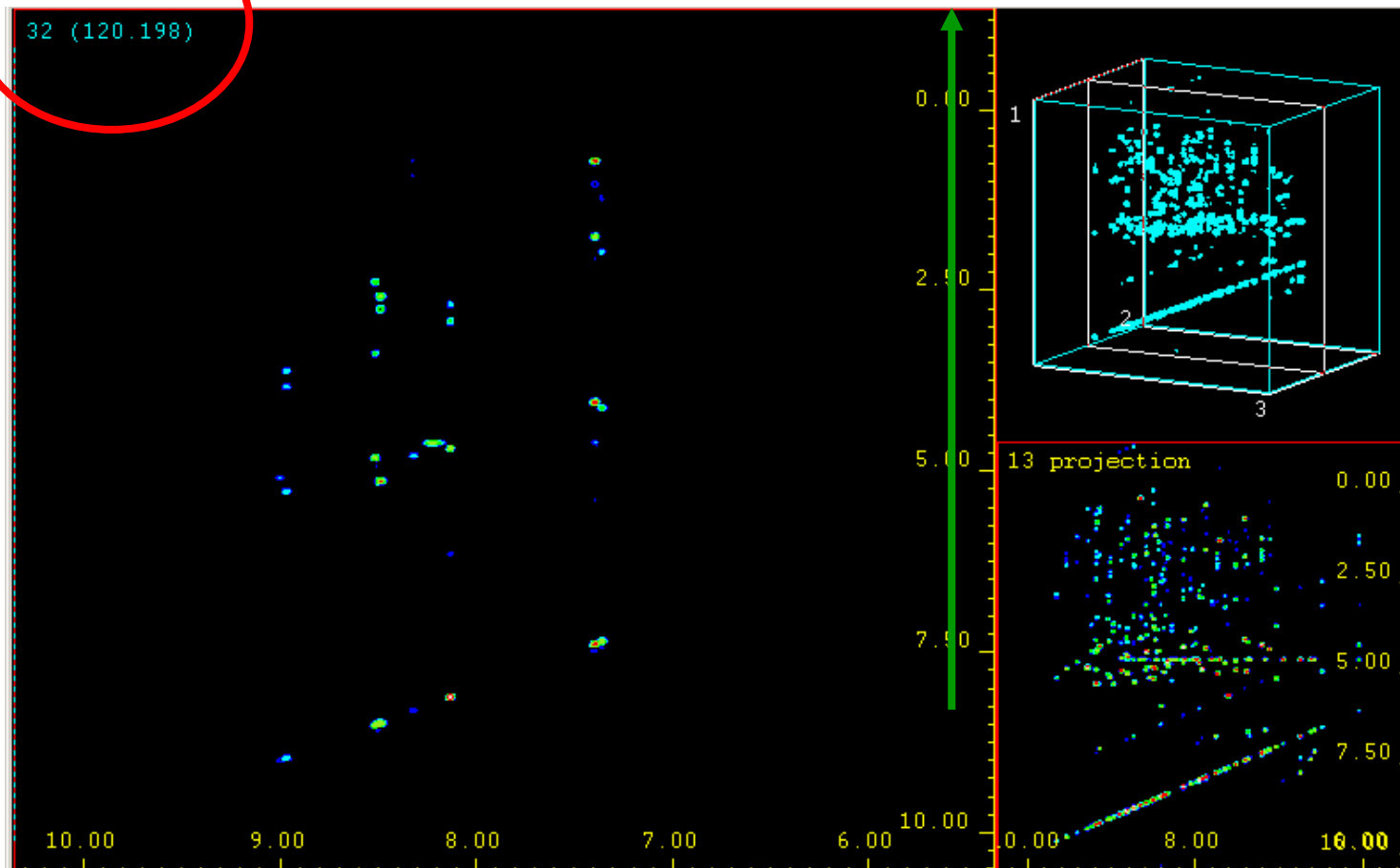


3D 15N-edit TOCSY-HSQC

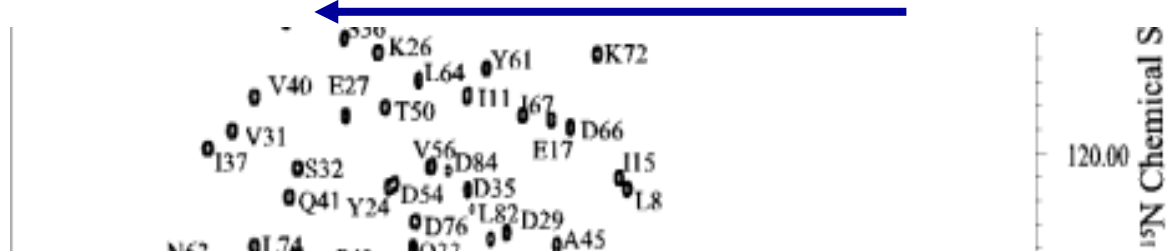
15N

32 (120.198)

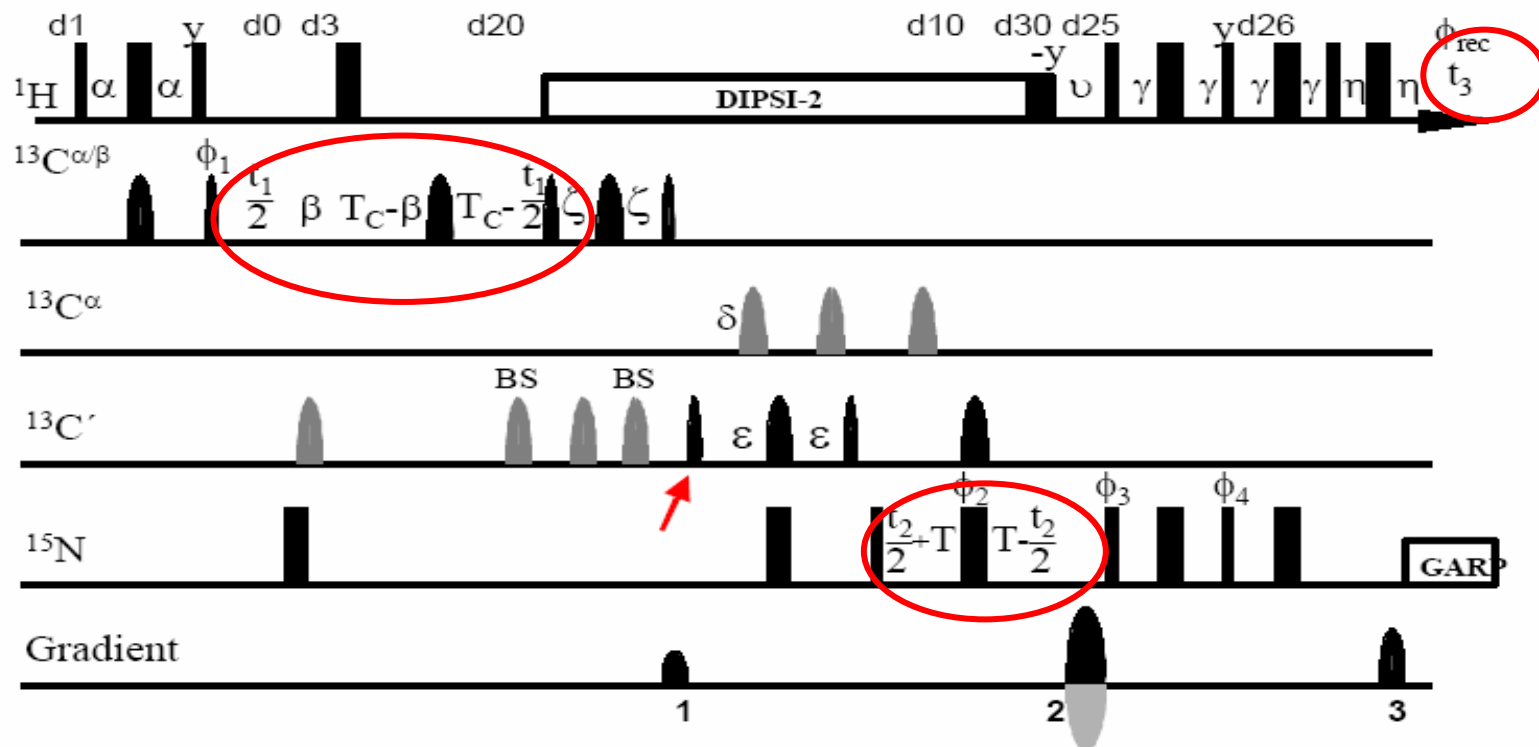
1H-all



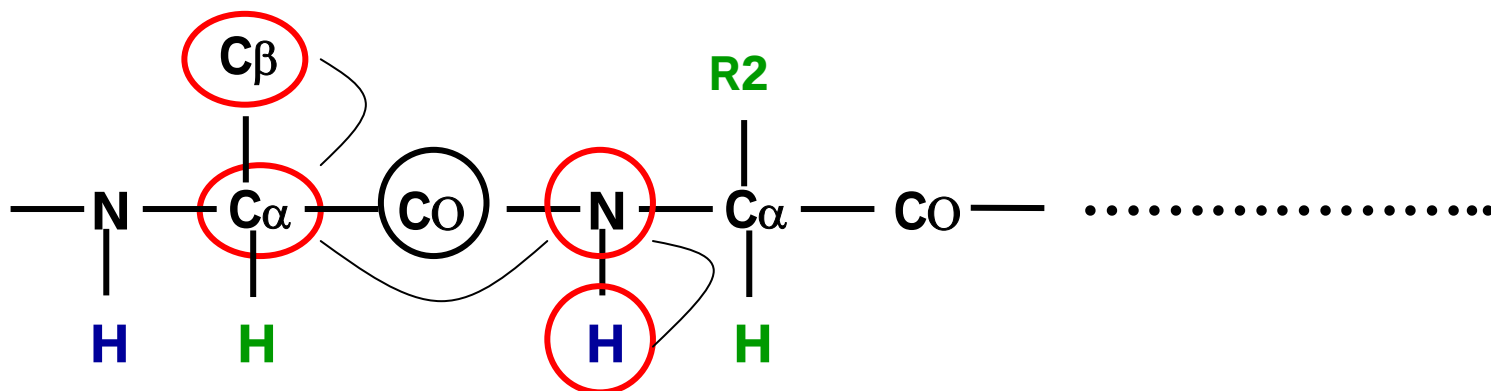
1H-NH



3D Hetero Nuclear 1H-15N-13C



$^1\text{H}^{\beta/\alpha} \xrightarrow{J_{\text{CH}}} ^{13}\text{C}^{\beta/\alpha}(t_1) \xrightarrow{J_{\text{C}\alpha\text{C}\beta}} ^{13}\text{C}^{\alpha} \xrightarrow{J_{\text{C}\alpha\text{C}'}} ^{13}\text{C}' \xrightarrow{J_{\text{C}'\text{N}}} ^{15}\text{N}(t_2) \xrightarrow{J_{\text{NH}}} ^1\text{H}(t_3)$

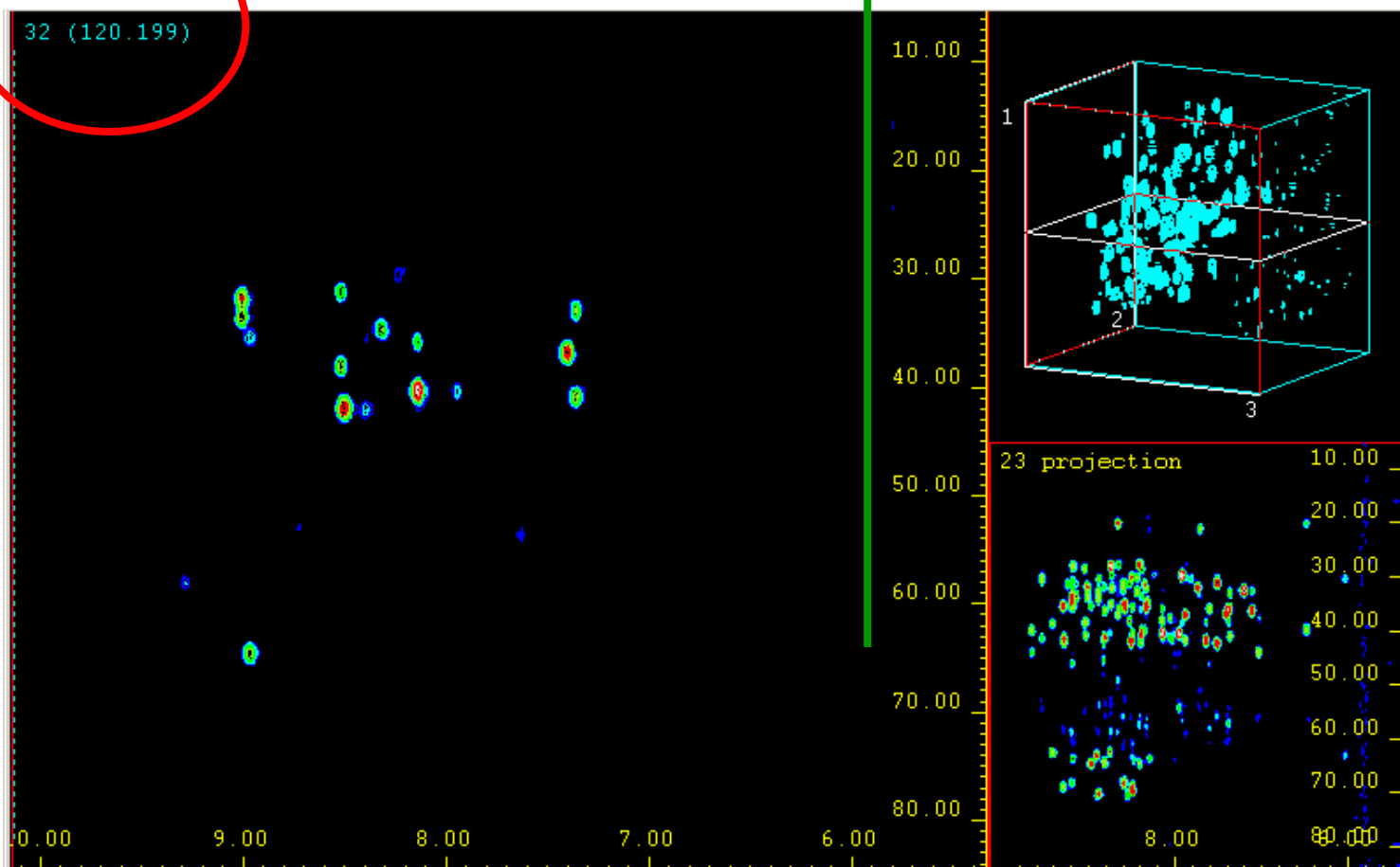


3D CBCA(CO)NH

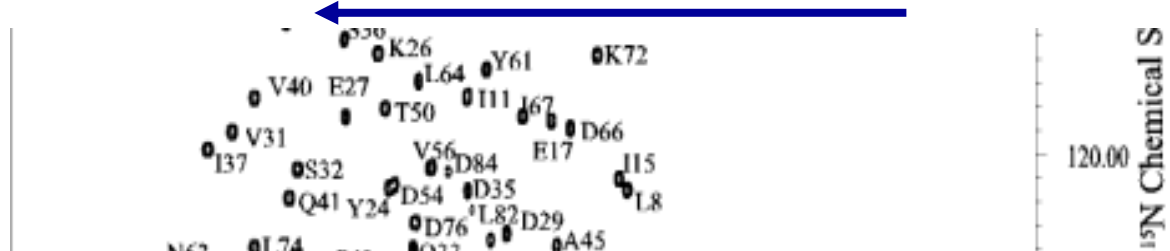
15N

32 (120.199)

13Ca & Cb



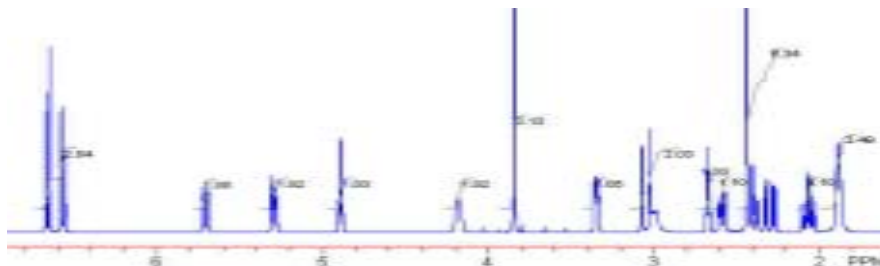
1H-NH



General Concept for 2D/3D experiment

(II) Set up using Xwinnmr

1D NMR

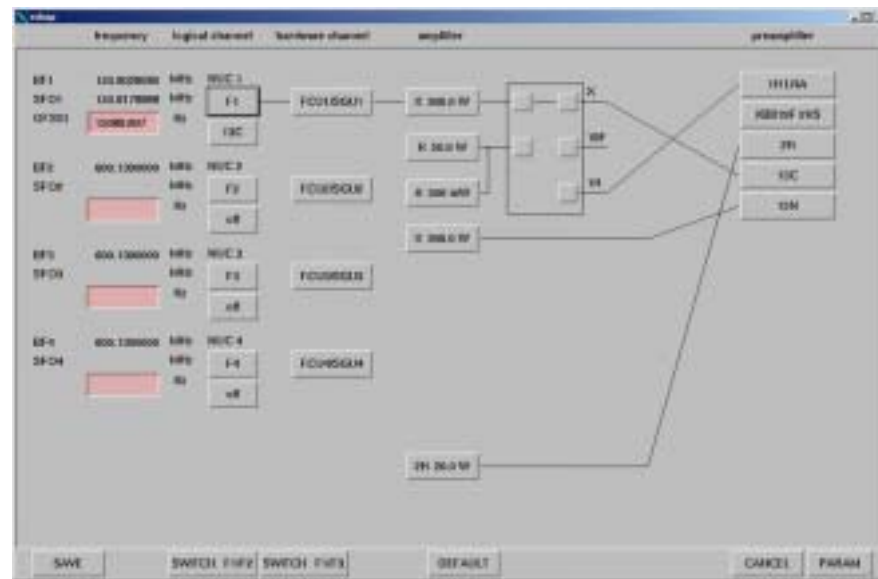
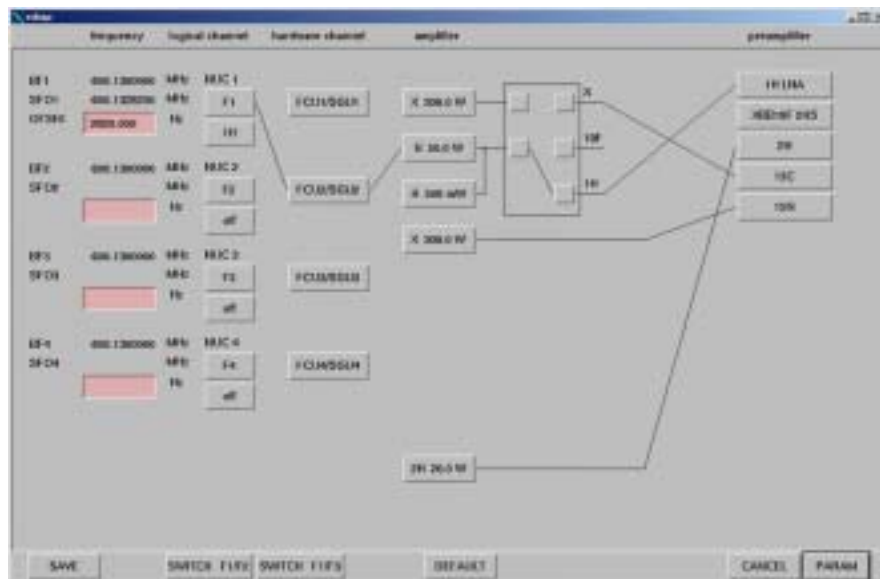


F1 dimension !!!

