Two Dimensional Homonuclear Correlation Spectroscopy

Gradient COSY

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INTRODUCTION

Correlation Spectroscopy

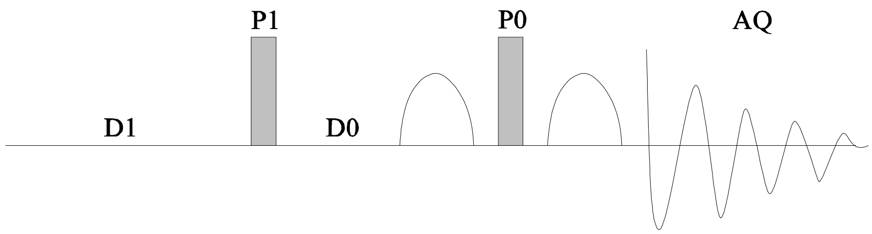
Correlation experiments reveal connections between spin coupled nuclei. Spin, or scalar, coupling between nuclei is established by the appearance of cross peaks (peaks off the diagonal) in the two-dimensional spectrum. Since spin coupled nuclei are usually separated by two or three bonds, the correlation experiment, by revealing the connectivity within the compound, is often enough to establish the chemical structure. Resonances which are coupled through four and five bonds will occasionally show cross peaks also. Although most correlation experiments are designed to emphasize short range coupling, there are experiments designed to emphasize long range correlations as well.

Two-dimensional spectra can be acquired in either of two modes. In the magnitude, or absolute valve mode, only the “real” part of the data is collected since the spectrum is not phased. In the phase sensitive mode, both the “real” and “imaginary” parts of the data are acquired so that the spectrum can be phased. An advantage of a phase sensitive spectrum is that the peaks are much narrower and the resolution greater. A disadvantage of a phase sensitive spectrum is that twice as much data must be recorded in order to extract the phase information. Thus, it takes twice the time to record a spectrum in phase sensitive mode.

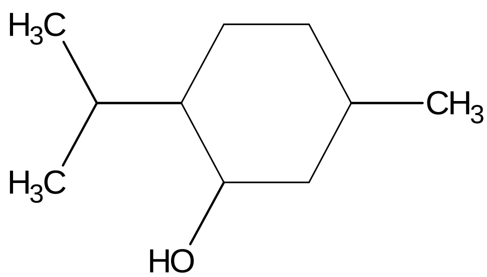
Gradient COSY

The experiment described here is the simplest of two-dimensional magnetic field gradient NMR experiments. Magnetic field gradients are sometimes called pulsed filed gradients, pfg. Normal COSY spectra require a minimum of four scans to distinguish the sign of a frequency in the second (F_1) dimension and select the desired coherence. If you require the peaks to have the correct shape (phase sensitive mode), you must collect eight scans. By introducing magnetic field gradients into the experiment, this can all be accomplished in one scan. Gradient methods can significantly shorten the time it takes to acquire a two-dimensional spectrum, provided you have a concentrated enough sample. An additional benefit of gradients is a significantly reduced t_1 noise. Gradient COSY spectra can be acquired in both magnitude and phase sensitive mode.

The pulse sequence for gradient-COSY is shown below.



The gradient COSY spectrum of Menthol is shown on a following page. The spectrum required about 11 minutes to collect.



Menthol

References.

1) **150** and More Basic NMR Experiments, A practical Course, S. Braun, H.-O. Kalinowski, S. Berger, Wiley-VCH, 1998, pages 477-480.

