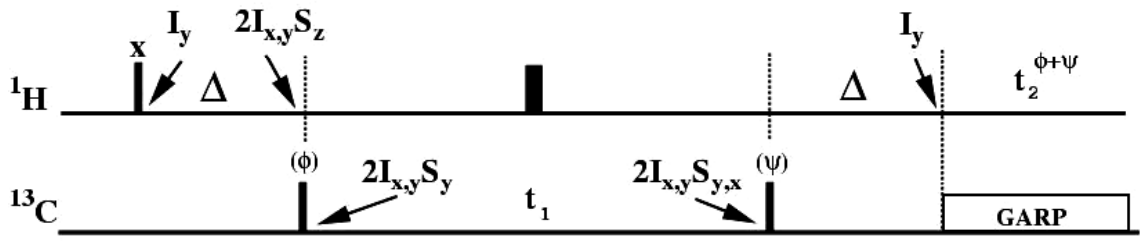
Basic heteronuclear correlations: HMQC

HMQC: heteronuclear multiple quantum correlation

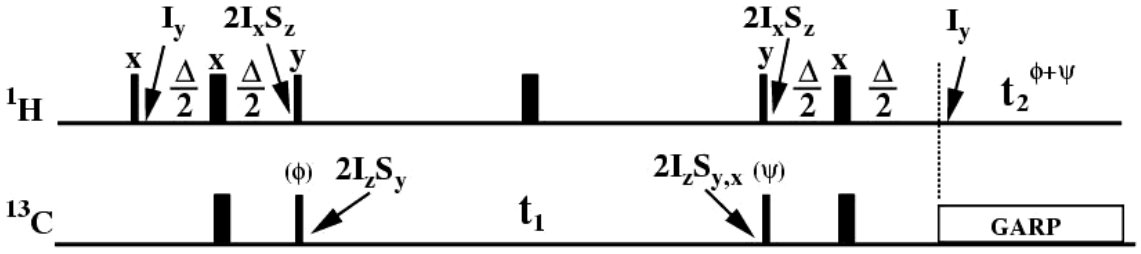


$$\phi = x\ -x$$
$$\psi = x\ \ x\ -x\ -x$$
$$\phi_{rec} = x\ -x\ -x\ \ x$$

- δ(^{1}H): refocused
- δ(^{13}C): evolution during t_1
- J(^{1}H,^{13}C): active during Δ
- J(H,H): active !
- J(C,C): active !
- Relaxation during t_1: multiple quantum line-narrowing

Basic heteronuclear correlations: HSQC

HSQC: heteronuclear single quantum correlation



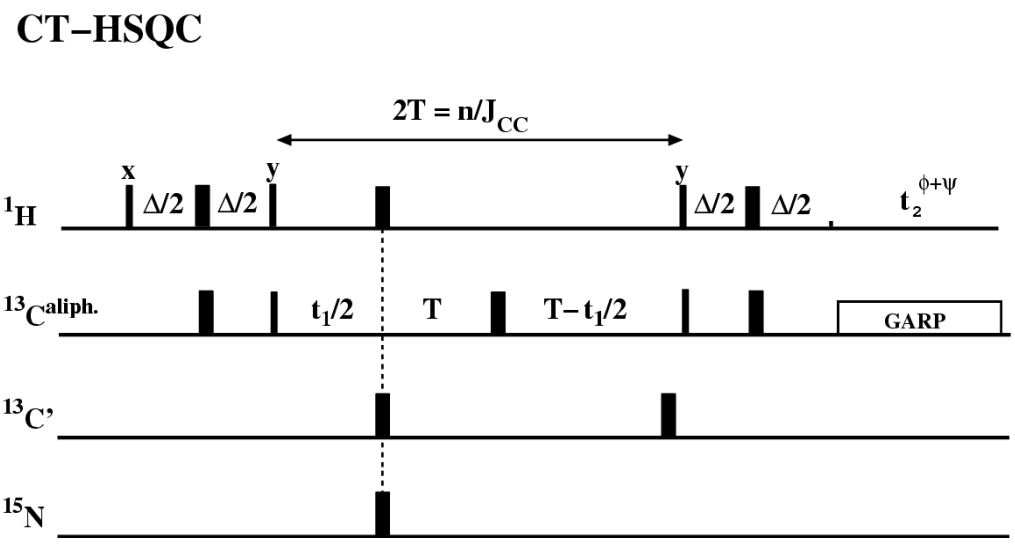
$$\phi = x\ -x$$
$$\psi = x\ \ x\ -x\ -x$$
$$\phi_{rec} = x\ -x\ -x\ \ x$$

- δ(^{1}H): refocused
- δ(^{13}C): evolution during t_1
- J(^{1}H,^{13}C): active during Δ
- J(H,H): not active
- J(C,C): active !
- Relaxation during t_1: T1(^{1}H), T2(^{13}C)