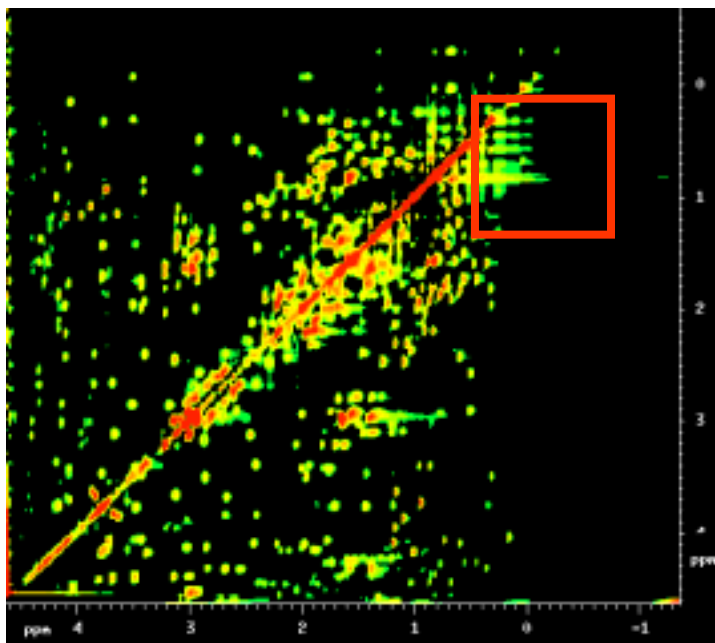
center (**o1**) at 4.5ppm

sw=6ppm

1.5ppm (=o1-sw/2) to 7.5ppm(=o1+sw/2)



2D NMR (homo)



F2 dimension !!!

F2: H (f1 channel)

O1(f1 channel): 1.8ppm (4.7)

2sw (F2 dimension): 6ppm (14)

F1: H (f1 channel)

O1(f1 channel): 1.8ppm (4.7)

1sw (F1 dimension): 6ppm (14)

edac

Acquisition Parameters

F2 F1

PULPROG noesyspph19

RG_mod 000

FnMODE States-TPPI

TD 2048 256

edac

Acquisition Parameters

F2 F1

NUCLEI edit edit

NUC1 1H 1H

O1 2814.00 Hz

SFO1 600.1328140 600.1328140 MHz

O1P 4.689 ppm

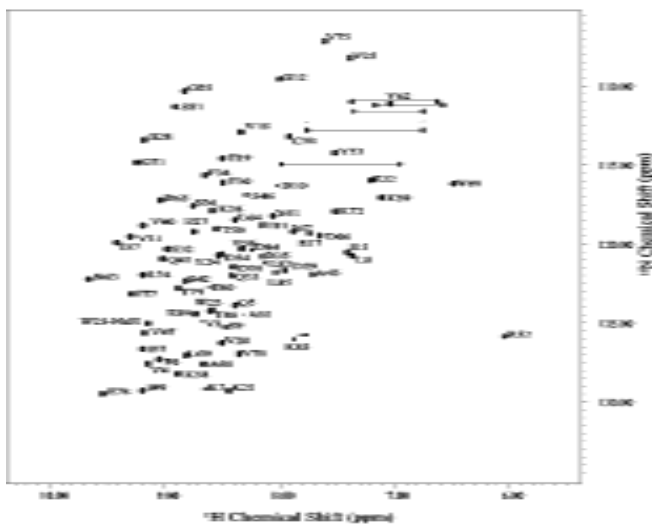
BF1 600.1300000 MHz

NUC2 off

edasp

	frequency	logical channel	hardware channel	amplifier
BF1	600.1300000 MHz	NUC 1		
SFO1	600.1328200 MHz	F1	FCU1/SGU1	X 300.0 W
OFSH1	2820.000 Hz	1H		H 50.0 W
BF2	600.1300000 MHz	NUC 2		
SFO2		F2	FCU2/SGU2	H 500 mW
		off		
BF3	600.1300000 MHz	NUC 3		
SFO3		F3	FCU3/SGU3	X 300.0 W
		off		

2D NMR (hetero)



F1 dimension !!!

F2 dimension !!!

F2: H (f1 channel)

O1(f1 channel): 8ppm (4.7)

2sw (F2 dimension): 5ppm (13)

F1: N (f3 channel)

O3(f3 channel): 120ppm (115)

1sw (F1 dimension): 40ppm (40)

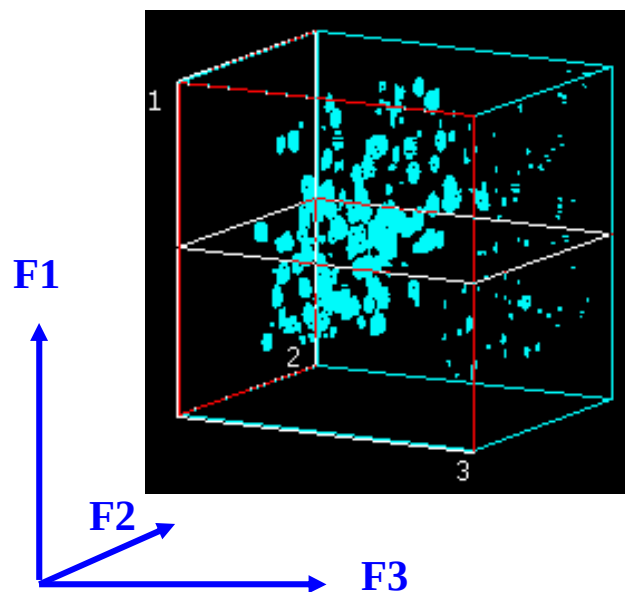
Two screenshots of Bruker software interfaces for NMR acquisition and hardware configuration.

Top Screenshot (edpar): Shows Acquisition Parameters for F2 and F1 channels. F2 is highlighted with a blue circle. Parameters include PULPROG (hsqcF3gpph19), AQ_mod (DQD), and F1MODE (States-TPPI).

Bottom Screenshot (xedasp): Shows hardware configuration for three channels (F1, F2, F3). The F1 channel is highlighted with a red box. The configuration includes frequency, logical channel, hardware channel, and amplifier settings.

	frequency	logical channel	hardware channel	amplifier
BF1	600.1300000 MHz	NUC 1	F1	FCU1/SGU1 — X 300.0 W
SFO1	600.1328200 MHz	1H		
OFSH1	2820.000 Hz			
BF2	150.9028090 MHz	NUC 2	F2	FCU2/SGU2 — H 50.0 W H 500 mW
SFO2	150.9106559 MHz	13C		
OFSX1	7846.946 Hz			
BF3	60.8106450 MHz	NUC 3	F3	FCU3/SGU3 — X 300.0 W
SFO3	60.8176382 MHz	15N		
OFSX2	6993.224 Hz			

3D NMR /triple



F3: H (f1 channel)

O1(f1 channel):4.7ppm

2 sw (F3 dimension): 14ppm

F2: N (f3 channel)

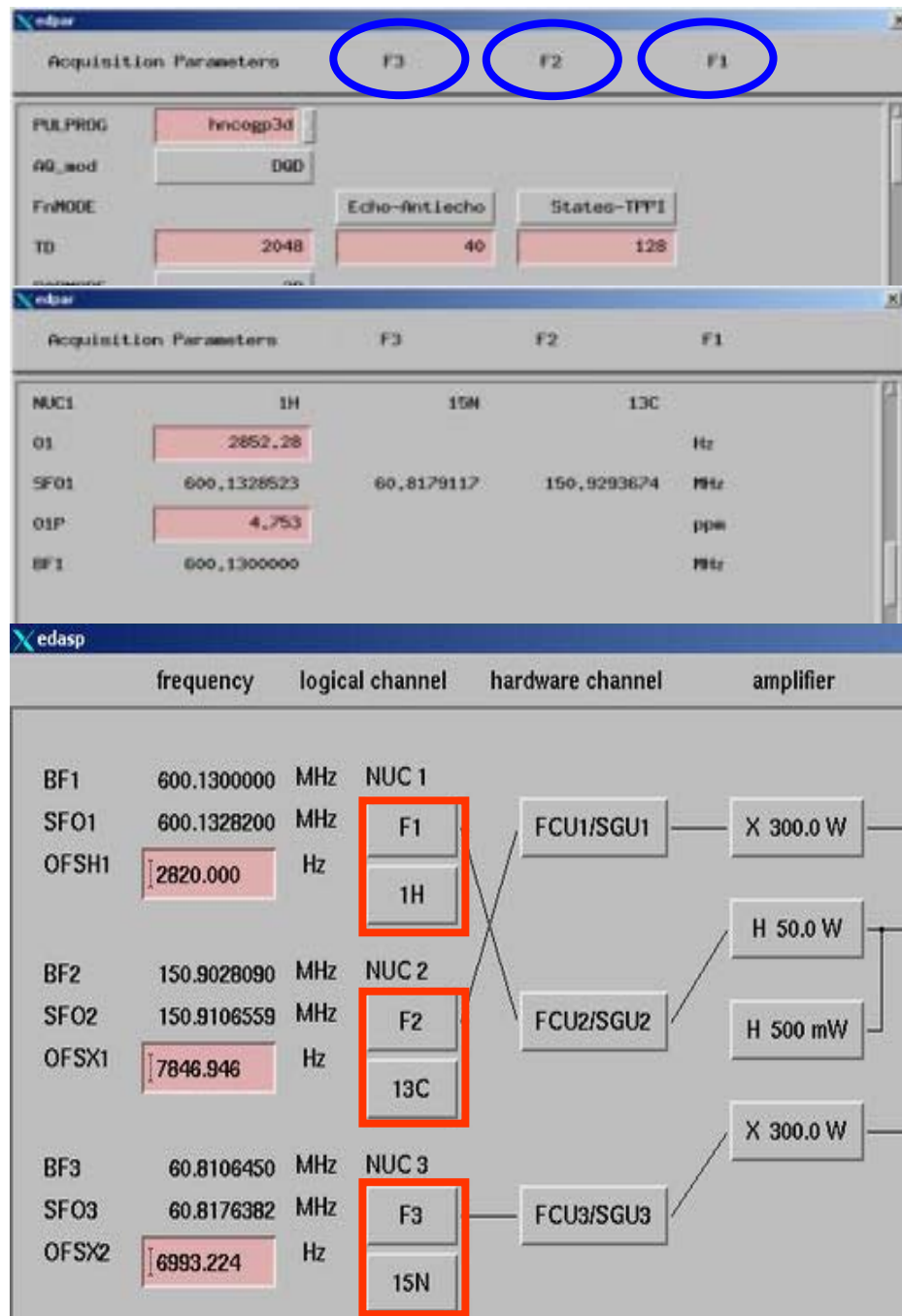
O3(f1 channel):120ppm

2 sw (F2 dimension): 35ppm

F1: C (f2 channel)

O2(f2 channel): 173ppm

1 sw (F1 diemnsion): 22ppm



Parameters in pulse sequence

(Bruker version)

How to set up experiment? (follow the guide)

- Experiment Name: **3D HNCO**
- Experiment Type: **Using Echo/antiecho , f1: H, f2:C, f3:N, F1(CO), F2(N), F3(H)**
- Standard Parameter Set: **std_3D_HNCO**
- Pulse Program: **hncogp3d**
- AQ parameters to check

1H pulses

p1 (high power, ex: 0db), **p1**(90deg at p1), **p2**(180deg at p1)

p19 (low power for dipsi2,pcpd1), **p26**(90deg at p19), **pcpd1**(90deg ,ex: 40-50usec)

sp1 (shape pulse power for Sinc.1000) , **p11**(pulse length for sp1, ex: 2m)

Others

o1 (for 1H), **o2** (for 13CO), **o3** (for 15N)

1 sw, 1 td (for F1 dimension, ie: 13C)

2 sw, 2 td (for F2 dimension, ie:15N)

3 sw, 3 td (for F3 dimension, ie:1H)

d1, rg, ns(=8*n), **ds** (16)

Users need to adjust parameters in “**red**” (meaning of the parameter in “**green**”)

What's going on? (inside the pulse program)

```
;hncogp3d  
;avance-version (02/05/31)  
;HNCO  
;3D sequence with  
;  inverse correlation for triple resonance using multiple  
;  inept transfer steps  
;  
; F1(H) -> F3(N) -> F2(C=O,t1) -> F3(N,t2) -> F1(H,t3)  
;  
;on/off resonance Ca and C=O pulses using shaped pulse  
;phase sensitive (t1)  
;phase sensitive using Echo/Antiecho gradient selection (t2)  
;using constant time in t2  
;(use parameterset HNCOGP3D)  
;  
;S. Grzesiek & A. Bax, J. Magn. Reson. 96, 432 - 440 (1992)  
;J. Schleucher, M. Sattler & C. Griesinger, Angew. Chem. Int. Ed. 32,  
; 1489-1491 (1993)  
;L.E. Kay, G.Y. Xu & T. Yamazaki, J. Magn. Reson. A109, 129-133 (1994)
```

Info & Ref.

```
prosol relations=<triple>
```

```
#include <Avance.incl>  
#include <Grad.incl>  
#include <Delay.incl>
```

Include files

**;;"d0=3u"
;;"d11=30m"
;;"d13=4u"**

**"d21=5.5m"
"d23=12m"
"d26=2.3m"**

**"in29=in10"
"in30=in10"**

**"d10=d23/2-p14/2"
"d29=d23/2-p14/2-p26-d21-4u"
"d30=d23/2-p14/2"**

**"DELTA=d0*2+larger(p14,p22)-p14"
"DELTA1=p16+d16+d13+4u"
"DELTA2=d23-d21-p26"
"DELTA3=d21-p16-d16-4u"**

**"spoff2=0"
"spoff3=0"
"spoff5=bf2*(cnst22/1000000)-o2"
"spoff8=0"**

Const. & Equ.