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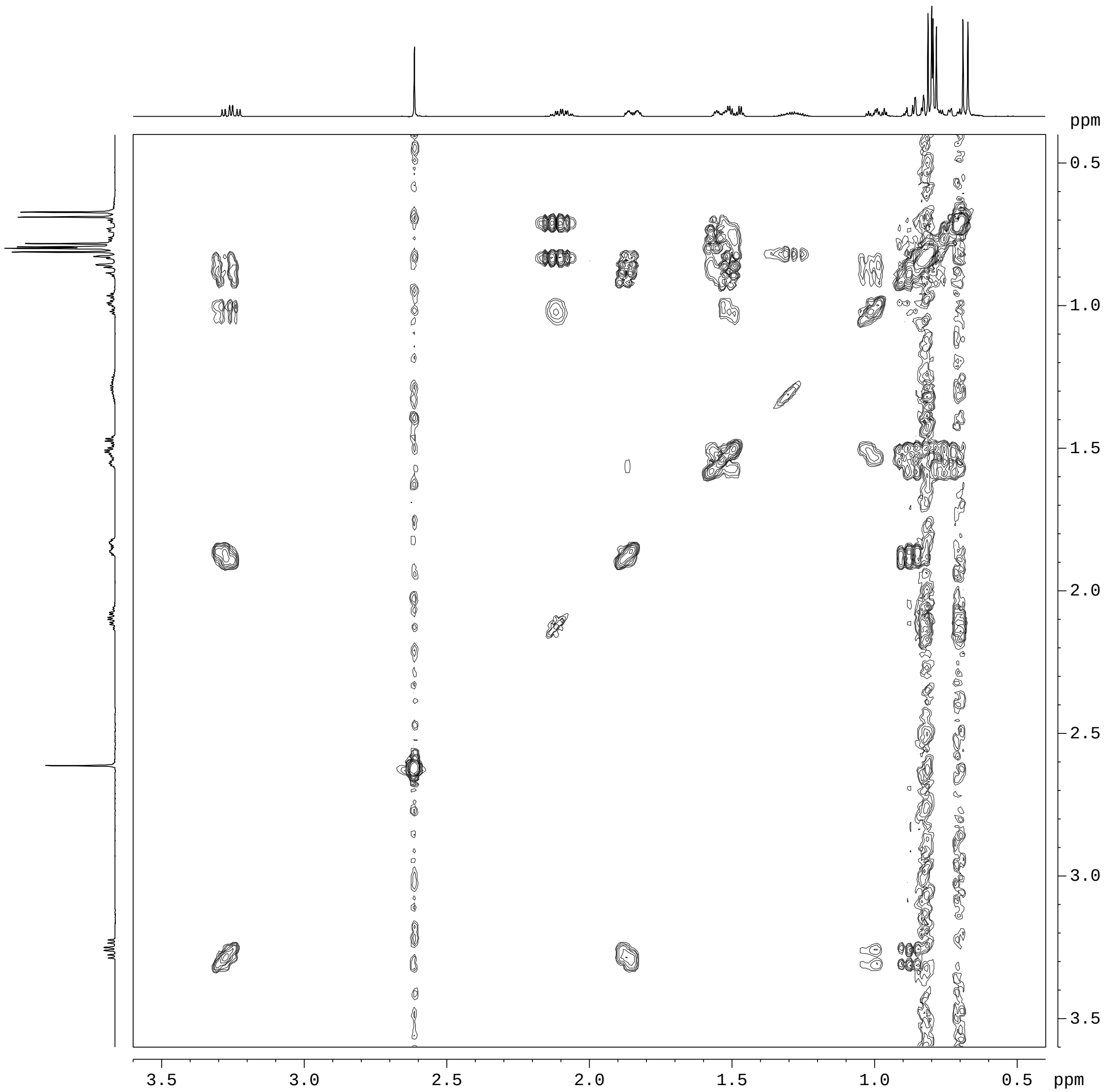


```
Current Data Parameters
NAME        Menthol_COSY
EXPNO                  5
PROCNO                 1

F2 - Acquisition Parameter
Date_             990223
Time               13.09
INSTRUM            spect
PROBHD     5 mm QNP  1H
PULPROG           cosygp
TD                  1024
SOLVENT            CDCl3
NS                     1
DS                    16
SWH             2003.205 Hz
FIDRES          1.956255 Hz
AQ             0.2556404 se
RG                     8
DW               249.600 us
DE                  6.00 us
TE                 300.0 K
d0            0.00000300 se
d13           0.00000300 se
D1            1.00000000 se
P1                  7.40 us
SFO1         400.1308094 MH
NUC1                  1H
PL1                -3.00 dB
P16              1000.00 us
GPX1                0.00 %
GPY1                0.00 %
GPZ1               10.00 %
GPNAM1         sine.100
D16           0.00025000 se
P0                  7.40 us
GPX2                0.00 %
GPY2                0.00 %
GPZ2               10.00 %
GPNAM2         sine.100
IN0           0.00049920 se

F1 - Acquisition parameter
ND0                    1
TD                   256
SFO1            400.1308 MH
FIDRES          7.825020 Hz
SW                 5.006 pp

F2 - Processing parameters
SI                   512
SF           400.1300092 MH
WDW                 SINE
SSB                    0
LB                  0.00 Hz
GB                     0
PC                  1.00

F1 - Processing parameters
SI                   512
MC2                   QF
SF           400.1300092 MH
WDW                 SINE
SSB                    0
LB                  0.00 Hz
GB                     0

2D NMR plot parameters
CX2                15.00 cm
CX1                15.00 cm
F2PLO              3.604 pp
F2LO             1441.94 Hz
F2PHI              0.396 pp
F2HI              158.63 Hz
F1PLO              3.604 pp
F1LO             1441.94 Hz
F1PHI              0.396 pp
F1HI              158.63 Hz
F2PPMCM          0.21381 pp
F2HZCM          85.55355 Hz
F1PPMCM          0.21381 pp
F1HZCM          85.55355 Hz
```