Constant-time HSQC

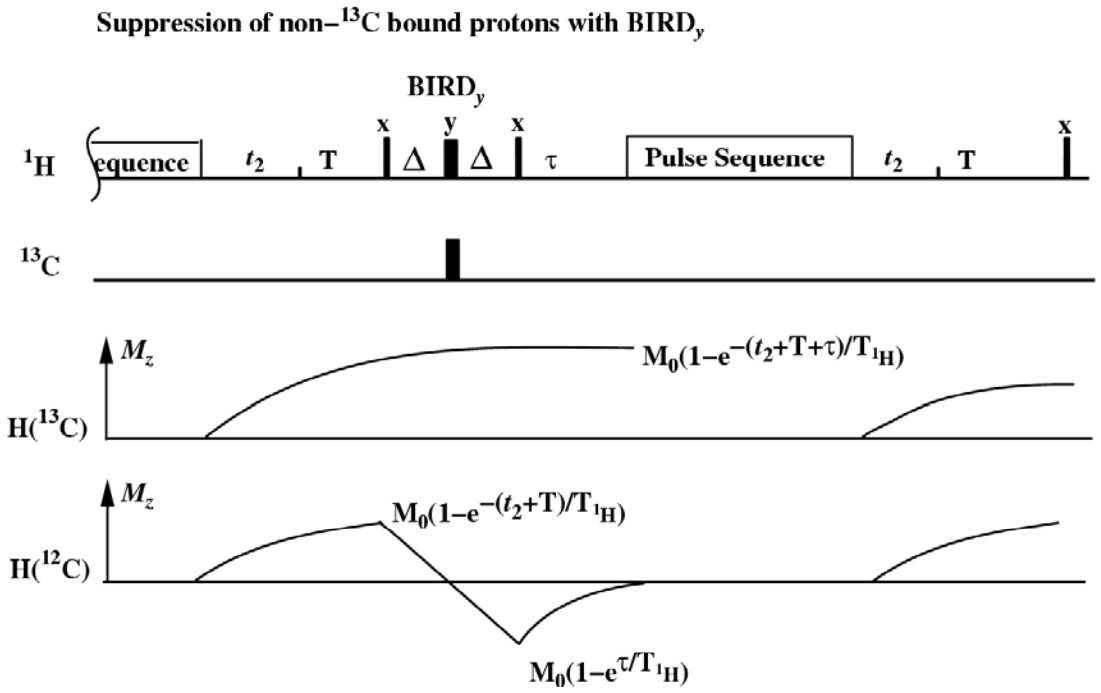
➔ Set $2T = n/J_{CC}$ to refocus evolution of homonuclear C,C couplings during 2T

➔ J-coupling evolution: $\cos(\pi J_{CC} 2T)^n = -1^n$



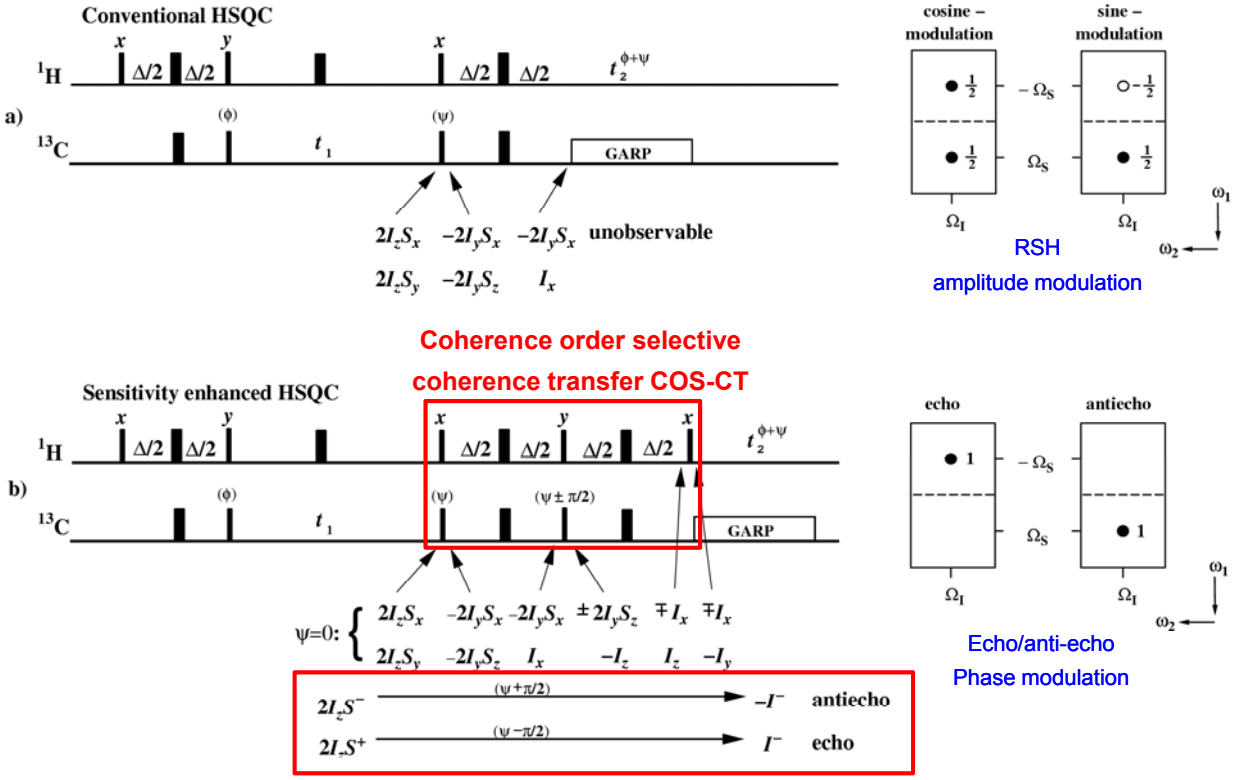
BIRD filter to suppress ^{12}C magnetization

Excellent ^{12}C suppression and fast acquisition (small, unlabeled molecules!)