1 d11 ze
 d11 pl16:f3
 2 d11 do:f3
 3 d1 **pl1:f1**
 p1 ph1
 d26 **pl3:f3**
 (center (p2 ph1) (**p22 ph1**):f3)
 d26 UNBLKGRAD
 (p1 ph2):f1

 4u pl0:f1
 (p11:sp1 ph1:r):f1
 4u
 p16:gp1
 d16

 (p21 ph3):f3
 d21 **pl19:f1**
 (p26 ph2):f1
 DELTA2 **cpds1:f1** ph1
 (center (**p14:sp3 ph1**):f2 (p22 ph1):f3)
 d23
 (p21 ph1):f3

 d13 do:f1
 (p26 ph7):f1
 4u
 p16:gp2
 d16

Pulse sequence

(p13:sp2 ph4):f2

d0

(center **(p14:sp5 ph1):f2** (p22 ph1):f3)

d0

4u

(p14:sp3 ph1):f2

DELTA

(p14:sp5 ph1):f2

4u

(p13:sp8 ph1):f2

4u

p16:gp3

d16

(p26 ph2):f1

20u cpds1:f1 ph1

(p21 ph1):f3

d30

(p14:sp5 ph1):f2

d30

(center (p14:sp3 ph1):f2 (p22 ph8):f3)

d10

(p14:sp5 ph1):f2

d29

4u do:f1

(p26 ph7):f1

4u

p16:gp4*EA

d16

DELTA3 pl1:f1

Pulse sequence

```

(center (p1 ph1) (p21 ph5):f3 )
d26
(center (p2 ph1) (p22 ph1):f3 )
d26
(center (p1 ph2) (p21 ph6):f3 )
d26
(center (p2 ph1) (p22 ph1):f3 )
d26
(p1 ph1)
DELTA1
(p2 ph1)
d13
p16:gp5
d16 pl16:f3
4u BLKGRAD
go=2 ph31 cpd3:f3
d11 do:f3 mc #0 to 2
    F1PH(rd10 & rd29 & rd30 & ip4, id0)
    F2EA(igrad EA & ip6*2, id10 & id29 & dd30)
Exit

```

```

ph1=0
ph2=1
ph3=0 0 0 0 0 0 0 2 2 2 2 2 2 2
ph4=0 2
ph5=0 0 2 2
ph6=3 3 1 1
ph7=3
ph8=0 0 0 0 2 2 2 2
ph31=0 2 2 0 0 2 2 0 2 0 0 2 2 0 0 2

```

Pulse sequence

Phase

;pl0 : 120dB

;pl1 : f1 channel - power level for pulse (default)

;pl3 : f3 channel - power level for pulse (default)

;pl16: f3 channel - power level for CPD/BB decoupling

;pl19: f1 channel - power level for CPD/BB decoupling

;sp1: f1 channel - shaped pulse 90 degree (H2O on resonance)

;sp2: f2 channel - shaped pulse 90 degree (C=O on resonance)

;sp3: f2 channel - shaped pulse 180 degree (C=O on resonance)

;sp5: f2 channel - shaped pulse 180 degree (Ca off resonance)

;sp8: f2 channel - shaped pulse 90 degree (C=O on resonance)

; for time reversed pulse

;p1 : f1 channel - 90 degree high power pulse

;p2 : f1 channel - 180 degree high power pulse

;p11: f1 channel - 90 degree shaped pulse [2 msec]

;p13: f2 channel - 90 degree shaped pulse

;p14: f2 channel - 180 degree shaped pulse

;p16: homospoil/gradient pulse [1 msec]

;p21: f3 channel - 90 degree high power pulse

;p22: f3 channel - 180 degree high power pulse

;p26: f1 channel - 90 degree pulse at pl19

remarks

```

;d0 : incremented delay (F1 in 3D)                [3 usec]
;d1 : relaxation delay; 1-5 * T1
;d10: incremented delay (F2 in 3D) = d23/2-p14/2
;d11: delay for disk I/O                          [30 msec]
;d13: short delay                                [4 usec]
;d16: delay for homospoil/gradient recovery
;d21: 1/(2J(NH))                                [5.5 msec]
;d23: 1/(4J(NCO))                               [12 msec]
;d26: 1/(4J'(NH))                               [2.3 msec]
;d29: incremented delay (F2 in 3D) = d23/2-p14/2-p26-d21-4u
;d30: decremented delay (F2 in 3D) = d23/2-p14/2
;cnst21: CO chemical shift (offset, in ppm)
;cnst22: Calpha chemical shift (offset, in ppm)
;o2p: CO chemical shift (cnst21)
;in0: 1/(2 * SW(CO)) = DW(CO)
;nd0: 2
;in10: 1/(4 * SW(N)) = (1/2) DW(N)
;nd10: 4
;in29: = in10
;in30: = in10
;NS: 8 * n
;DS: >= 16
;td1: number of experiments in F1
;td2: number of experiments in F2      td2 max = 2 * d30 / in30
;FnMODE: States-TPPI (or TPPI) in F1
;FnMODE: echo-antiecho in F2

```

remarks

```
;cpds1: decoupling according to sequence defined by cpdprg1
;cpd3: decoupling according to sequence defined by cpdprg3
;pcpd1: f1 channel - 90 degree pulse for decoupling sequence
;pcpd3: f3 channel - 90 degree pulse for decoupling sequence
```

```
;use gradient ratio:  gp 1 : gp 2 : gp 3 : gp 4 : gp 5
;                    60 : -40 : 10 : 80 : 8.1
```

```
;for z-only gradients:
;gpz1: 60%
;gpz2: -40%
;gpz3: 10%
;gpz4: 80%
;gpz5: 8.1%
```

```
;use gradient files:
;gpnam1: SINE.100
;gpnam2: SINE.100
;gpnam3: SINE.100
;gpnam4: SINE.100
;gpnam5: SINE.100
```

remarks

How to optimize condition? (only when there's no standard set available)

Steps: new → edasp → eda (select pulse program) → getprosol → ased

The screenshot shows the 'Xedpar' window with 'Acquisition Parameters' for a 3D experiment. The parameters are organized into columns F3, F2, and F1. The 'PULPROG' is set to 'hncogp3d'. The 'FnMODE' is set to 'Echo-Antiecho' (highlighted with a blue box) and 'States-TPPI'. The 'TD' is 2048, 'PARMODE' is 3D, 'NS' is 8, 'DS' is 16, 'TD0' is 1, 'ZGPTNS' is '-DLABEL_CN', 'D' is '** Array **', 'P' is '** Array **', 'NDO' is 2 (highlighted with a blue box), 'INO' is 0.00015000, 'ND10' is 4 (highlighted with a blue box), 'IN10' is 0.00011420, and 'SW' is 13.9790, 35.9950, 22.0854 ppm.

Parameter	F3	F2	F1
PULPROG	hncogp3d		
AQ_mod	DQD		
FnMODE	Echo-Antiecho States-TPPI		
TD	2048	40	128
PARMODE	3D		
NS	8		
DS	16		
TD0	1		
ZGPTNS	-DLABEL_CN		
D	** Array **		
P	** Array **		
NDO	2		
INO	0.00015000		
ND10	4		
IN10	0.00011420		
SW	13.9790	35.9950	22.0854

- follow pulse program
- maybe modify
- $nd0$ = number of d0 in the increment loops of 2D/3D
- $in0 = 1/(nd0 * sw)$

The screenshot shows the 'Xedpar' window with 'Acquisition Parameters' for a 3D experiment. The parameters are organized into columns F3, F2, and F1. The 'NUC1' is 1H, 15N, 13C (highlighted with a blue box). The 'O1' is 2852.28 Hz. The 'SF01' is 600.1328523, 60.8179117, 150.9293674 MHz.

Parameter	F3	F2	F1
NUC1	1H	15N	13C
O1	2852.28		
SF01	600.1328523	60.8179117	150.9293674

Steps: new → edasp → eda (select pulse program) → **getprosol** → **ased**

edpar

F2 - Acquisition Parameters

PULPROG	hncogp3d		pulse program for acquisition
TD	2048		time domain size
NS	8		number of scans
DS	16		number of dummy scans
SWH	8389.26	Hz	sweep width in Hz
AQ	0.1221704	sec	acquisition time
RG	256		receiver gain
DW	59.600	usec	dwel time
DE	6.00	usec	pre-scan-delay
d0	0.00000300	sec	d0=3u
D1	1.00000000	sec	relaxation delay: 1-5 * T1
d10	0.00587200	sec	d10=d23/2-p14/2
d11	0.03000000	sec	d11=30m
d13	0.00000400	sec	d13=4u
D16	0.00020000	sec	delay for homospoil/gradient recovery
d21	0.00550000	sec	d21=5.5m

SAVE 2-COL Parameter Next CANCEL

• sample dependent

Steps: new → edasp → eda (select pulse program) → **getprosol** → **ased**

edpar

F2 - Acquisition Parameters

d23	0.01200000	sec	d23=12m
d26	0.00230000	sec	d26=2.3m
d29	0.00032800	sec	d29=d23/2-p14/2-p26-d21-4u
d30	0.00587200	sec	d30=d23/2-p14/2
DELTA	0.00000600	sec	DELTA=d0*2+larger(p14,p22)-p14
DELTA1	0.00120800	sec	DELTA1=p16+d16+d13+4u
DELTA2	0.00646000	sec	DELTA2=d23-d21-p26
DELTA3	0.00429600	sec	DELTA3=d21-p16-d16-4u
IN0	0.00015000	sec	$1/(2 * SW(C0)) = DW(C0)$
IN10	0.00011420	sec	$1/(4 * SW(N)) = (1/2) DW(N)$
in29	0.00011420	sec	in29=in10
in30	0.00011420	sec	in30=in10
MCREST	0.00000000	sec	MCREST = d11 - d11
MCWRK	0.00300000	sec	MCWRK = 0.100000*d11
ST1CNT	64		ST1CNT = td1 / (2)
ST2CNT	20		ST2CNT = td2 / (2)

SAVE 2-COL Parameter Next CANCEL

Steps: new → edasp → eda (select pulse program) → **getprosol** → **ased**

edpar

F2 - Acquisition Parameters

===== CHANNEL f1 =====

CPDPRG1	dipsi2	file name for cpd1
NUC1	1H	nucleus for channel 1
P1	9.45	f1 channel - 90 degree high power puls
P2	18.90	f1 channel - 180 degree high power puls
P11	1000.00	f1 channel - 90 degree shaped pulse
P26	40.00	f1 channel - 90 degree pulse at pl19
PCPD1	40.00	f1 channel - 90 degree pulse for decoup
PL0	120.00	120dB
PL1	-5.00	f1 channel - power level for pulse (def
PL19	7.53	f1 channel - power level for CPD/BB dec
SF01	600.1328523	frequency of observe channel
SP1	30.89	f1 channel - shaped pulse 90 degree (
SPNAM1	Sinc1.1000	file name for sp1
SPOAL1	0.500	phase alignment of freq. offset in SP1
SPOFF1	0.00	offset frequency for SP1

SAVE 2-COL Parameter Next CANCEL

• use standard

• sample dependent

Steps: new → edasp → eda (select pulse program) → **getprosol** → **ased**

edpar

F2 - Acquisition Parameters

===== CHANNEL f2 =====

CNST22	54.0000000		Calpha chemical shift (offset, in ppm)
NUC2	13C		nucleus for channel 2
P13	320.00	usec	f2 channel - 90 degree shaped pulse
P14	256.00	usec	f2 channel - 180 degree shaped pulse
PL2	-6.00	dB	power PL2
SF02	150.9293674	MHz	frequency of 2nd channel
SP2	-4.40	dB	f2 channel - shaped pulse 90 degree (
SP3	-3.65	dB	f2 channel - shaped pulse 180 degree (
SP5	-3.65	dB	f2 channel - shaped pulse 180 degree (
SP8	-4.40	dB	f2 channel - shaped pulse 90 degree (
SPNAM2	Q5.1000		file name for sp2
SPNAM3	Q3.1000		file name for sp3
SPNAM5	Q3.1000		file name for sp5
SPNAM8	Q5tr.1000		file name for sp8
SPOAL2	0.500		phase alignment of freq. offset in SP2

SAVE 2-COL Parameter Next CANCEL

• use standard

Steps: new → edasp → eda (select pulse program) → **getprosol** → **ased**

edpar

F2 - Acquisition Parameters

SP0AL5	0.500		phase alignment of freq. offset in SP5
SP0AL8	0.500		phase alignment of freq. offset in SP8
spoff2	0.00	Hz	spoff2=0
spoff3	0.00	Hz	spoff3=0
spoff5	-18409.70	Hz	spoff5=bf2*(cnst22/1000000)-o2
spoff8	0.00	Hz	spoff8=0
===== CHANNEL f3 =====			
CPDPRG3	garp		file name for cpd3
NUC3	15N		nucleus for channel 3
P21	38.80	usec	f3 channel - 90 degree high power puls
P22	77.60	usec	f3 channel - 180 degree high power puls
PCPD3	190.00	usec	f3 channel - 90 degree pulse for decoupl
PL3	-5.00	dB	f3 channel - power level for pulse (def
PL16	8.80	dB	f3 channel - power level for CPD/BB dec
SF03	60.8179117	MHz	frequency of 3rd channel
===== GRADIENT CHANNEL =====			

SAVE 2-COL Parameter Next CANCEL

• use standard

Steps: new → edasp → eda (select pulse program) → **getprosol** → **ased**

edpar

F2 - Acquisition Parameters

===== GRADIENT CHANNEL =====

GPNAM1	smsq.100	SINE.100
GPNAM2	smsq.100	SINE.100
GPNAM3	smsq.100	SINE.100
GPNAM4	smsq.100	SINE.100
GPNAM5	smsq.100	SINE.100
GPX1	0.00 %	x-gradient strength 1
GPX2	0.00 %	x-gradient strength 2
GPX3	0.00 %	x-gradient strength 3
GPX4	0.00 %	x-gradient strength 4
GPX5	0.00 %	x-gradient strength 5
GPY1	0.00 %	y-gradient strength 1
GPY2	0.00 %	y-gradient strength 2
GPY3	0.00 %	y-gradient strength 3
GPY4	0.00 %	y-gradient strength 4
GPY5	0.00 %	y-gradient strength 5

SAVE 2-COL Parameter Next CANCEL

- follow pulse program
- maybe modify

Steps: new → edasp → eda (select pulse program) → **getprosol** → **ased**

edpar

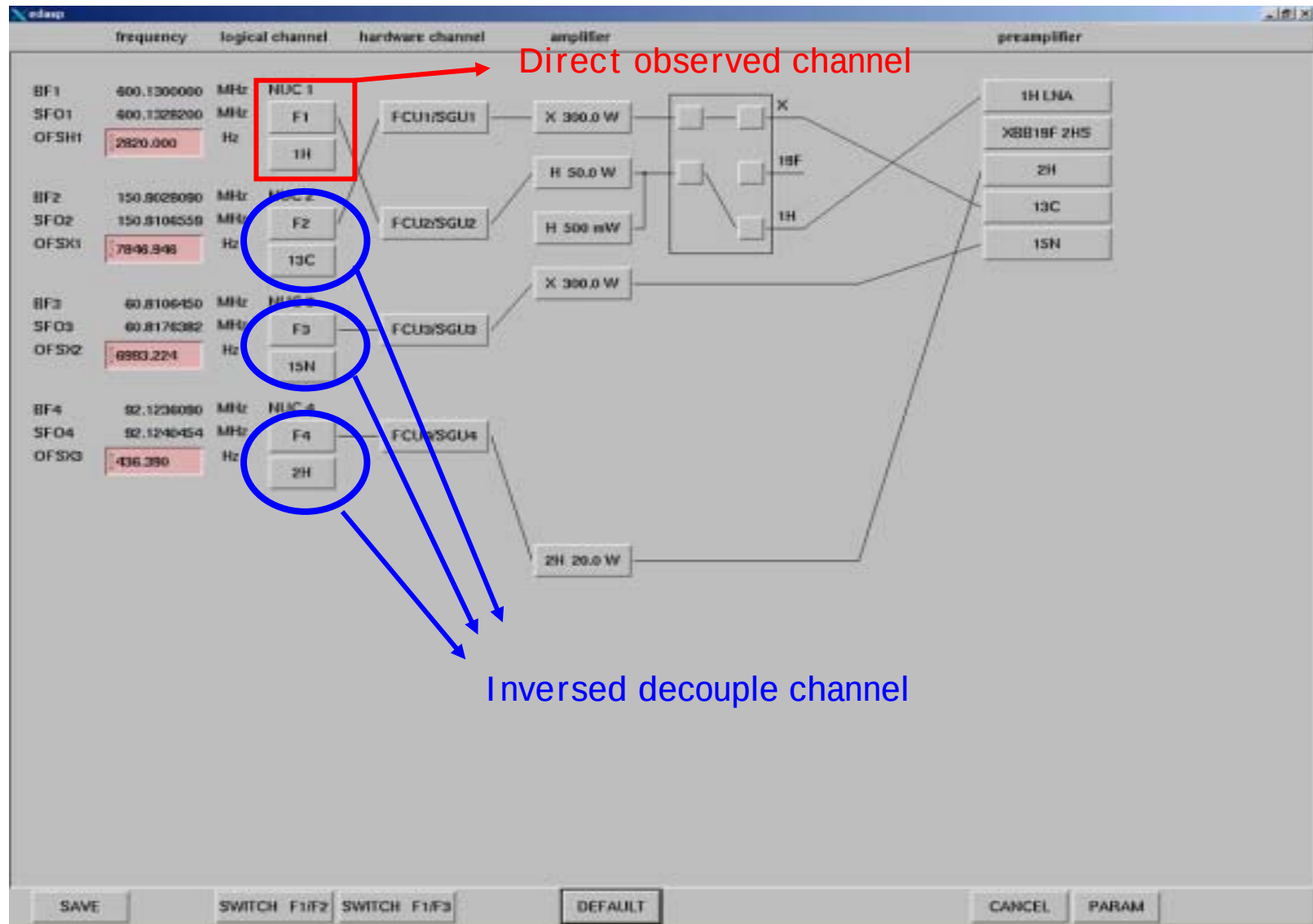
F2 - Acquisition Parameters

GPX1	0.00	%	x-gradient strength 1
GPX2	0.00	%	x-gradient strength 2
GPX3	0.00	%	x-gradient strength 3
GPX4	0.00	%	x-gradient strength 4
GPX5	0.00	%	x-gradient strength 5
GPY1	0.00	%	y-gradient strength 1
GPY2	0.00	%	y-gradient strength 2
GPY3	0.00	%	y-gradient strength 3
GPY4	0.00	%	y-gradient strength 4
GPY5	0.00	%	y-gradient strength 5
GPZ1	60.00	%	60%
GPZ2	-40.00	%	-40%
GPZ3	10.00	%	10%
GPZ4	80.00	%	80%
GPZ5	8.10	%	8.1%
P16	1000.00	usec	homospoil/gradient pulse