Gradient COSY of Menthol

EXPERIMENTAL SETUP

The concentration of the sample should be high enough that you can record a high signal to noise ^{1}H NMR spectrum in 16 - 32 scans.

Record a ^{1}H spectrum.

NEW (or EDC)	Create a new data set for your sample.
NAME name	Data set name.
EXPNO 1	Experiment number (must be 1).
PROCNO 1	Process data number (must be 1).
[SAVE]	Save the data set.
RPAR +proton all	Read in the standard proton parameters.
NS, etc.	Adjust NS and other parameters as necessary.
RGA, ZG	Acquire some data.
FT, APK, ref	Fourier transform, phase and reference the spectrum.
Zoom	Zoom in on the region of interest. The expanded region need not contain a reference peak.
[sw-sfo1]	Set the sweep width and spectrometer frequency to cover the zoomed region. Reduce TD if the acquisition time (AQ) is unnecessarily large.
RGA, ZG	Acquire a spectrum of the zoomed region.
FT, etc.	Fourier transform, phase etc.

Setting the conditions for ^{1}H-^{1}H correlation.

The following AU program sets up all of the parameters for gradient cosy.

XAU su_gcosy	Set up gradient cosy (g cosy).
[OK]	Answer “Delete `meta.ext` files ?” with [OK].

The AU program sets the following file parameters. Use EDC to display the results.

File parameter		
EXPNO	1 5	Experiment number 1, for 1-D proton. Experiment number 5, for g cosy.

The AU program sets the following acquisition parameters. Use EDA to display the results.

Acquisition Parameters		
Time domain 2 (F2)		
PULPROG	cosygp	PULse PROGram for m cosy.
TD	1024	Time Domain points.
NS	1	Number of Scans (should be 1, 2, 4, 8 etc.).
DS	16	number of Dummy Scans for 1st row.
D1	2 sec	relaxation Delay.
GPZ1	10.0	strength of the first Gradient Pulse along Z.
GPZ2	10.0	strength of the second Gradient Pulse along Z.
Time domain 1 (F1)		
TD	256	Time Domain points.
ND0	1	Number of D0 periods per cycle.
IN0	1 / SW	Increment in t1 (calculated).
SW		SW of ^{1}H spectrum (same as for F2).
SFO1		^{1}H frequency (same as for F2).

ASED — Check that acquisition parameters are set correctly. A brief description of other parameters, not described above, is given in the pulse program at the end of this document.

[SAVE] — Save the acquisition parameters.

<u>EXPT</u> — Calculates the approximate length of time to do the experiment. This may help you to decide if you want to collect more or fewer slices or scans or points etc. The Menthol spectrum required 11 minutes.

DATA ACQUISITION

[Spin on/off]	TURN THE SPINNER OFF (press the Spin On/Off button on the BOSS keyboard).
ZG	Start the acquisition.

DATA PROCESSING

The AU program sets the following processing parameters. Use EDP to check or modify the values.

Processing Parameters		
Time/frequency domain 2 (F2)		
SI	512	the SIze in F2 (zero fill rows).
WDW	SINE	Sine multipication.
SSB	0	Unshifted sine bell.
PH_mod	NO	No phase correction.
PKNL	TRUE	Required with digital filter.
BC_mod	QUAD	Background correct quadrature data.
Time/frequency domain 1 (F1)		
SI	512	the SIze in F1 (zero fill columns).
WDW	SINE	Sine multipication.
SSB	0	Unshifted sine bell.
PH_mod	MC	Magnitude calculation.
BC_mod	NO	No background correction.
MC2	QF	Forward quadrature FFT.
OFFSET		Frequency offset (same as for F2).
SF		Spectrometer frequency (same as for F2).