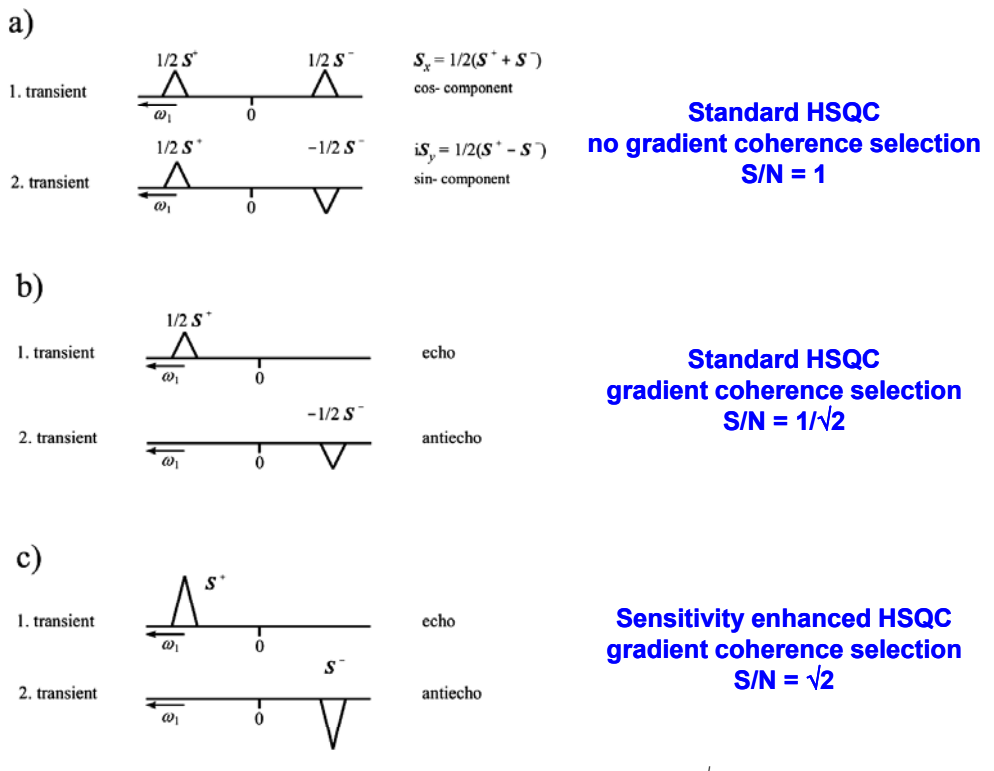


Bax Subramanian JMR (1986) 67, 565-9
Sattler et al JACS (1992) 114, 1126-7.

Sensitivity enhancement



Sensitivity enhancement / gradient coherence selection



S/N=: I/√n (I: Σ(intensities), n: # signals)

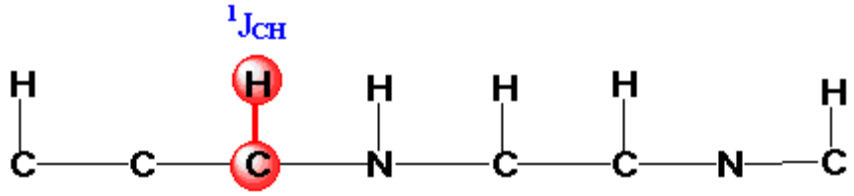
Go to Tutorial
Go to SpecWizard

2D Experiments

2D HSQC

DESCRIPTION

The **2D HSQC (Heteronuclear Single-Quantum Correlation) experiment** permits to obtain a 2D heteronuclear chemical shift correlation map between directly-bonded 1H and X-heteronuclei (commonly, 13C and 15N) . It is widely used because it is based on proton-detection, offering high sensitivity when compared with the conventional carbon-detected 2D HETCOR experiment. Similar results are obtained using the 2D HMQC experiment.



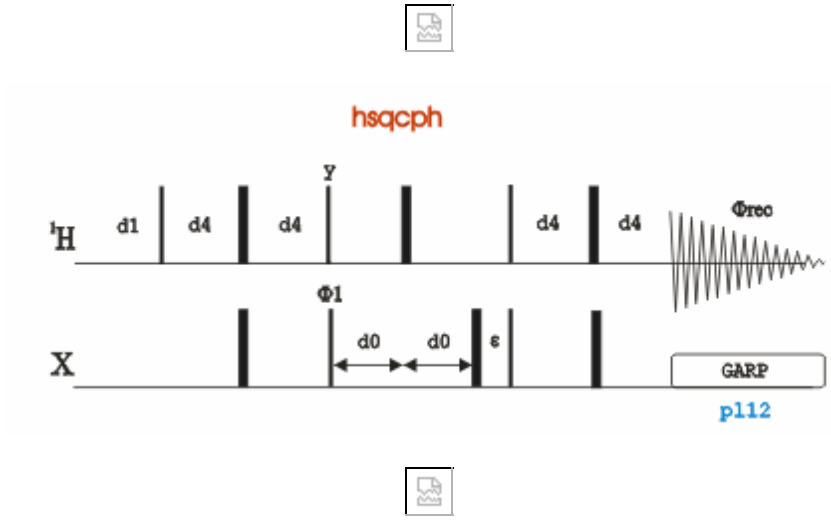
REQUIREMENTS

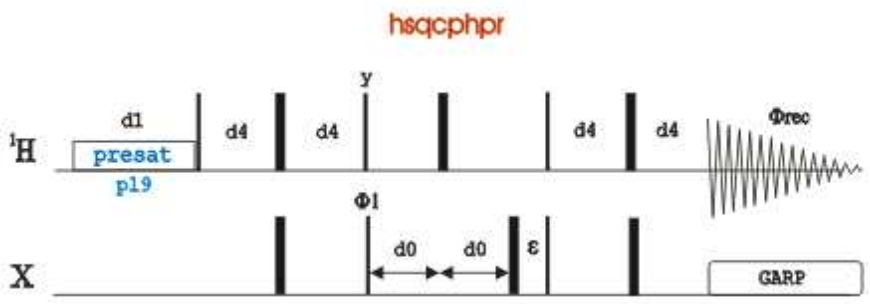
Easy implementation on any AVANCE spectrometer equipped with an inverse probehead

VERSIONS

The basic 2D HSQC pulse sequence consists of four independent parts (**80CPL185** and **86JMR546-69**):

1. An **initial INEPT pulse train** transfers polarization from 1H to X via 1J(XH) (see INEPT block).
2. The antiphase 13C magnetization evolves during the **variable evolution t1 period** under the effect of X chemical shift. Heteronuclear 1H-X couplings are refocused by applying a 180º 1H pulse at the middle of this period (see 1H-decoupled X evolution block).
3. A **retro-INEPT pulse train** converts X magnetization to in-phase 1H magnetization (see reverse-INEPT block).
4. **Proton acquisition** is performed with X decoupling (see 1H-acquisition with X-decoupling).