SAVE 2-COL Parameter Next CANCEL

• follow pulse program

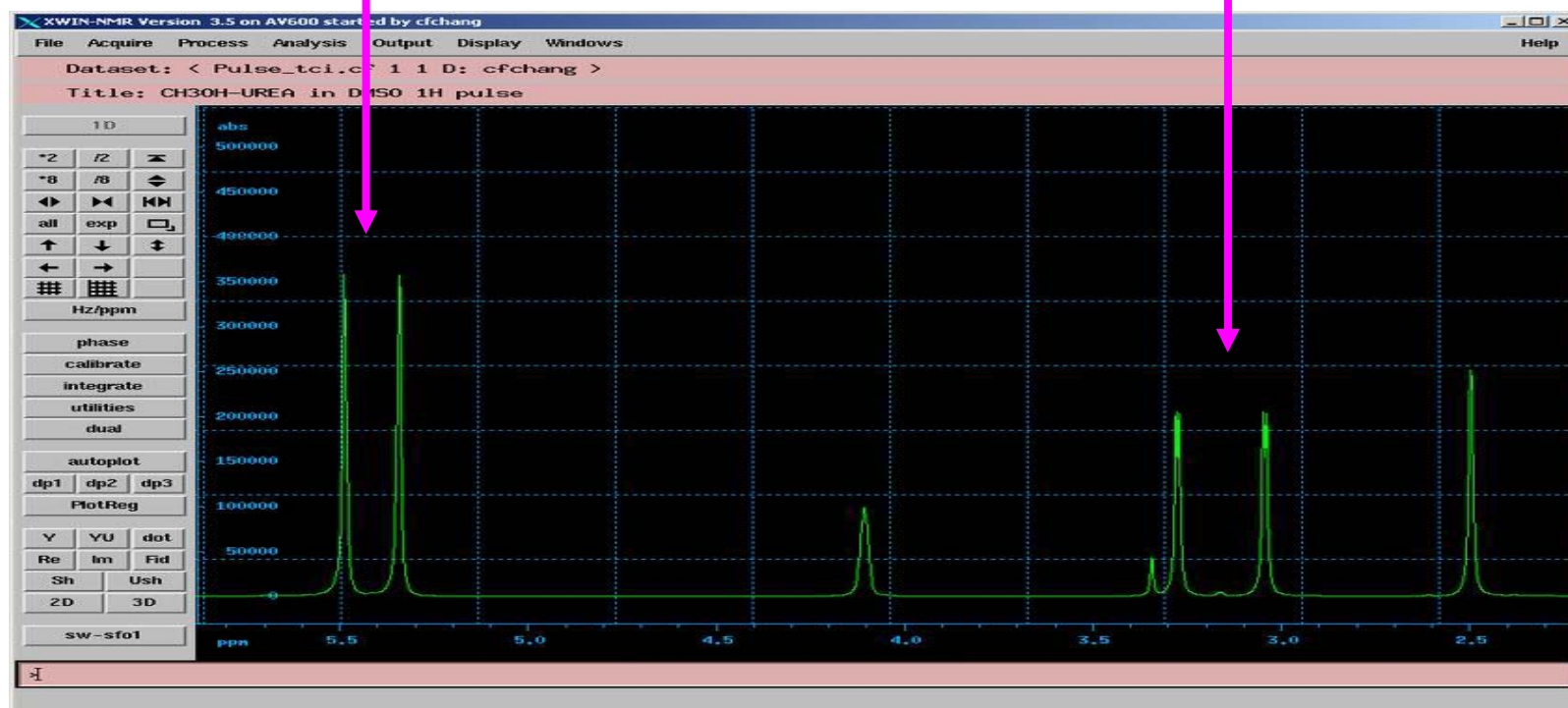
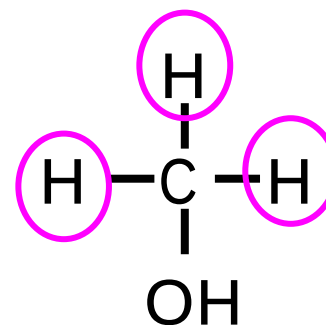
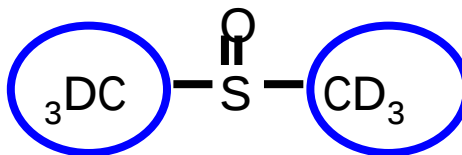
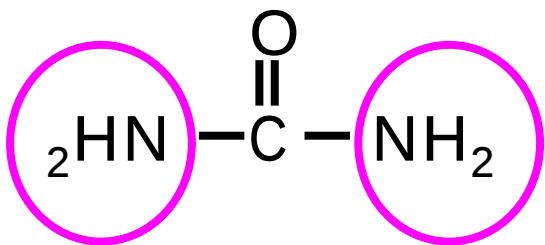
Pulse Calibration for decoupled channels (parameters in “prosol”)



Pulse Calibration for decoupled channel

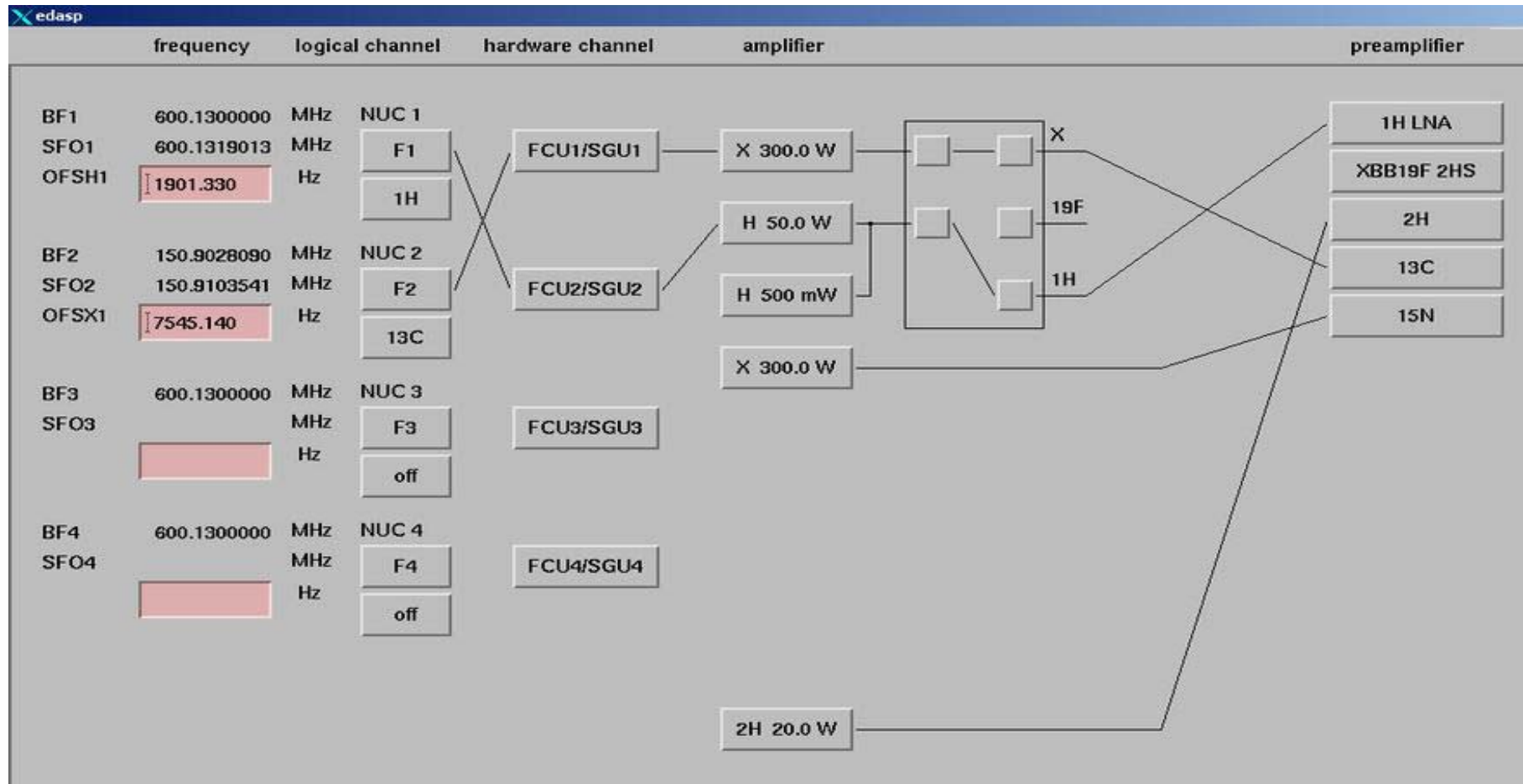
(I) Hard Pulses ($^{13}\text{C}/^{15}\text{N}$)

Sample: 0.1M $^{13}\text{CH}_3\text{OH}$ + 0.1M ^{15}N -urea in DMSO-d_6



“Decouple channel” for 13C hard pulse

Step 1: edasp



Decouple channel for ^{13}C hard pulse (decpg90)

Step 2: eda (parameter set up , PL2 vs. P3)

Left Window: F2 - Acq

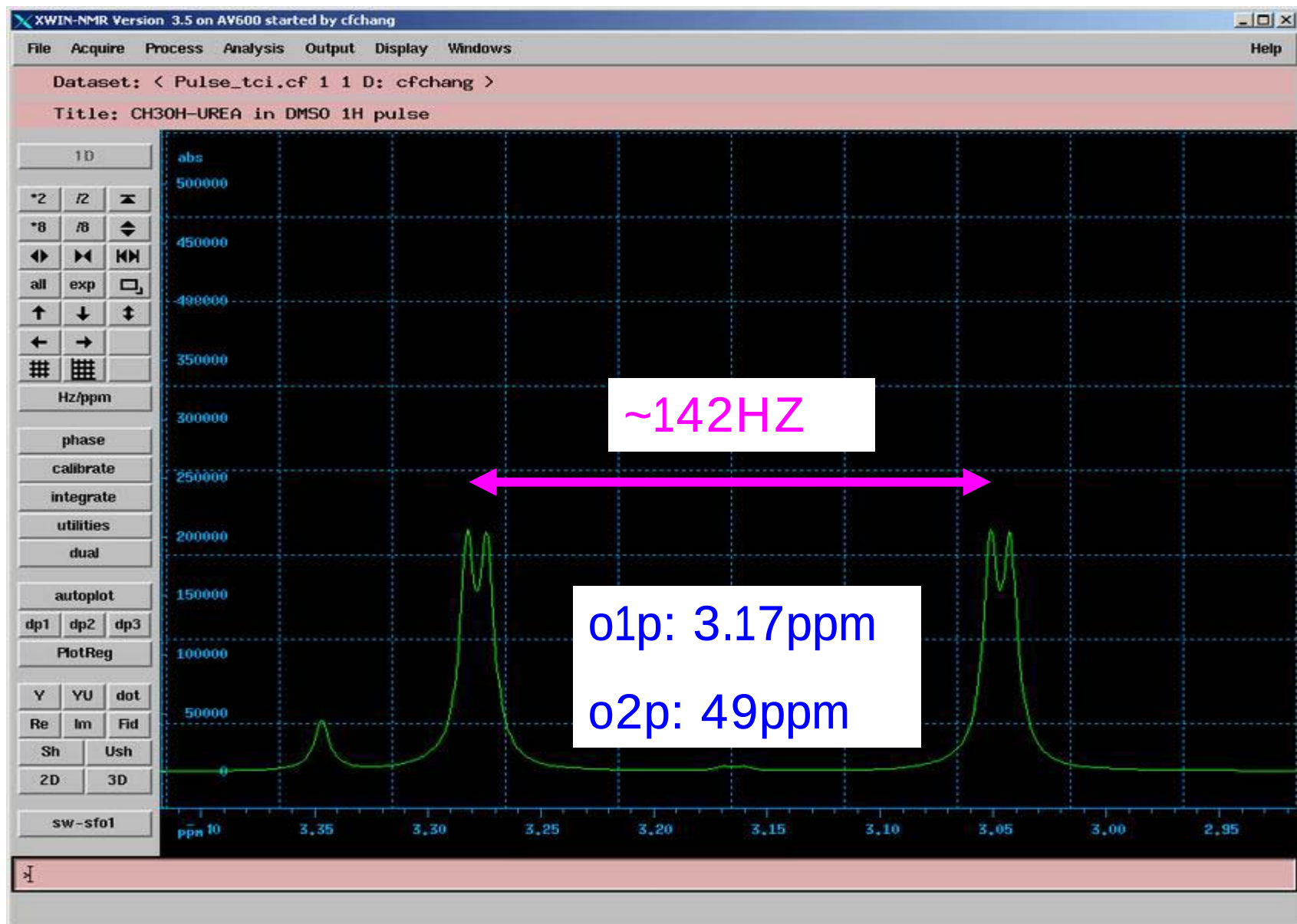
Parameter	Value	Unit
PULPROG	decpg90	
TD	16384	
NS	1	
DS	0	
SWH	1201.92	Hz
AQ	6.8162098	sec
RG	40.3	
DW	416.000	usec
DE	6.00	usec
CNST2	142.0000000	
D1	5.00000000	sec
d2	0.00352113	sec
MCREST	0.00000000	sec
MCWRK	0.01500000	sec
TD0	1	

Right Window: F2 - Acquisition Parameters

Parameter	Value	Unit	Description
CNST2	142.0000000		= J(XH)
D1	5.00000000	sec	relaxation delay: 1-5 * T1
d2	0.00352113	sec	d2=1s/(cnst2*2)
MCREST	0.00000000	sec	MCREST = 30m - 30m
MCWRK	0.01500000	sec	MCWRK = 0.500000*30m
TD0	1		Dimension of accumulation loop
===== CHANNEL f1 =====			
NUC1	1H		nucleus for channel 1
P1	9.75	usec	f1 channel - 90 degree high power puls
PL1	-5.00	dB	f1 channel - power level for pulse (def
SF01	600.1319013	MHz	frequency of observe channel
===== CHANNEL f2 =====			
NUC2	13C		nucleus for channel 2
P3	5.00	usec	f2 channel - 90 degree high power puls
PL2	-6.00	dB	f2 channel - power level for pulse (def
SF02	150.9103541	MHz	frequency of 2nd channel

Buttons: SAVE, 2-COL, Parameter, Next, CANCEL

$^{13}\text{CH}_3\text{OH}$ + 0.1M ^{15}N -urea in DMSO- d_6



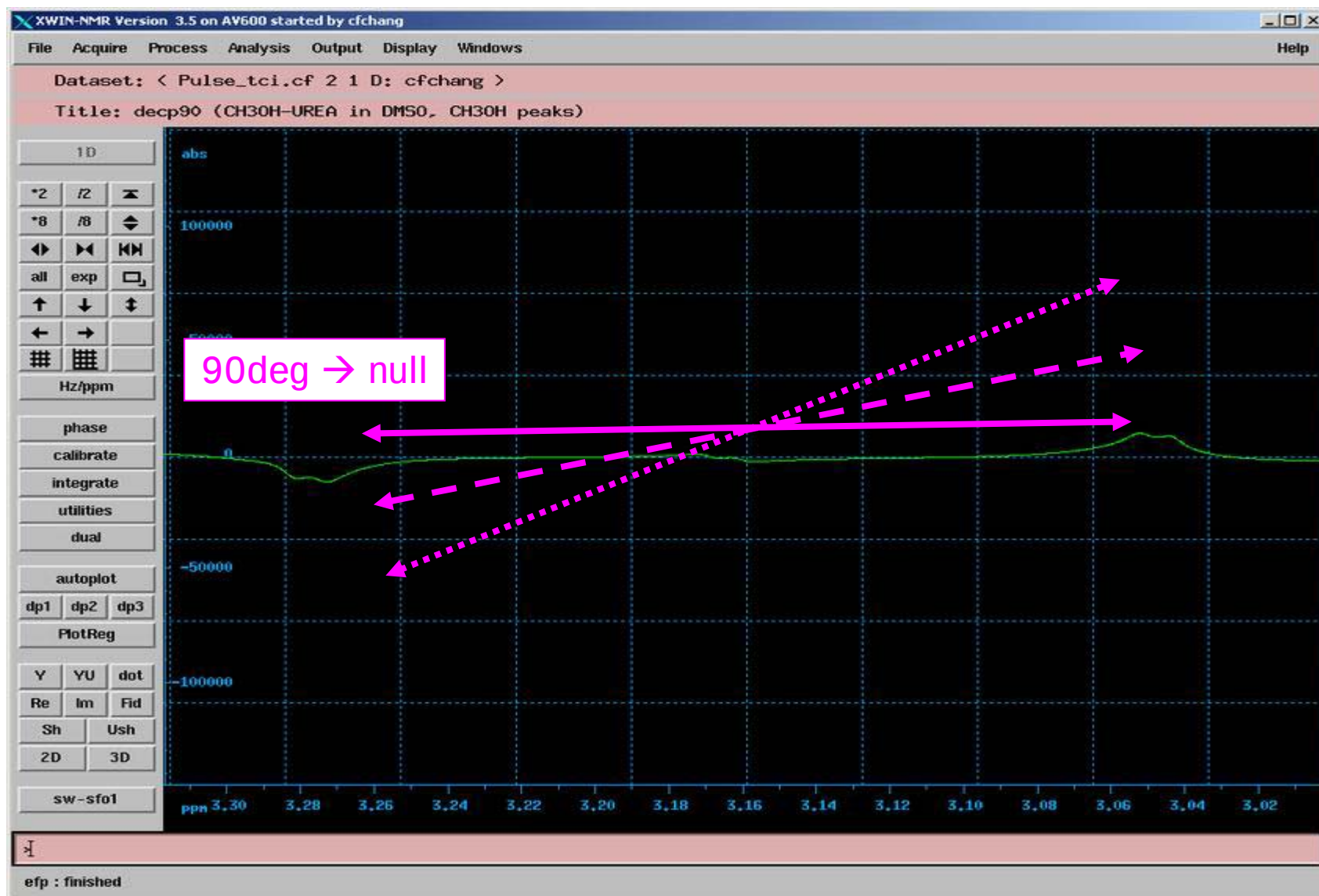
Decouple channel for ^{13}C hard pulse (dec90)

Step 3: zg ,em, ft (pl2=-6dB, p3=5us)