XFB	Background correct, window, zero fill, Fourier transform, phase and reference. The whole kaboodle in one command. It is OK to execute this command on a partial data set, during acquisition.

2-D CONTOUR DISPLAY

X-Y Expansion the spectrum.

[Limits]	Set the limits of the plot region. NOTE: For homo-nuclear correlation experiments, (COSY, NOESY, etc.) set F1LO = F2LO and F1HI = F2HI so the display will be symmetric and the diagonal peaks will appear on the diagonal line.
[PlotReg]	Forces XWinNMR to display the plot region.

Setting the intensity scale.

[DefPlot]	Sets the intensity scale for plotting to be the same as the currently displayed intensity scale.
[contours]	Displays the equi-intensity contours of the data.
[intensities]	Displays ranges of intensity as a color map.

PLOTTING

The AU program sets the following plotting parameters. Use EDG to check or modify the values. Once EDG has started, press either the [EDPROJ2] or [EDPROJ1] buttons.

Plotting Parameters		
Projection for Frequency domain 2		
PF2DU	z	Disk Unit of the data set.
PF2USER	username	Your user name.
PF2NAME	name	Name of the data set.
PF2EXP	1	Experiment number of the ^{1}H spectrum.
PF2PROC	1	Process number of the ^{1}H spectrum.
Projection for Frequency domain 1		
PF1DU	z	Disk Unit of the data set.
PF1USER	username	Your user name.
PF1NAME	name	Name of the data set.
PF1EXP	1	Experiment number of the ^{1}H spectrum.
PF1PROC	1	Process number of the ^{1}H spectrum.
Additional plotting parameters		
PF1CY	2.5	The value of CY for the F1 projection. This can be used to increase or decrease the height of the spectrum displayed in the F1 projection. (PF1CY = 5.0 will double the height of the 1-D spectrum).
PF2CY	2.5	The value of CY for the F2 projection.

SETTI — Set the title for the 2D spectrum.

PLOT — Plot the contours of the 2D spectrum along with the hi-resolution ^{1}H spectrum in experiment #1.

Additional Processing Parameters		
Frequency domain		
F1LO	bbb	Set to the value for the bottom of the spectrum (ppm).
F1HI	ttt	Set to the value for the top of the spectrum (ppm).
F2LO	lll	Set to the value for the left side of the spectrum (ppm).
F2HI	rrr	Set to the value for the right side of the spectrum (ppm).

Additional Processing Commands	
Frequency domain	
ABS1	Automatic Baseline Subtraction for F1 (uses F1LO, F1HI).
ABS2	Automatic Baseline Subtraction for F2 (uses F2LO, F2HI).
SYM	Symmetrize the spectrum. This “averages” the upper and lower triangles of the matrix. CAUTION! This is an easy way to introduce artifacts into your spectrum.

Pulse Program for Gradient COSY

```
;cosygp
;avance-version
;2D homonuclear shift correlation
;using gradient pulses for selection

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

"d0=3u"
"d13=3u"

1 ze
2 d1
3 p1 ph1
  d0
  50u UNBLKGRAD
  p16:gp1
  d16
  p0 ph2
  d13
  p16:gp2
  d16
  4u BLKGRAD
  go=2 ph31
  d1 wr #0 if #0 id0 zd
  lo to 3 times td1
exit

ph1=0 2
ph2=0 0 2 2
ph31=0 2

;pl1 : f1 channel - power level for pulse (default)
;p0 : f1 channel -  20 to 90 degree high power pulse
;p1 : f1 channel -  90 degree high power pulse
;p16: homospoil/gradient pulse
;d0 : incremented delay (2D)                          [3 usec]
;d1 : relaxation delay; 1-5 * T1
;d13: short delay                                     [3 usec]
;d16: delay for homospoil/gradient recovery
;in0: 1/(1 * SW) = 2 * DW
;nd0: 1
;NS: 1 * n
;DS: 16
;td1: number of experiments
;MC2: QF
;use gradient ratio:       gp 1 : gp 2
;                            10 :   10
```