A number of alternatives have been proposed:

- Use of spin-lock pulses for purging undesired coherences (**88JMR569-76** and **89JMR608-85** and **91JMR394-94**).
- Improved sensitivity can be obtained using the PEP methodology (**91JMR151-93** and **93AR1**). A second retro-INEPT pulse train is inserted prior to acquisition in order to select orthogonal components evolving during the t1 period.
- Multiplicity-editing by inserting an heteronuclear spin-echo during the t1 period (**90JMR589-90** and **91JMR665-91**).
- The decoupled HSQC (or Double INEPT) experiment in which the initial INEPT and the last retro-INEPT pulse trains are replaced by refocused INEPT pulse trains in order to achieve in-phase carbon magnetization during the t1 period (**90JMR304-86** and **90JMR488-87**).
- Constant-Time HSQC experiment to remove 13C-13C scalar coupling during the t1 period.
- Gradient-enhanced versions (see ge-2D HSQC experiment).
- Removing axial peaks artifacts by modified phase cycling (**94JMRA246-109** and **96JMRB91-110**).
- Semi-selective versions to achieve better resolution in the indirect dimension (see Semi-selective HSQC experiment)

EXPERIMENTAL DETAILS

The 2D HSQC experiment can be recorded in routine/automation modes. Minor changes are required if a predefined parameter set is available.The interpulse d2 delay is optimized to 1/2*JCH (3.3-3.8 ms).

More details on practical implementation of ge-2D HSQC experiments on AVANCE spectrometers can be found in the corresponding tutorials:

- **2D HSQC experiment (phase-sensitive)**
- **2D HSQC experiment (phase-sensitive with presaturation)**

SPECTRA

The HSQC spectrum correlates chemical shifts of heteronucleus X (F1 dimension) and protons (F2 dimension) via the direct heteronuclear coupling 1J(XH). Carbon decoupling is usually performed during proton acquisition and the corresponding satellites colapse to a single resonance showing all proton-proton couplings.

See spectra

NMRSIM: 2D HSQC

RELATED TOPICS

Comparison of different 2D inverse correlations have been carried out (**90JMR304-86** and **90JMR488-87**).

Related sequences:

- 2D HSMQC experiment (**90JMR346-86**).
- 2D Double DEPT sequence (**89METH134** and **91JMR151-93**).
- 2D Inverse experiments
- 2D Inverse gradient-enhanced experiments

More about HSQC ....

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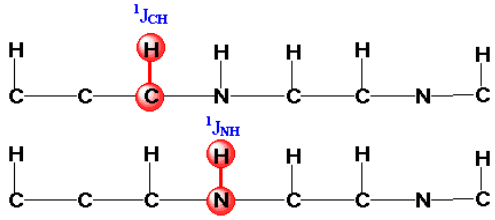
Go to Tutorial
Go to Experiment Wizard

2D Experiments
ge-2D HSQC

DESCRIPTION

The **ge-2D HSQC experiment** is the gradient-enhanced version of the conventional 2D HSQC experiment in which coherence selection is achieved by means of PFG. Thus, clean 2D HSQC spectra can be recorded in a single scan per t_1 increment without need for phase cycle when sample concentration is high. Other advantages are the optimal dynamic range, improved water and artefact suppression, and reduced t_1 noise in the minimally required experiment time. The HSQC experiment allows to trace out directly bonded ^{1}H-X pairs via the large $^1J_{HX}$ coupling constant



REQUIREMENTS

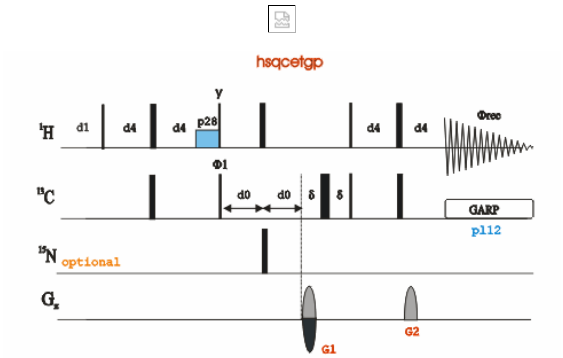
Easy implementation on any AVANCE spectrometer equipped with pulsed field gradients (PFGs) and inverse probehead.

VERSIONS

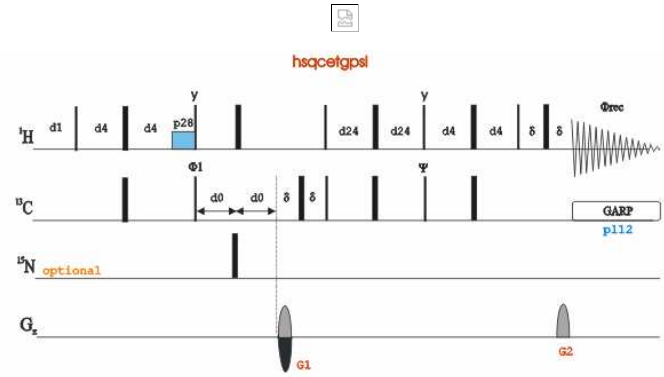
In addition of the advantages to use PFGs, an important aspect when PFG are incorporated in the HSQC pulse sequence, are the *sensitivity* and *resolution* requirements. The effect on the sensitivity of several gradient-based HSQC and other related schemes have been extensively discussed (95JB11-6 , 95JMRB235-108 , 94JMRA70-111 , 96JMRA64-122 , 94JB301 , 93ANG1489 , and 96JMRA17-1191). It is possible, for instance, to design six different basic versions of the 2D ^{1}H-X HSQC experiments using PFGs. The use of each version will depend on the sample under study.

- The dephasing gradient G1 is applied into the variable t_1 period, obtaining a magnitude mode spectrum (91JACS9688 and 92JMR282-100). This is an excellent option for routine samples in which sensitivity and resolution are not critical. In this case, two gradients with a 4:1 ratio select N-type data (shaded line corresponding to a p_1=+1) whereas a 4:-1 ratio would select P-type data (continuous line corresponding to a p_1=-1).