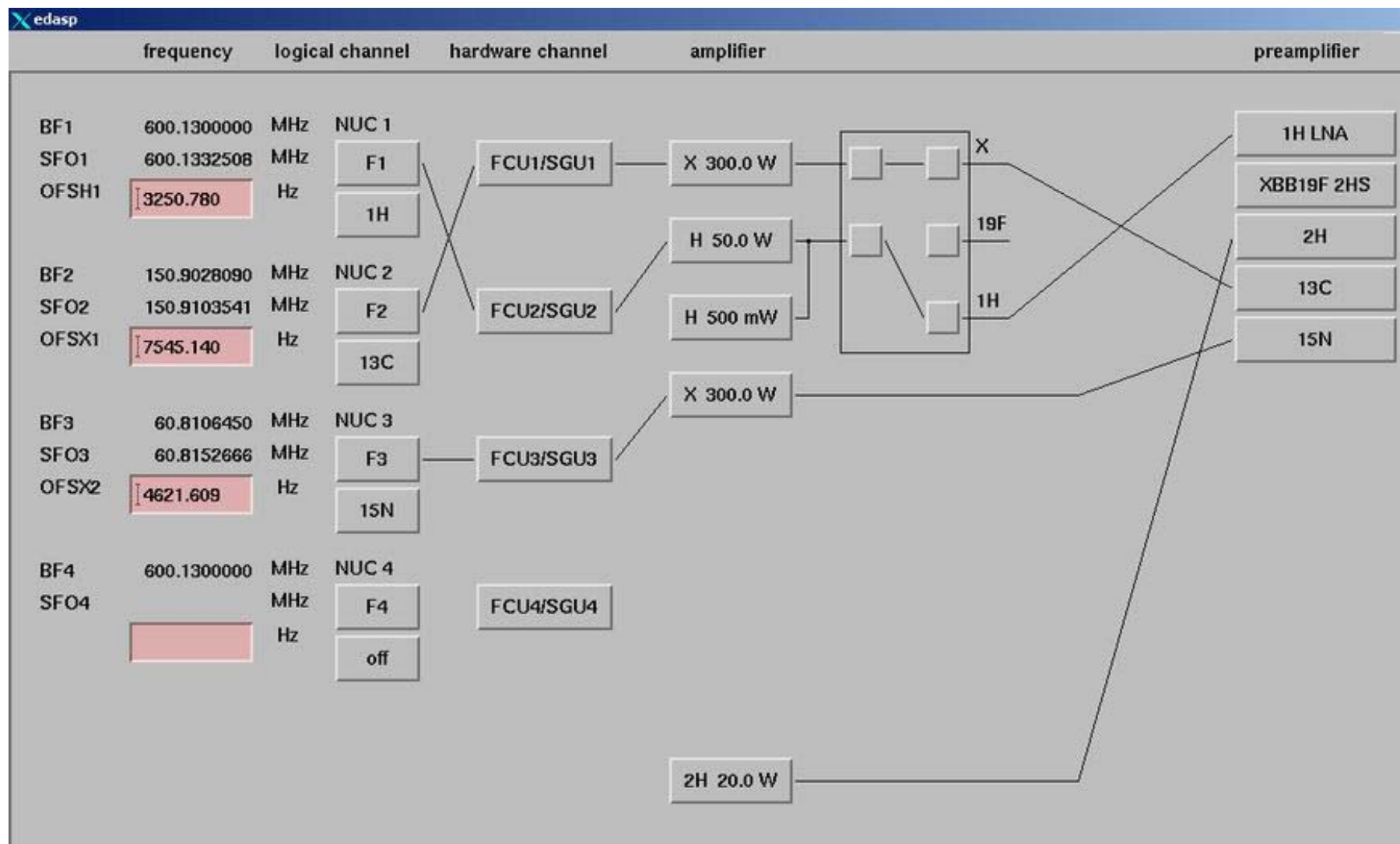
Decouple channel for ^{13}C hard pulse (dec90)

Step 4: zg, em, fp (adjust p3 until reach null)



“Decouple channel” for ^{15}N hard pulse

Step 1: edasp



Decouple channel for ^{15}N hard pulse (dec90f3)

Step 2: eda (parameter set up, PL3 vs. P21)

The image displays two screenshots of the 'edpar' software interface, showing parameter setup for a ^{15}N hard pulse.

Left Window (F2 - Acquisition Parameters):

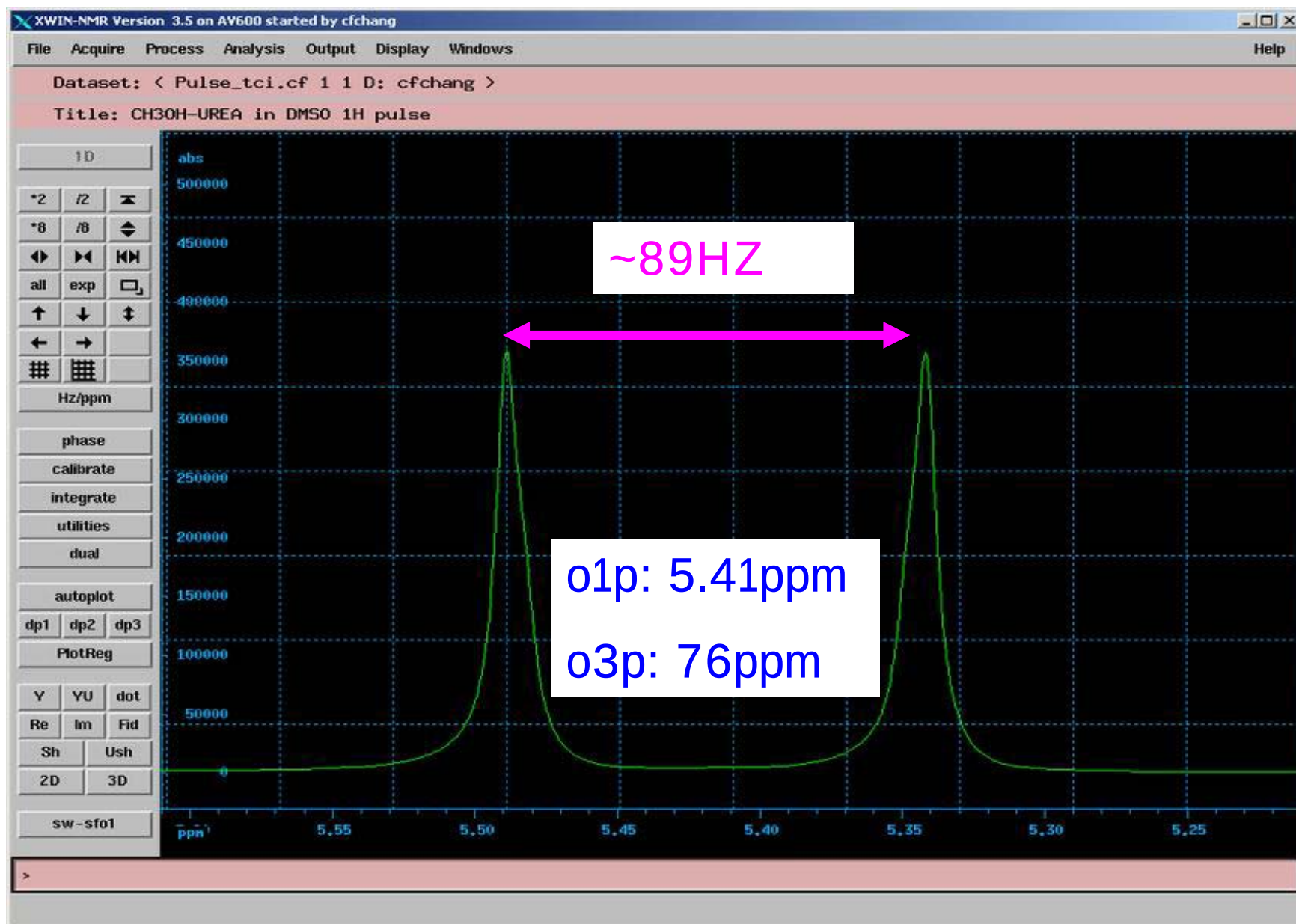
- PULPROG: **dec90f3** (circled in green)
- TD: 16384
- NS: 1
- DS: 0
- SWH: 600.96 Hz
- AQ: 13.6323700 sec
- RG: 64
- DW: 832.000 usec
- DE: 6.00 usec
- CNST2: **89.0000000** (circled in green)
- D1: 2.00000000 sec
- d2: 0.00561798 sec
- MCREST: 0.00000000 sec
- MCWRK: 0.01500000 sec
- TD0: 1

Right Window (F2 - Acquisition Parameters):

- CNST2: 89.0000000 = J(XH)
- D1: 2.00000000 sec relaxation delay: $1-5 \cdot T_1$
- d2: 0.00561798 sec $d2=1s/(cnst2*2)$
- MCREST: 0.00000000 sec MCREST = 30m - 30m
- MCWRK: 0.01500000 sec MCWRK = 0.500000*30m
- TD0: 1 Dimension of accumulation loop
- ===== CHANNEL f1 =====
- NUC1: 1H nucleus for channel 1
- P1: **9.45** usec f1 channel - 90 degree high power puls
- PL1: **-5.00** dB f1 channel - power level for pulse (def
- SF01: 600.1332508 MHz frequency of observe channel
- ===== CHANNEL f3 =====
- NUC3: 15N nucleus for channel 3
- P21: **10.00** usec f3 channel - 90 degree high power puls
- PL3: **-5.00** dB f3 channel - power level for pulse (def
- SF03: 60.8152666 MHz frequency of 3rd channel

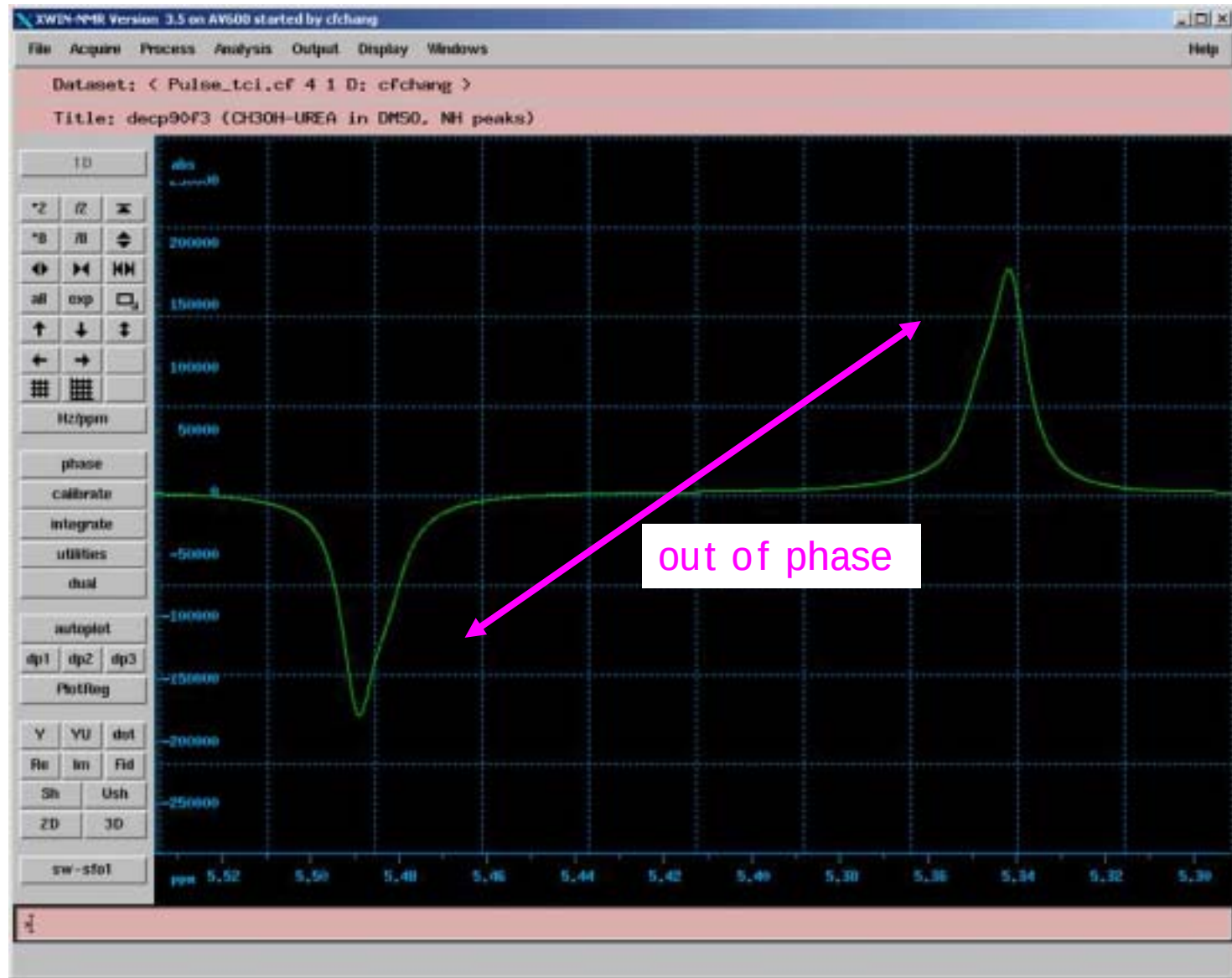
Buttons: SAVE, 2-COL, Parameter, Next, CANCEL

$^{13}\text{CH}_3\text{OH}$ + ^{15}N -urea in DMSO-d₆



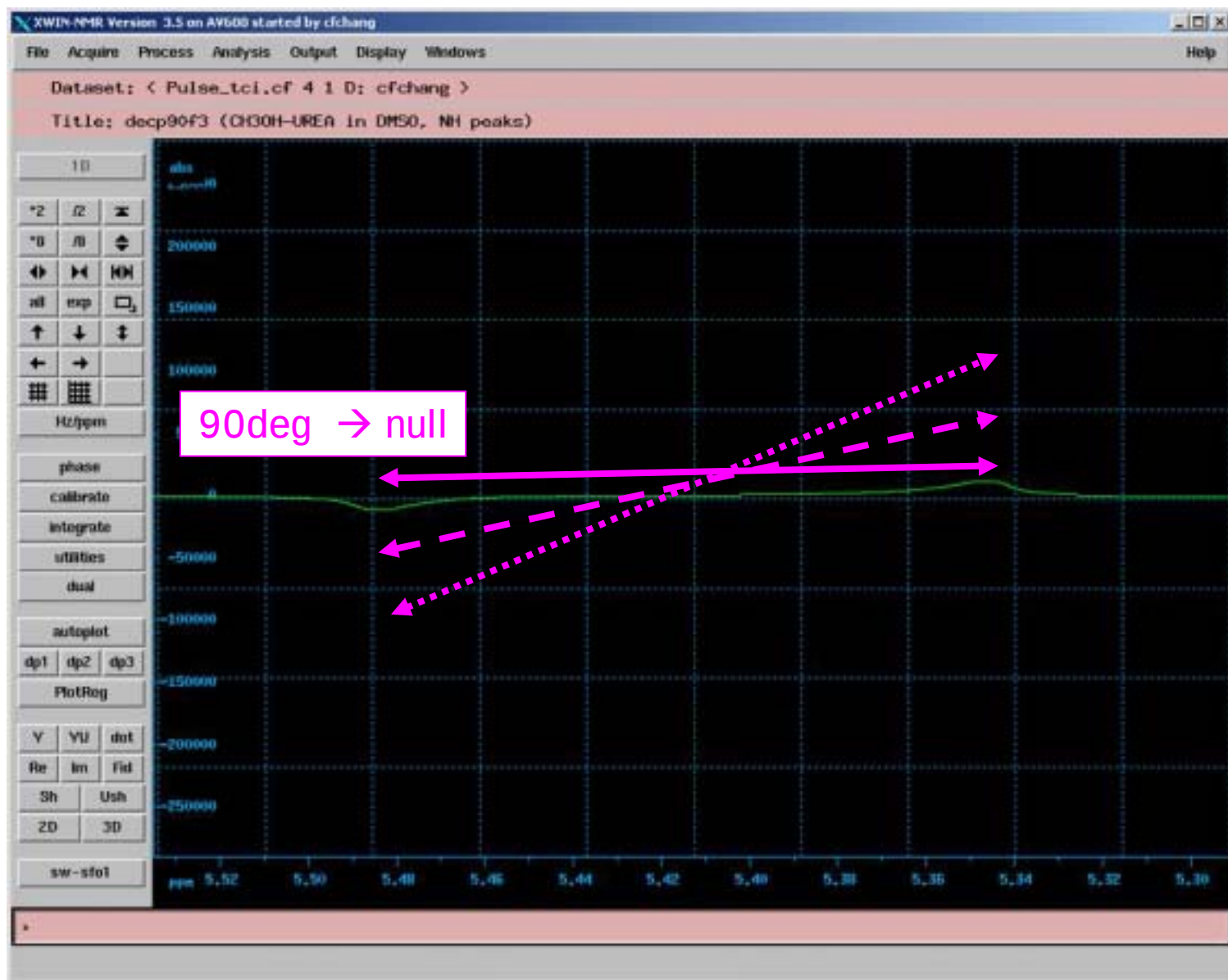
Decouple channel for ¹⁵N hard pulse (decpg90f3)

Step 3: zg ,em, ft (pl3=-5dB, p21=5us)



Decouple channel for ^{15}N hard pulse (dec90f3)

Step 4: zg, em, fp (adjust p21 until reach null)



Pulse Calibration for decoupled channel

(I I) ^{13}C Shape Pulses

How to optimize the pulse length for 13C shape pulse?