- The echo-antiecho version of this experiment uses the same sequence, but the intensity of the refocusing gradient G_2 is inverted on alternated scans to obtain the N- and P-type data separately (92JMR207-98 and 92JMR660-98). After proper processing, this approach allows for obtain phase-sensitive spectra but with sensitivity losses by a factor of square(2) with respect to the classical phase-cycled experiment.



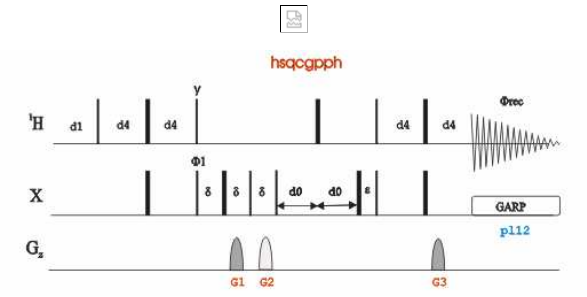
- Use of the PEP methodology (91JMR429-91 , 91JMR151-93 , and 93AR1). This is the best option to record phase-sensitive 2D HSQC spectra with maximum sensitivity (92JACS10663). The selection procedure use the same principles described for the echo-antiecho approach, but the pulse sequence must be modified adding a second retro-INEPT block in order to select both orthogonal components of the magnetization (I_zS_x and I_zS_y) present during t_1. This basic scheme is widely applied to improve the sensitivity in other related multidimensional experiments (95JB11-6 , 95JMRB235-108 , 94JB301 , 93ANG1489 , and 96JMRA171-119).



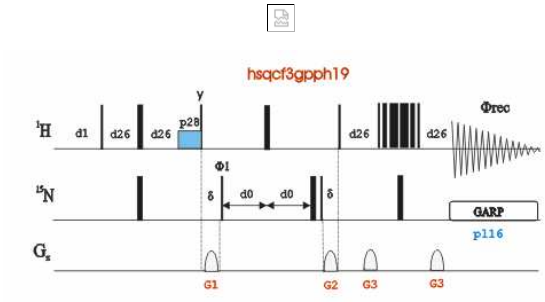
In triple resonance experiments applied to labeled proteins, sensitivity can be further enhanced by simultaneous acquisition (95JB97). Some examples of modified PEP-HSQC experiments have been described to measure accurate 1JNH (96JMRB245-112 and 96JMRB269-112), measure a set of relaxation parameters in ^{15}N-^{1}H spin systems (96JMRB245-112-111 and 96JMRB269-101) or to observe exchange broadened signals (96JB223).

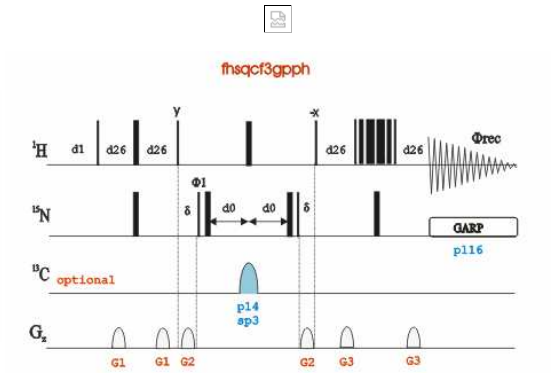
- A X filter-z (consisting of 90°(X)-PFG-90°(X)) is applied between the t_1 period and the dephasing G_1 gradient (93MRC287). A phase-sensitive spectrum is obtained using the classical acquisition and processing procedures (for instance, TPPI), but theoretical sensitivity loss by a factor of two is obtained compared to the phase-cycled experiment. However, this loss can be partially recovered applying the PEP methodology (a second retro-INEPT block shifted 90º in phase) without the need to apply the echo-antiecho approach (

95JMRB285-108).



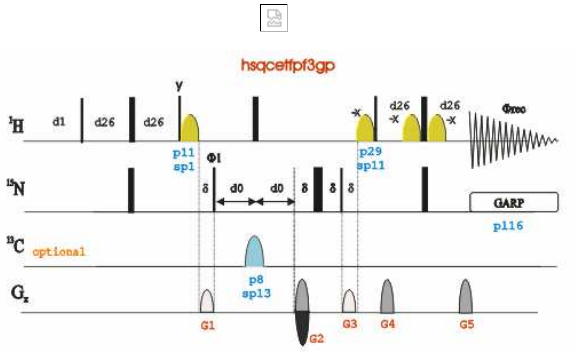
- Use of PFG as purge elements in the original phase cycle sequence. These PFG are placed between the simultaneous 90° pulses of ^{1}H and X in order to select the corresponding IzSz magnetization (93JMRA113-101 and 93JMRB239-102). Although this is not a pure selection procedure, this approach reduces the number of phase cycle steps and minimizes the suppression artefacts without affecting the overall sensitivity with comparison to the phase cycled analog.
- Much attention has been given to the effect of water suppression on the signal intensity of relatively rapidly exchanging protons, as the NH protons in proteins. As a general approach, the WATERGATE block (92JB661 and 93JMRA241-102) is usually applied during the retro-INEPT block of the standard HSQC pulse sequence in order to improve solvent suppression in aqueous samples.

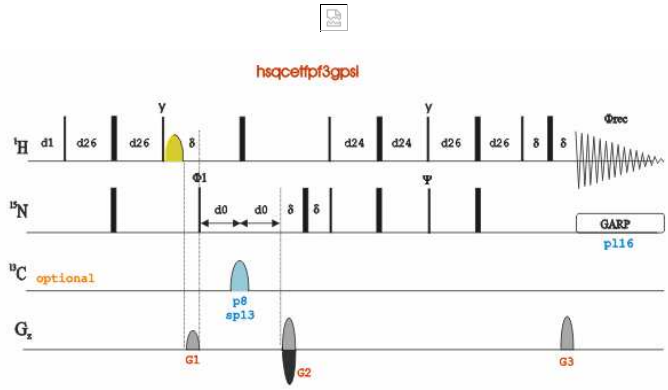


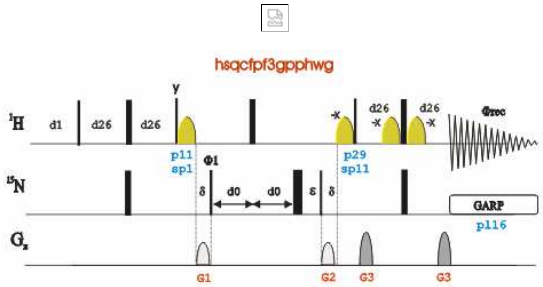


WATERGATE can also be combined with the water flip-back approach (93JACS12593) which ensures that the water magnetization is little perturbed and oriented along the +z axis during most of the experiment, especially just prior to acquisition, to minimize the saturation of water. This is commonly applied to proteins and nucleic acids dissolved in H_2O.