Excitation

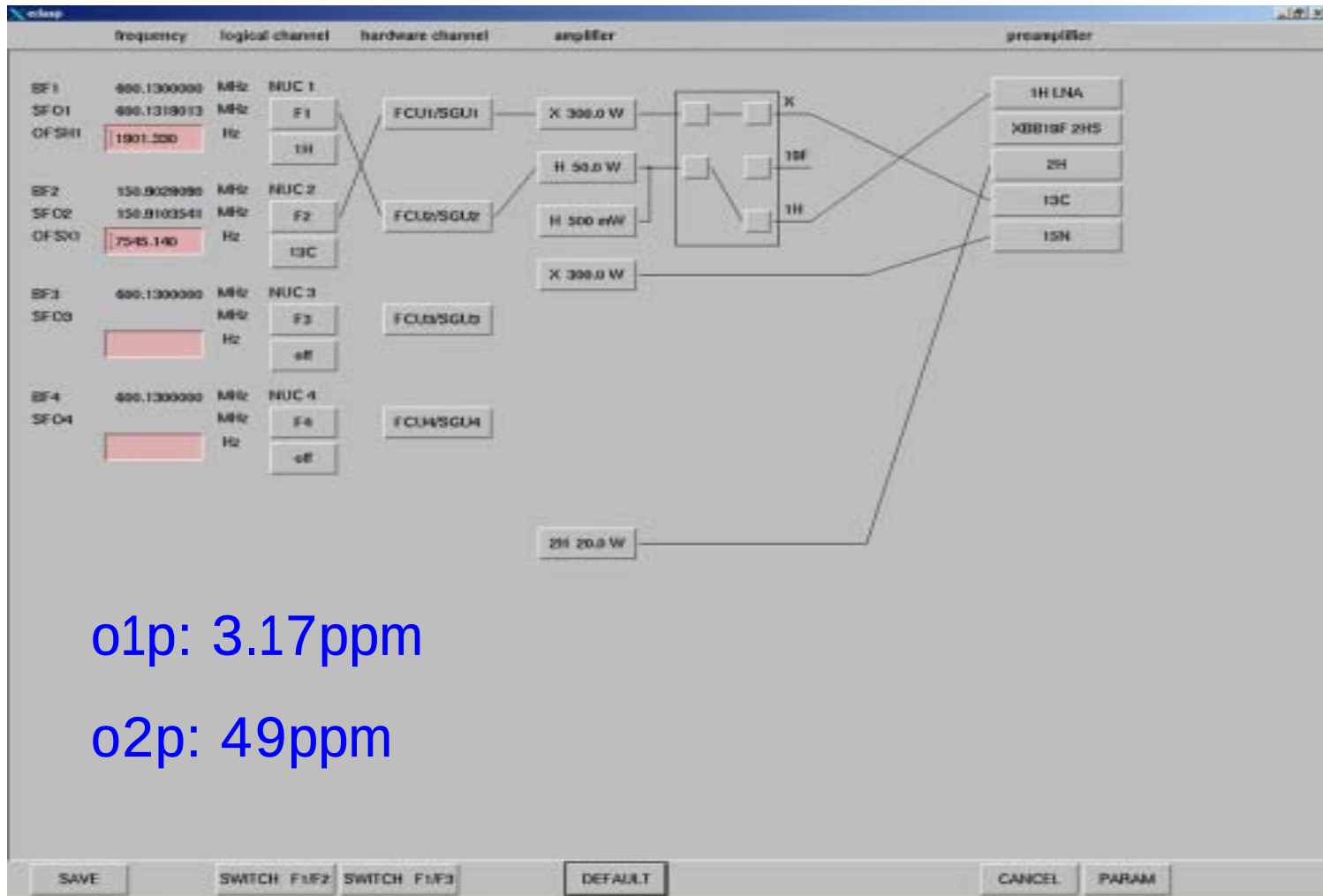
Shape	Pulse length	Cover frequency	500MHz (125Hz)	600MHz (150Hz)	800MHz (200Hz)
Q5.1000	410us	15073Hz	120ppm	100ppm	75ppm
Q5.1000	320u	19312Hz	154ppm	128ppm	96ppm

Inversion

Shape	Pulse length	Cover frequency	500MHz (125Hz)	600MHz (150Hz)	800MHz (200Hz)
Q3.1000	256us	13320Hz	106ppm	89ppm	66.6ppm
Q3.1000	768us	4440Hz	35ppm	29.6ppm	22.2ppm
Reburp.1000	1500u	3093Hz	24.75ppm		
Reburp.1000	1250u	3712Hz		24.75ppm	

Decouple channel for ^{13}C 90 deg shape pulse

Step 1: edasp



o1p: 3.17ppm

o2p: 49ppm

Decouple channel for ^{13}C 90 deg shape pulse (decp90sp)

Step 2: eda (parameter set up , p13 vs. sp2 for SPNAM2)

The image shows two side-by-side windows from the 'edpar' software, both titled 'F2 - Acquisition Parameters'.

Left Window (F2 - Acquisition Parameters):

Parameter	Value	Unit
PULPROG	decp90sp	
TD	32768	
NS	1	
DS	0	
SWH	600.96	Hz
AQ	27.2638588	sec
RG	24	
DW	832.000	usec
DE	6.00	usec
CNST2	142.0000000	
D1	5.00000000	sec
d2	0.00352113	sec
MCREST	0.00000000	sec
MCWRK	0.01500000	sec
TD0	1	

Right Window (F2 - Acquisition Parameters):

MCWRK = 0.01500000 sec
MCWRK = 0.500000*30m

TD0 = 1
Dimension of accumulation loop

===== CHANNEL f1 =====

NUC1 = 1H
nucleus for channel 1

f1 channel - 90 degree high power puls

f1 channel - power level for pulse (def

frequency of observe channel

===== CHANNEL f2 =====

NUC2 = ^{13}C
nucleus for channel 2

f2 channel - 90 degree shaped pulse

power PL2

frequency of 2nd channel

f2 channel - shaped pulse 90 degree (

file name for sp2

phase alignment of freq. offset in SP2

offset frequency for SP2

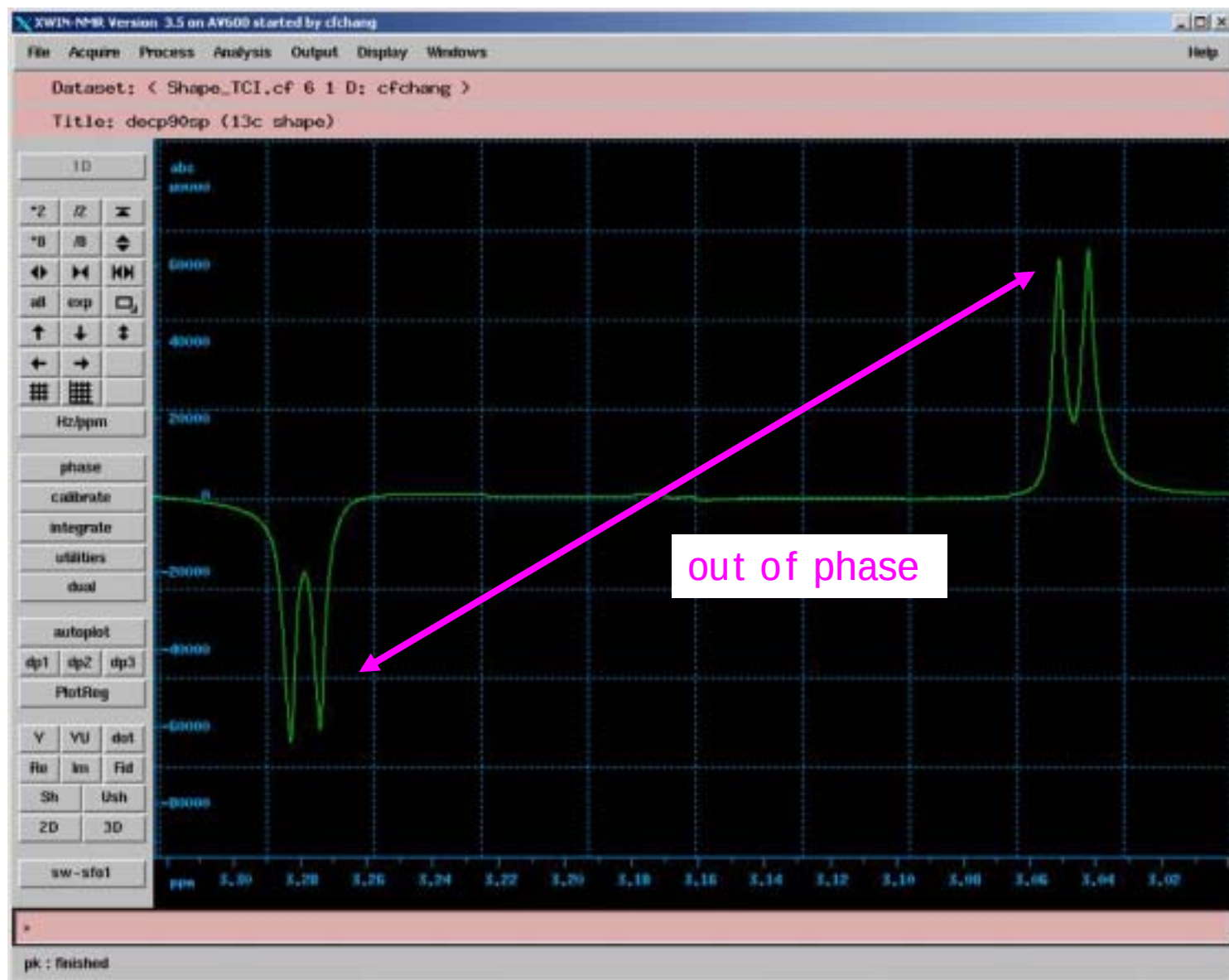
Parameters highlighted in the image:

- decp90sp** (PULPROG)
- CNST2** (142.0000000)
- P1** (9.45 usec)
- PL1** (-5.00 dB)
- SF01** (600.1319012 MHz)
- P13** (410.00 usec)
- PL2** (120.00 dB)
- SP2** (10.00 dB)
- SPNAM2** (05.1000)

Buttons at the bottom: SAVE, 2-COL, Parameter, Next, CANCEL

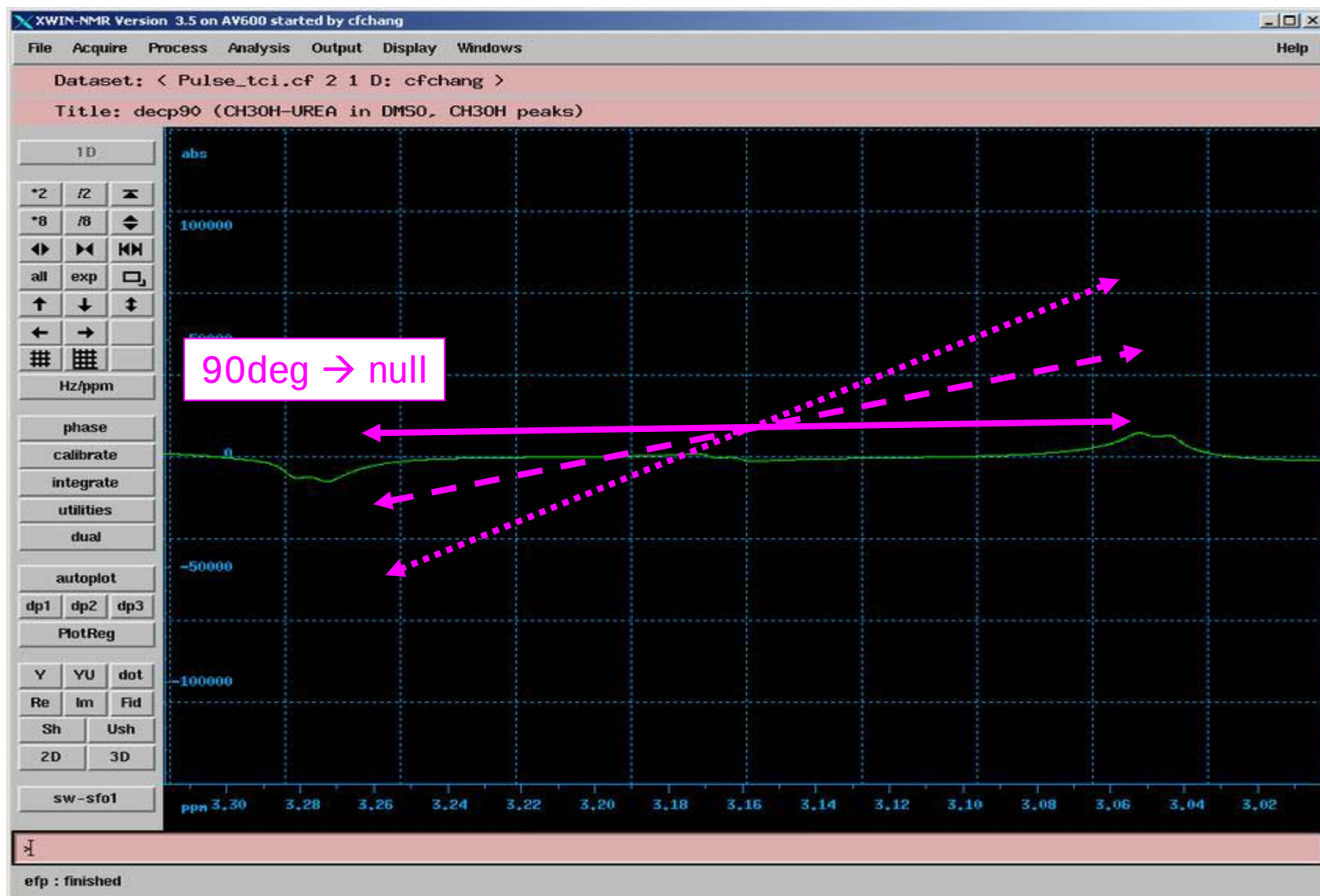
Decouple channel for ^{13}C 90 deg shape pulse (dec90sp)

Step 3: zg ,em, ft (p13= 410us , sp2= 10dB , SPNAM2= Q5.1000)



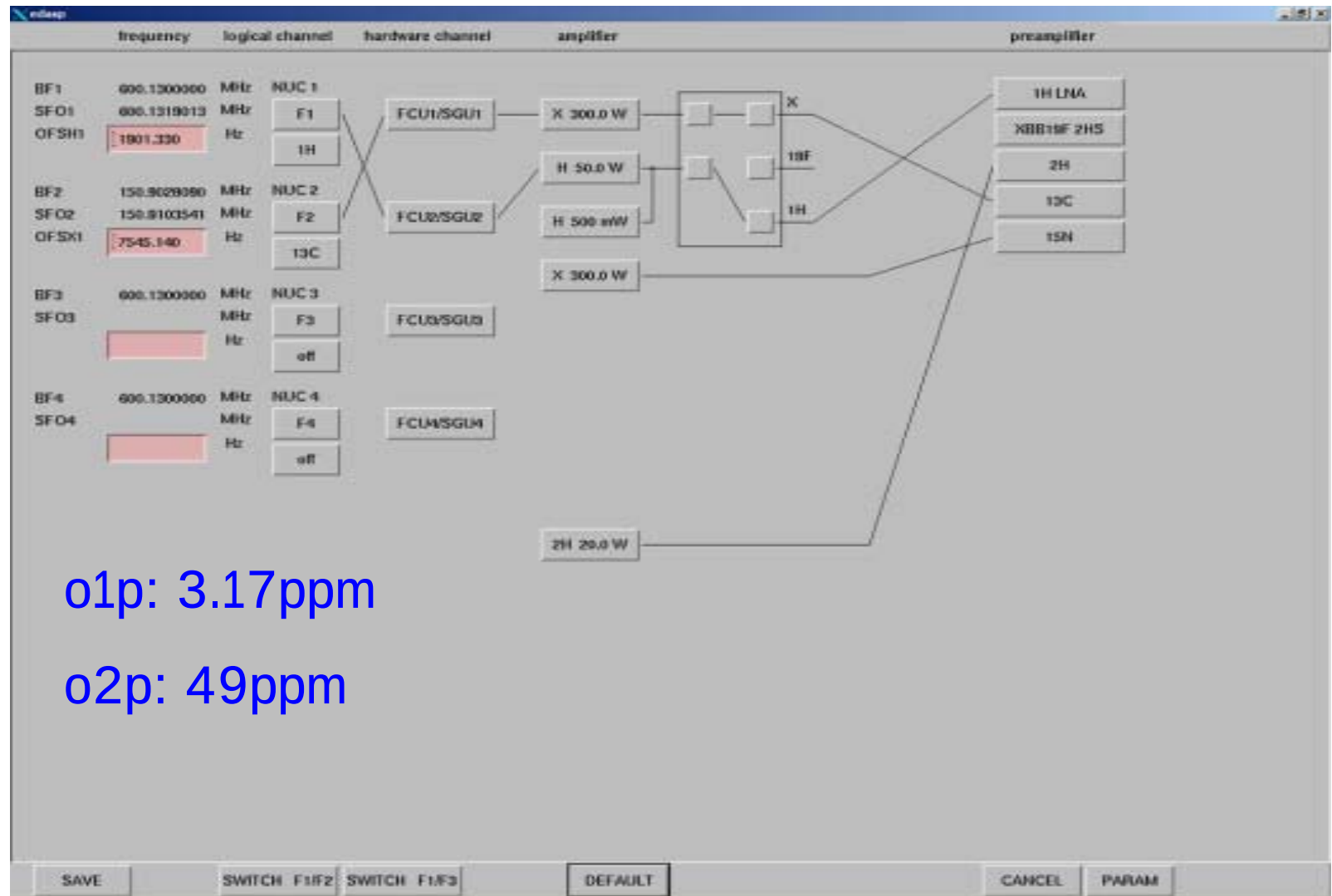
Decouple channel for ^{13}C 90 deg shape pulse (dec90sp)

Step 4: zg, em, fp (adjust sp2 until reach null)



Decouple channel for 13C180 deg shape pulse

Step 1: edasp



o1p: 3.17ppm

o2p: 49ppm

Decouple channel for ^{13}C 180 deg shape pulse (decp180sp)

Step 2: eda (parameter set up , p14 vs. sp3 for SPNAM3)

The image shows two windows from the 'edpar' software. The left window, titled 'F2 -', displays a list of parameters and their values. The right window, titled 'F2 - Acquisition Parameters', provides detailed settings for two channels: NUC1 (1H) and NUC2 (13C).