Other related approaches have been proposed to avoid the saturation transfer from water in HSQC experiments (93JMRB315-101 , 94JMRA178-107 , 94JMRB45-105 , 95JMRB94-108 , and 96JACS5510).







Other related versions:

- Simultaneous acquisition of ^{1}H-^{13}C and ^{1}H-^{15}N 2D HSQC spectra can be obtained using the *Time-Shared* method
- Incorporation of an X-filter prior to acquisition to remove ABX strong coupling signals (99JMR89-138)
- Improved resolution in the F_1 dimension can be achieved by applying a band-selective carbon pulse ((see Semi-selective HSQC experiment). This approach can be applied in any version of the mentioned HSQC experiment.

EXPERIMENTAL DETAILS

The ge-2D HSQC experiment is usually recorded in routine and automation modes. Minor changes are required if a predefined parameter set is available. The user must define the strength, the duration, and the shape of the gradients and the recovery delay.

More details on practical implementation of ge-2D HSQC experiments on AVANCE spectrometers can be found in the corresponding tutorials:

- Tutorials: 2D inverse experiments
- Tutorials: 2D gradient-based inverse experiments

SPECTRA

The HSQC spectrum correlates chemical shifts of heteronucleus X (F_2 dimension) and protons (F_1 dimension) via the direct heteronuclear coupling $^1J(XH)$. The effective suppression of unwanted ^1H-^{12}C or ^1H-^{14}N magnetization by means of PFGs allows to obtain ultra-clean 2D spectra from which clear analysis can be done.

See Some Examples

RELATED TOPICS

Related experiments:

- 2D Inverse experiments
- 2D Inverse gradient-enhanced experiments

More about 2D HSQC ....

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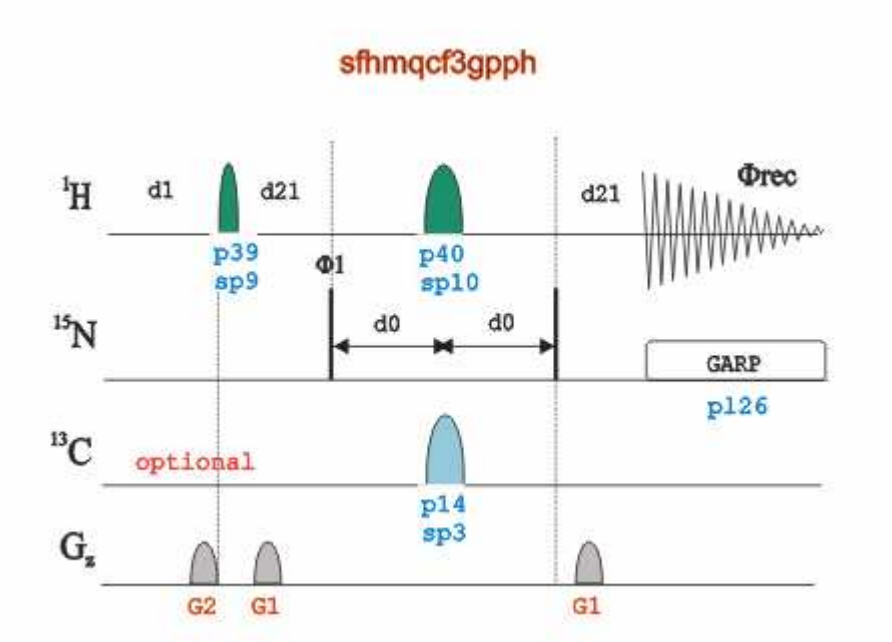
Previous pp:**seqtrhncagp3d**
Next pp:**shmbcgpndqf**

Pulse Programs

NMRSIM Pulse Diagram
Relevant parameters: **ased**
More info on **sfhmqcf3gpph**

Pulse Diagram



Pulse Program

```
;sfhmqcf3gpph
;avance-version (09/10/26)
;SOFAST HMQC
;2D H-1/X correlation via heteronuclear zero and double quantum
; coherence
;phase sensitive
;with decoupling during acquisition
;
;P.Schanda and B. Brutscher, J. Am. Chem. Soc. 127, 8014 (2005)
;
;$CLASS=HighRes
;$DIM=2D
;$TYPE=
;$SUBTYPE=
;$COMMENT=


prosol relations=<triple>


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


"d11=30m"
"d12=20u"
"d21=1s/(cnst4*2)"


"in0=inf1"
```

"d0=in0/2-p21*4/3.1415"

"DELTA1=d21-p16-d16-p39*cnst39"
"DELTA2=p39*cnst39-de-4u"

"spoff23=bf1*(cnst19/1000000)-o1"
"spoff24=bf1*(cnst19/1000000)-o1"

1 ze
d11 pl26:f3
2 d1 do:f3
3 d12 pl3:f3
50u UNBLKGRAD

p16:gp2
d16

(p39:sp23 ph1):f1
p16:gp1
d16

ifdef LABEL_CN
(center (p40:sp24 ph2):f1 (p8:sp13 ph1):f2 (DELTA1 p21 ph3 d0 p21 ph4 DELTA1):f3)
else
(center (p40:sp24 ph2):f1 (DELTA1 p21 ph3 d0 p21 ph4 DELTA1):f3)
endif /*LABEL_CN*/