Left Window (F2 -) Parameters:

Parameter	Value	Unit
PULPROG	dec180sp	
TD	32768	
NS	2	
DS	2	
SWH	1201.92	Hz
AQ	13.6319542	sec
RG	32	
DW	416.000	usec
DE	6.00	usec
CNST2	142.0000000	
D1	3.00000000	sec
d2	0.00352113	sec
d20	0.00012800	sec
d21	0.00339313	sec
MCREST	0.00000000	sec
MCWRK	0.01500000	sec

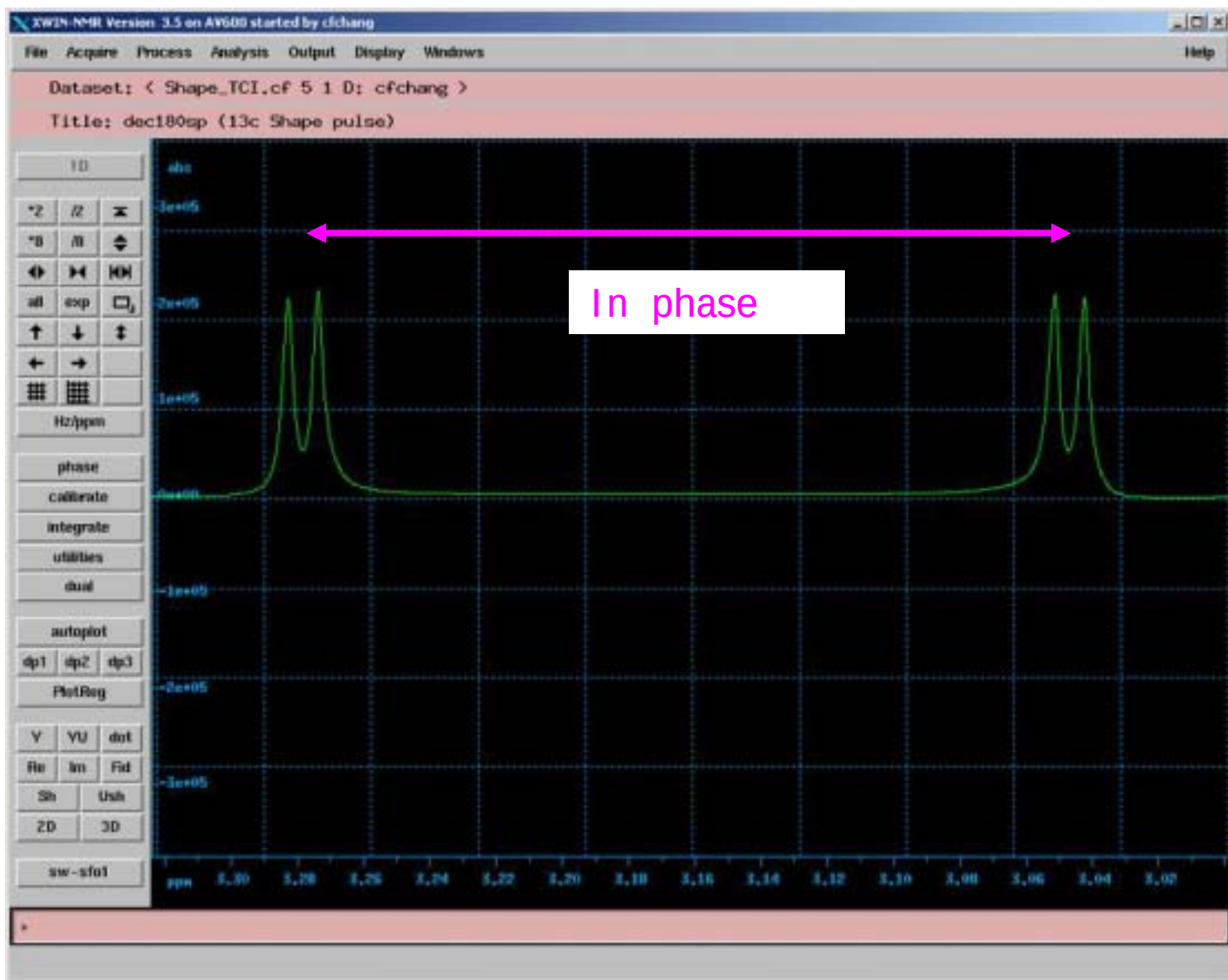
Right Window (F2 - Acquisition Parameters) Parameters:

Parameter	Value	Unit	Description
NUC1	1H		nucleus for channel 1
P1	9.45	usec	f1 channel - 90 degree high power puls
p2	18.90	usec	p2=p1*2
PL1	-5.00	dB	f1 channel - power level for pulse (def
SF01	600.1319064	MHz	frequency of observe channel
===== CHANNEL f2 =====			
NUC2	13C		nucleus for channel 2
P3	15.00	usec	f2 channel - 90 degree high power puls
P14	256.00	usec	f2 channel - 180 degree shaped pulse
PL0	120.00	dB	120dB
PL2	-6.00	dB	f2 channel - power level for pulse (def
SF02	150.9103541	MHz	frequency of 2nd channel
SP3	10.00	dB	f2 channel - shaped pulse 180 degree (
SPNAM3	03.1000		file name for sp3
SPOAL3	0.500		phase alignment of freq. offset in SP3
SPOFF3	0.00	Hz	offset frequency for SP3

Buttons: SAVE, 2-COL, Parameter, Next, CANCEL

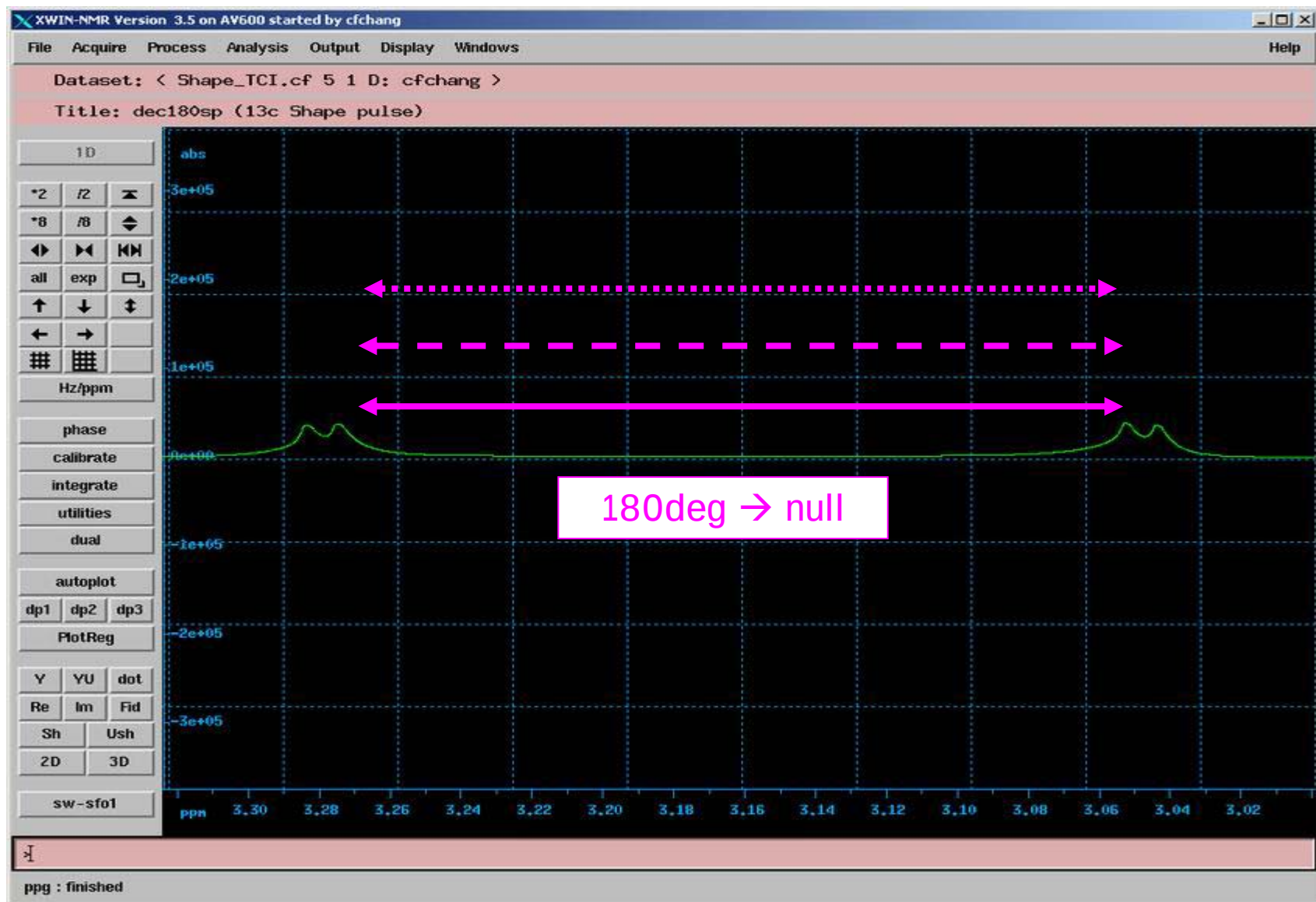
Decouple channel for 13C180 deg shape pulse (decpl80sp)

Step 3: zg ,em, ft (p14= 256us , sp3= 10dB , SPNAM3= Q3.1000)



Decouple channel for ^{13}C 180 deg shape pulse (decp180sp)

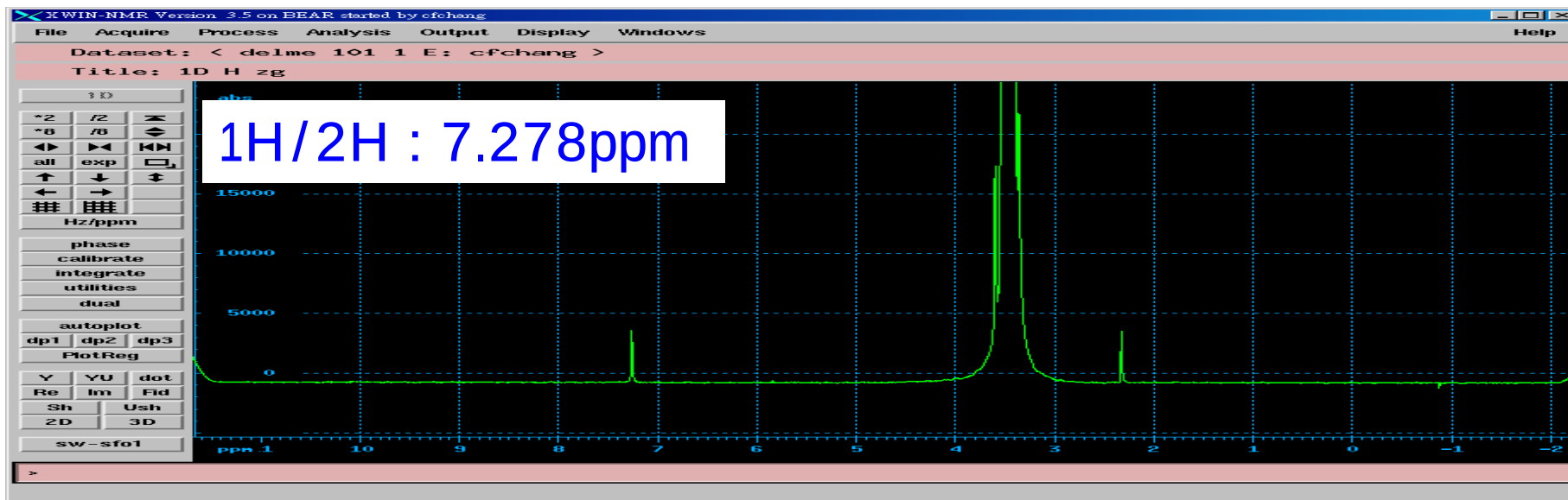
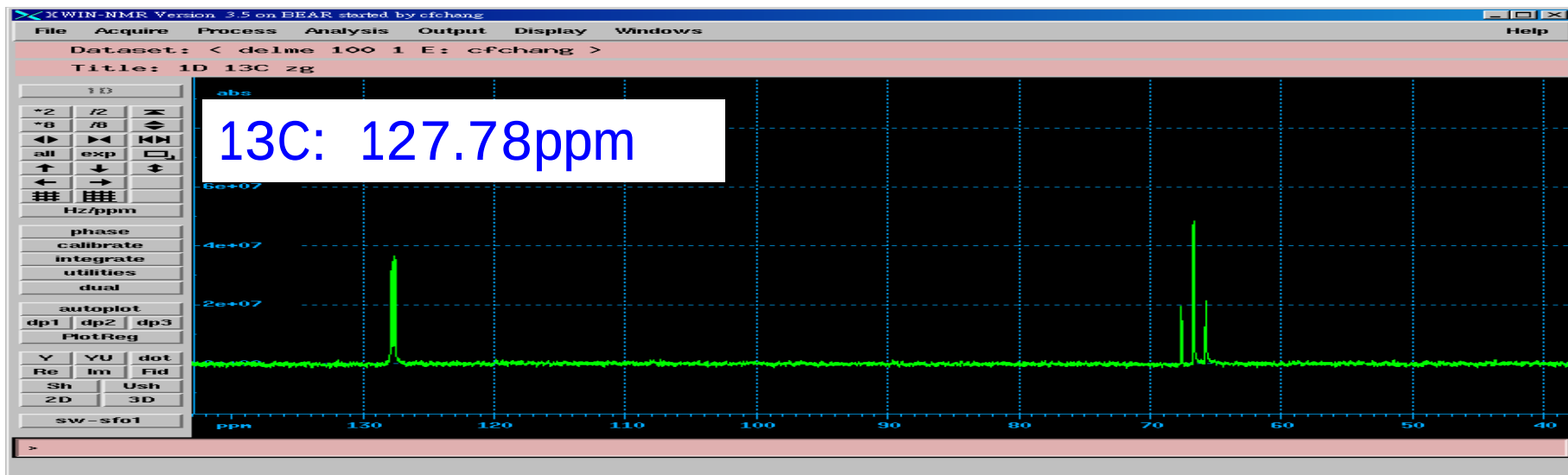
Step 4: zg, em, fp (adjust sp3 until reach null)



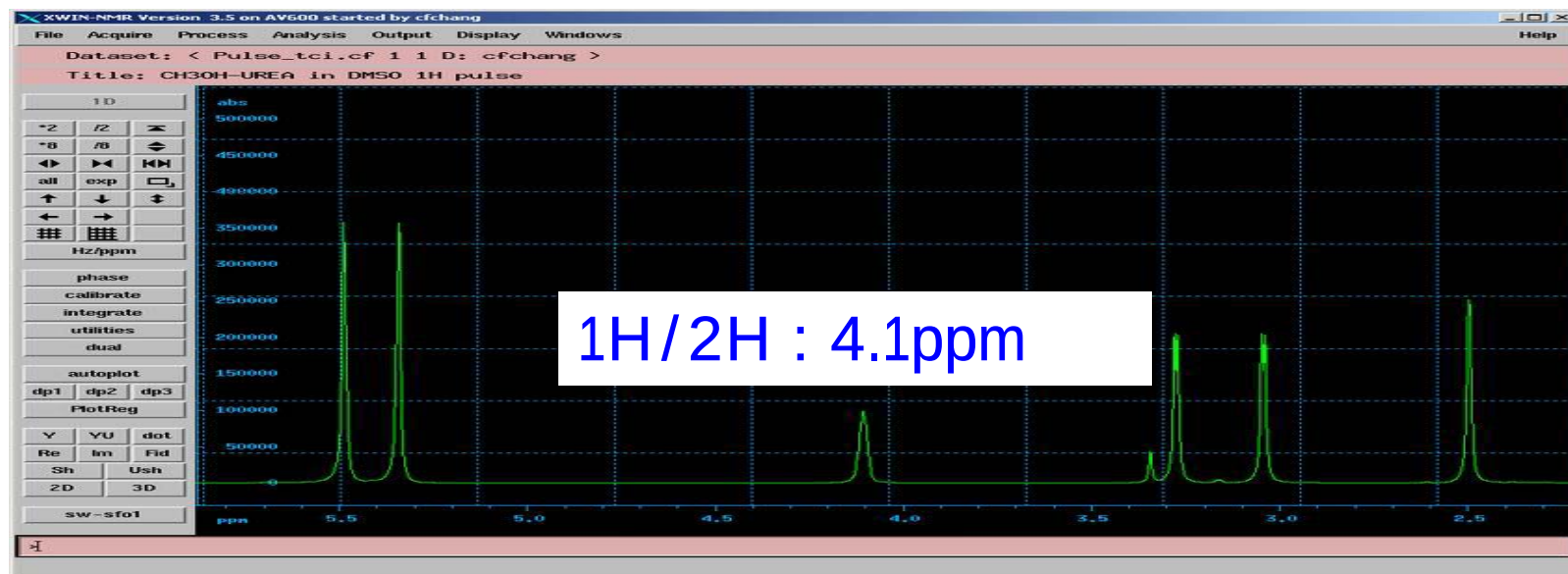
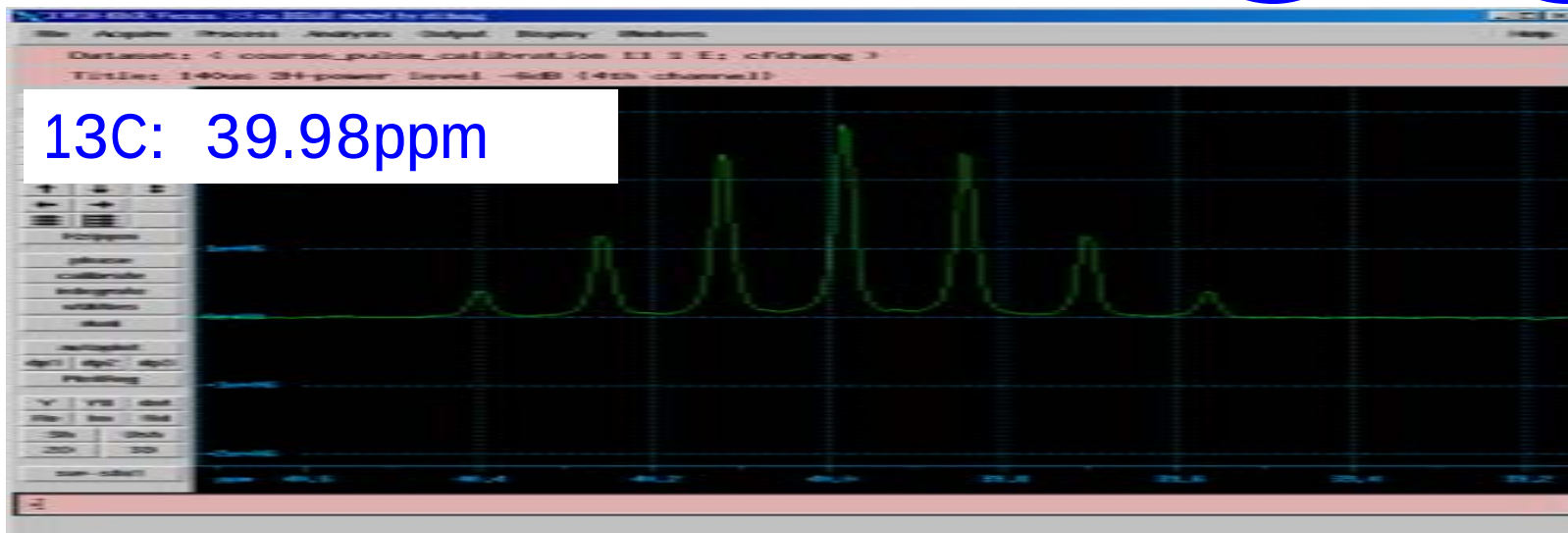
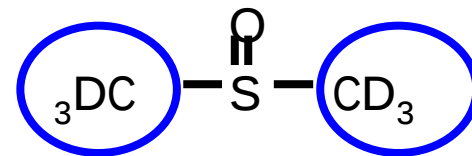
Pulse Calibration for decoupled channel

(III) 2H Hard Pulses

Sample: 60% Benzene-d₆

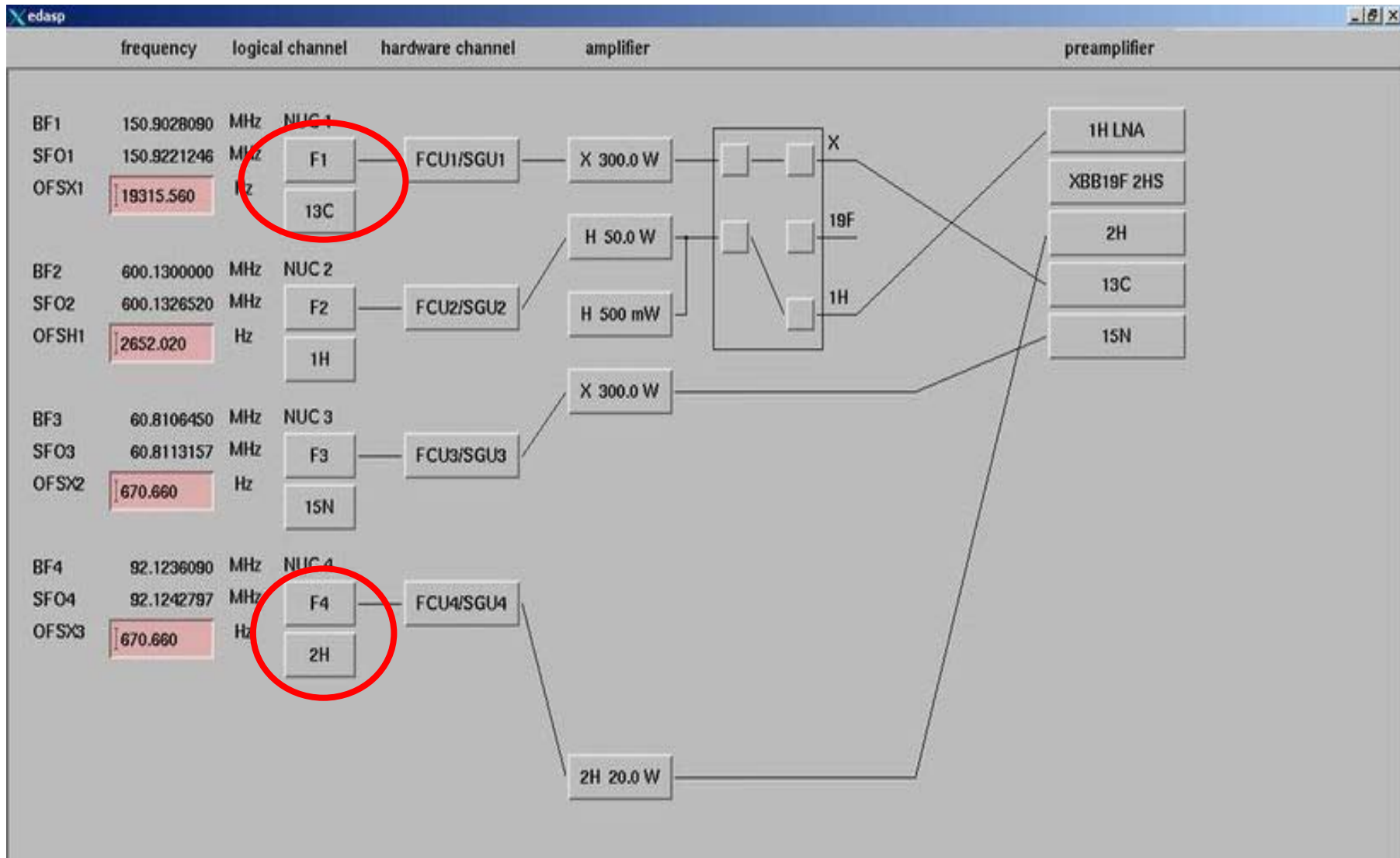


Sample: 0.1M $^{13}\text{CH}_3\text{OH}$ + 0.1M ^{15}N -urea in $\text{DMSO-}d_6$



Decouple channel for 2H hard pulse

Step 1: edasp



Decouple channel for 2H hard pulse (dec902hf4)

Step 2: eda (parameter set up , PL4 vs. P23)

The image shows two windows from the 'edpar' software. The left window is titled 'F2 - Ac' and the right window is titled 'F2 - Acquisition Parameters'. Both windows display a list of parameters and their values. The values are entered in red boxes. The parameters are listed in two columns. The left column lists parameters and their values, while the right column lists parameters and their units and descriptions. The parameters are: PULPROG, TD, NS, DS, SWH, AQ, RG, DW, DE, CNST5, D1, d4, d11, d12, MCREST, MCWRK, NUC1, P1, PL1, SF01, NUC4, P23, PL4, and SF04. The values are: decp902hf4, 16384, 1, 0, 1502.40 Hz, 5.4529781 sec, 7298.2, 332.800 usec, 6.00 usec, 22 or 24, > 60sec, 0.01041667 sec, 0.03000000 sec, 0.00002000 sec, 0.00000000 sec, 0.01500000 sec, 13C, 15.50 usec, -5.00 dB, 150.9221246 MHz, 2H, 300.00 usec, 10.00 dB, and 92.1242797 MHz. The parameters are grouped into two channels: CHANNEL f1 and CHANNEL f4. The parameters for CHANNEL f1 are: P1, PL1, SF01, NUC1, and the parameters for CHANNEL f4 are: P23, PL4, SF04. The parameters are also grouped into two sections: F2 - Ac and F2 - Acquisition Parameters. The parameters are listed in two columns: the left column lists parameters and their values, and the right column lists parameters and their units and descriptions. The parameters are: PULPROG, TD, NS, DS, SWH, AQ, RG, DW, DE, CNST5, D1, d4, d11, d12, MCREST, MCWRK, NUC1, P1, PL1, SF01, NUC4, P23, PL4, and SF04. The values are: decp902hf4, 16384, 1, 0, 1502.40 Hz, 5.4529781 sec, 7298.2, 332.800 usec, 6.00 usec, 22 or 24, > 60sec, 0.01041667 sec, 0.03000000 sec, 0.00002000 sec, 0.00000000 sec, 0.01500000 sec, 13C, 15.50 usec, -5.00 dB, 150.9221246 MHz, 2H, 300.00 usec, 10.00 dB, and 92.1242797 MHz. The parameters are grouped into two channels: CHANNEL f1 and CHANNEL f4. The parameters for CHANNEL f1 are: P1, PL1, SF01, NUC1, and the parameters for CHANNEL f4 are: P23, PL4, SF04. The parameters are also grouped into two sections: F2 - Ac and F2 - Acquisition Parameters. The parameters are listed in two columns: the left column lists parameters and their values, and the right column lists parameters and their units and descriptions. The parameters are: PULPROG, TD, NS, DS, SWH, AQ, RG, DW, DE, CNST5, D1, d4, d11, d12, MCREST, MCWRK, NUC1, P1, PL1, SF01, NUC4, P23, PL4, and SF04. The values are: decp902hf4, 16384, 1, 0, 1502.40 Hz, 5.4529781 sec, 7298.2, 332.800 usec, 6.00 usec, 22 or 24, > 60sec, 0.01041667 sec, 0.03000000 sec, 0.00002000 sec, 0.00000000 sec, 0.01500000 sec, 13C, 15.50 usec, -5.00 dB, 150.9221246 MHz, 2H, 300.00 usec, 10.00 dB, and 92.1242797 MHz. The parameters are grouped into two channels: CHANNEL f1 and CHANNEL f4. The parameters for CHANNEL f1 are: P1, PL1, SF01, NUC1, and the parameters for CHANNEL f4 are: P23, PL4, SF04.

Parameter	Value	Unit	Description
PULPROG	decp902hf4		
TD	16384		
NS	1		
DS	0		
SWH	1502.40	Hz	
AQ	5.4529781	sec	
RG	7298.2		
DW	332.800	usec	
DE	6.00	usec	
CNST5	22 or 24		
D1	> 60sec	sec	
d4	0.01041667	sec	d4=1s/(cnst5*4)
d11	0.03000000	sec	d11=30m
d12	0.00002000	sec	d12=20u
MCREST	0.00000000	sec	MCREST = 30m - 30m
MCWRK	0.01500000	sec	MCWRK = 0.500000*30m
TD0	1		Dimension of accumulation loop
NUC1	13C		nucleus for channel 1
P1	15.50	usec	f1 channel - 90 degree high power puls
PL1	-5.00	dB	f1 channel - power level for pulse (def
SF01	150.9221246	MHz	frequency of observe channel
NUC4	2H		nucleus for channel 4
P23	300.00	usec	f4 channel - 90 degree high power puls
PL4	10.00	dB	f4 channel - power level for pulse (def
SF04	92.1242797	MHz	frequency of 4th channel

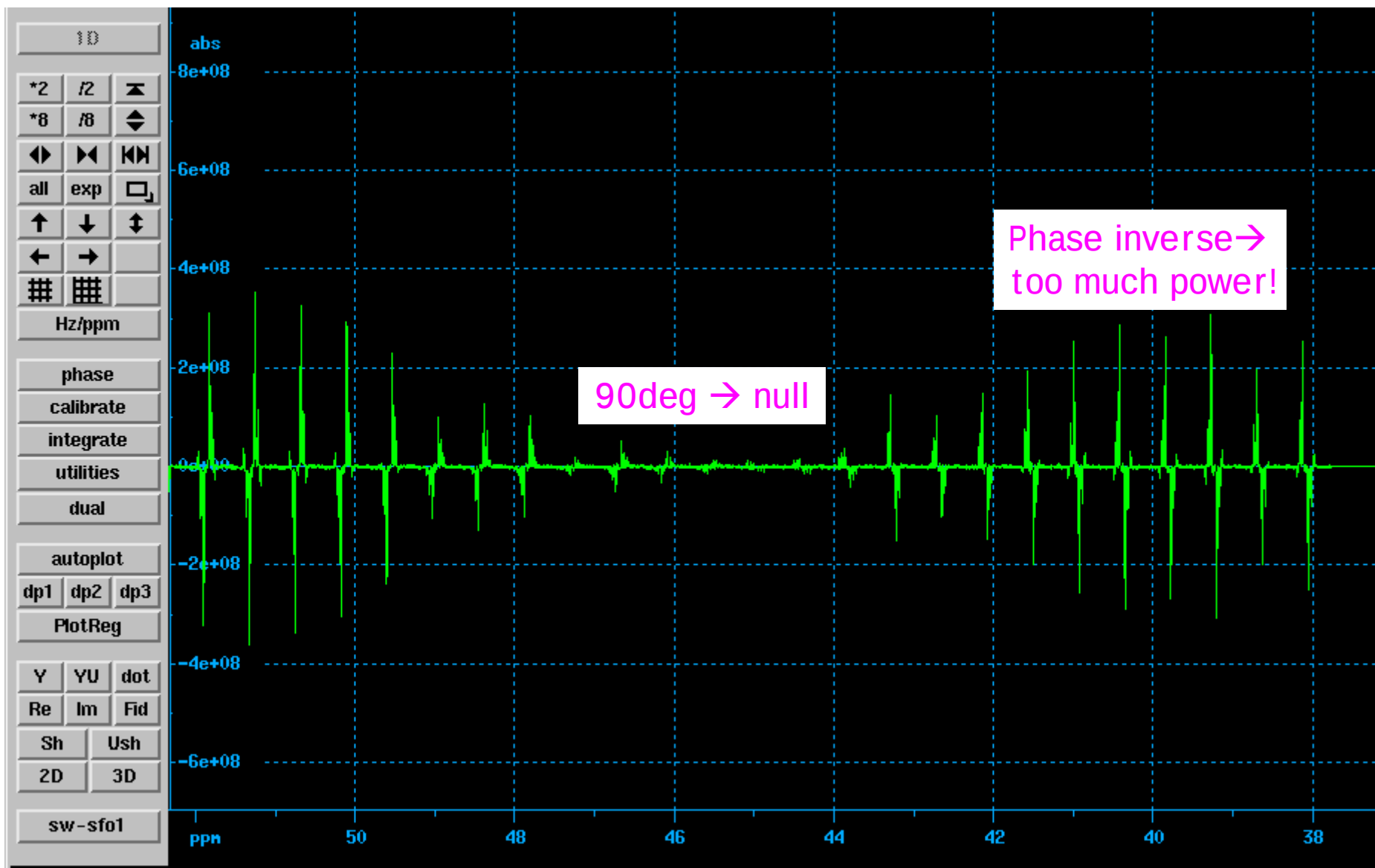
Decouple channel for 2H hard pulse (decouple902hf4)

Step 3: zg ,em, ft (pl4=10dB, p23=200~300us)



Decouple channel for 2H hard pulse (dec902hf4)

Step 4: zg, em, fp (fix p23, adjust pl4 until reach null)



2005 NMR Training Course Lab Session

分組名單

示範人員	游春愛、方珮如	吳文晉	張七鳳
儀器名稱	AV600	AV500	AV600(in Chem)
2:00 ~	何孟儒(陳金榜) 黃愈志(陳金榜) 楊玉良(吳世雄) 蔡丞坤(黃太煌) 吳順家(磊葳)	駱啓仁(林達顯) 王誌慶(林達顯) 邱垂霖(程家維) 劉育男(呂平江) 吳英彥(磊葳)	盧弘捷(甘魯生) 饒淑娟(生化所) 林素卿(陳長謙) 詹雅玲(錢偉鈞) 羅彥中(王惠鈞)

P.S.由於此次**Lab**上機課程以有經驗者優先, 初學者請於下次基礎課程再上機.謝謝合作 !!

2005 NMR Training Course Lab Session

Outline (you should know how to shim, wobb, 1H pulse calibration..... already)

1. Hard pulse calibration for $^{13}\text{C}/^{15}\text{N}$ (decouple channel)
 - a. rpar 1D ^1H experiment
 - b. edasp → eda (select pulse program) → ased (set up parameters, cnst=142 or 89)
 - c. Look for 90deg pulse
2. Shape pulse calibration for ^{13}C (decouple channel)
 - a. rpar 1D ^1H experiment
 - b. edasp → eda (select pulse program) → ased (set up parameters, cnst=142)
 - c. Look for 90deg or 180deg pulse
3. Standard HNC0 type experiment (hncogpwg3d)
 - a. rpar → getprosol → look for “users” parameters
 - b. double check “decouple pulses” which you obtain from (1) & (2)
4. New experiment from scratch

new → eda (change to hncogp3d, and other parameters) → getprosol
→ ased (set up parameters)