DELTA2
p16:gp1
d16 pl26:f3
4u BLKGRAD
go=2 ph31 cpd3:f3
d1 do:f3 mc #0 to 2
F1PH(calph(ph3, +90), caldel(d0, +in0))
exit

ph1=0
ph2=0
ph3=0 2
ph4=0 0 2 2
ph31=0 2 2 0

;pl3 : f3 channel - power level for pulse (default)
;pl26: f3 channel - power level for CPD/BB decoupling (low power)
;sp13: f2 channel - shaped pulse 180 degree (adiabatic)
;sp23: f1 channel - shaped pulse 120 degree
; (Pc9_4_120.1000 or Q5.1000)
;sp24: f1 channel - shaped pulse 180 degree (Rsnob.1000)
;p8 : f2 channel - 180 degree shaped pulse for inversion (adiabatic)
;p16: homospoil/gradient pulse [1 msec]
;p21: f3 channel - 90 degree high power pulse
;p39: f1 channel - 120 degree shaped pulse for excitation
; Pc9_4_120.1000 (120o) (3.0ms at 600.13 MHz)
; (or Q5.1000 (90o) (2.0ms at 600.13 MHz))
;p40: f1 channel - 180 degree shaped pulse for refocussing
; Rsnob.1000 (1.0ms at 600.13 MHz)

```
;d0 : incremented delay (2D) = in0/2-p21*4/3.1415
;d1 : relaxation delay
;d11: delay for disk I/O [30 msec]
;d12: delay for power switching [20 usec]
;d16: delay for homospoil/gradient recovery
;d21 : 1/(2J)NH
;cnst4: = J(NH)
;cnst19: H(N) chemical shift (offset, in ppm)
;cnst39: compensation of chemical shift evolution during p39
; Pc9_4_120.1000: 0.529
; Q5.1000: -0.07
;inf1: 1/SW(N) = 2 * DW(N)
;in0: 1/ SW(N) = 2 * DW(N)
;nd0: 1
;NS: 2 * n
;DS: 16
;aq: <= 50 msec
;td1: number of experiments
;FnMODE: States-TPPI, TPPI, States or QSEC
;cpd3: decoupling according to sequence defined by cpdprg3: garp4.p62
;pcpd3: f3 channel - 90 degree pulse for decoupling sequence
; use pulse of >= 350 usec


;use gradient ratio: gp 1 : gp 2
; 11 : 7


;for z-only gradients:
;gpz1: 11%
;gpz2: 7%

;use gradient files:
;gpnam1: SMSQ10.100
;gpnam2: SMSQ10.100


;preprocessor-flags-start
;LABEL_CN: for C-13 and N-15 labeled samples start experiment with
; option -DLABEL_CN (eda: ZGOPTNS)
;preprocessor-flags-end


;Processing

;PHC0(F1): 90
;PHC1(F1): -180
;FCOR(F1): 1



;$Id: sfhmqcf3gpph,v 1.8.2.2 2009/12/14 12:35:13 ber Exp $
```

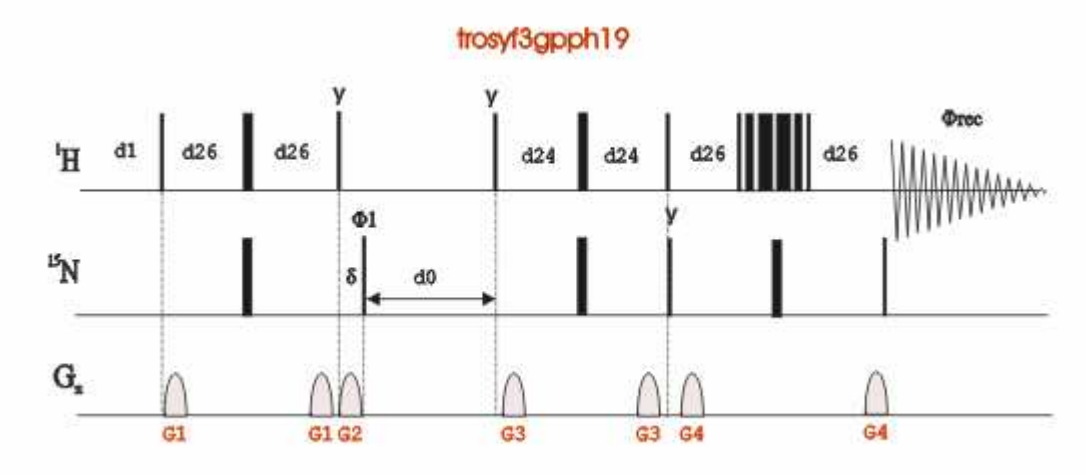
NMRGuide 4.3 - TOPSPIN 3.0
Written by Teodor Parella

Previous pp:**trosyf3gpidphwg**
Next pp:**trosyf3gpphsi19**

Pulse Programs

NMRSIM Pulse Diagram
Relevant parameters: **ased**
More info on **trosyf3gpph19**

Pulse Diagram



Pulse Program

```
;trosyf3gpph19
;avance-version (09/04/17)
;2D H-1/X correlation via TROSY
;phase sensitive
;using f3 - channel
;water suppression using 3-9-19 pulse sequence with gradients
;(use parameterset TROSYF3GPPH19)
;
;K. Pervushin, R. Riek, G. Wider & K.Wuethrich, Proc. Natl. Acad.
; Sci. USA, 12366-12371 (1997)
;M. Piotto, V. Saudek & V. Sklenar, J. Biomol. NMR 2, 661 - 666 (1992)
;V. Sklenar, M. Piotto, R. Leppik & V. Saudek, J. Magn. Reson.,
; Series A 102, 241 -245 (1993)
;
;$CLASS=HighRes
;$DIM=2D
;$TYPE=
;$SUBTYPE=
;$COMMENT=


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


"p2=p1*2"
"p22=p21*2"
"d12=20u"
"d26=1s/(cnst4*4)"


"d0=3u"

"in0=inf1"
```

"DELTA=d19-p22/2"
"DELTA1=d26-p16-d16-p27*2.385-d19*5+p22/2"
"DELTA2=d26-p16-d16-p27*2.154-p0*0.231-d19*5+p22/2-8u-p21"
"DELTA3=d26-p16-4u"

1 ze
2 d1
3 d12 pl1:f1
50u UNBLKGRAD
(p1 ph1)
4u
p16:gp1
DELTA3
(center (p2 ph1) (p22 ph6):f3)
4u
p16:gp1
DELTA3
(p1 ph2)
3u
p16:gp2
d16
(p21 ph3):f3
d0
(p1 ph5)
4u
p16:gp3
DELTA3
(center (p2 ph1) (p22 ph1):f3)
4u
p16:gp3
DELTA3
(center (p1 ph1) (p21 ph4):f3)
DELTA1
p16:gp4
d16 pl18:f1
p27*0.231 ph7
d19*2
p27*0.692 ph7
d19*2
p27*1.462 ph7
DELTA
(p22 ph1):f3
DELTA
p27*1.462 ph8
d19*2
p27*0.692 ph8
d19*2
p0*0.231 ph8
4u
p16:gp4
d16
4u BLKGRAD
DELTA2
(p21 ph9):f3
go=2 ph31
d1 mc #0 to 2 F1PH(calph(ph3, +90) & calph(ph6, +90), caldel(d0, +in0))
exit

ph1=0
ph2=1

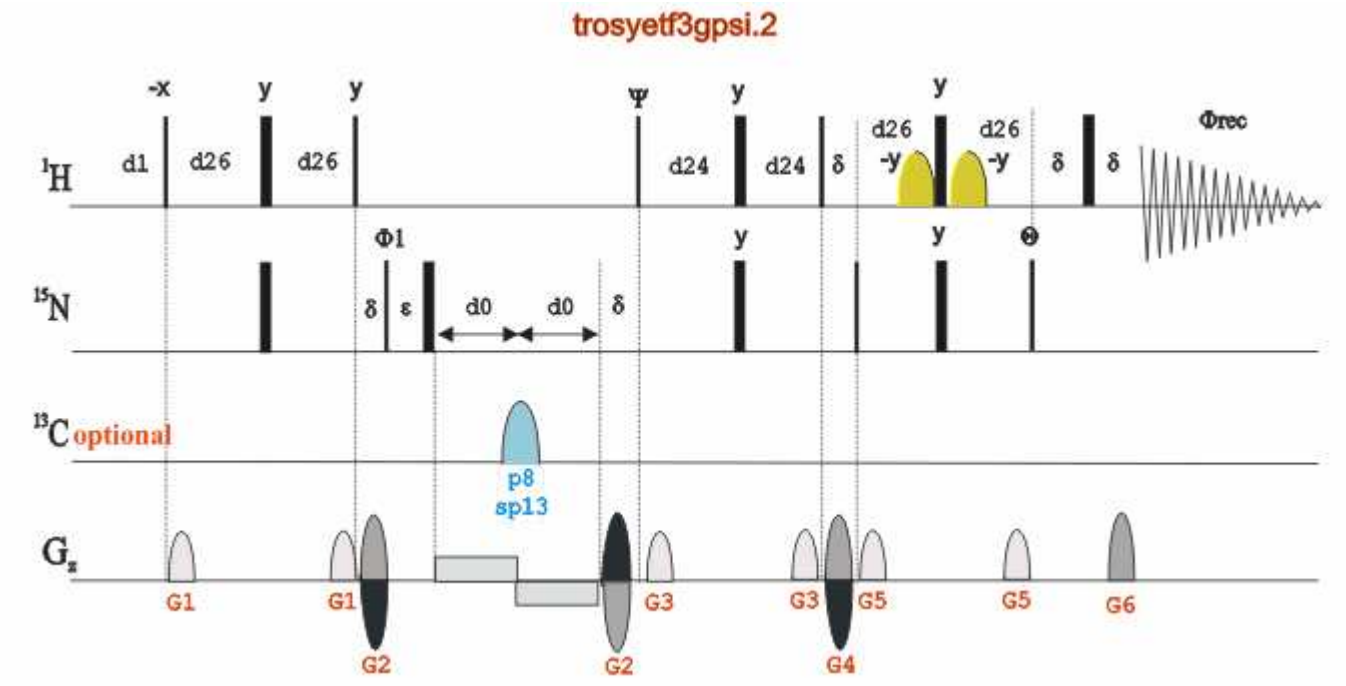
```
ph3=1 3 2 0
ph4=1
ph5=1 1 1 1 3 3 3 3
ph6=0
ph7=0
ph8=2
ph9=0 0 0 0 2 2 2 2
ph31=0 2 3 1 0 2 1 3


;pl1 : f1 channel - power level for pulse (default)
;pl3 : f3 channel - power level for pulse (default)
;pl18: f1 channel - power level for 3-9-19-pulse (watergate)
;p0 : f1 channel - 90 degree pulse at pl18
; use for fine adjustment
;p1 : f1 channel - 90 degree high power pulse
;p2 : f1 channel - 180 degree high power pulse
;p16: homospoil/gradient pulse
;p21: f3 channel - 90 degree high power pulse
;p22: f3 channel - 180 degree high power pulse
;p27: f1 channel - 90 degree pulse at pl18
;d0 : incremented delay (2D) [3 usec]
;d1 : relaxation delay; 1-5 * T1
;d12: delay for power switching [20 usec]
;d16: delay for homospoil/gradient recovery
;d19: delay for binomial water suppression
; d19 = (1/(2*d)), d = distance of next null (in Hz)
;d26 : 1/(4J)YH
;cnst4: = J(YH)
;inf1: 1/SW(X) = 2 * DW(X)
;in0: 1/(1 * SW(X)) = 2 * DW(X)
;nd0: 1
;NS: 8 * n
;DS: 16
;td1: number of experiments
;FnMODE: States-TPPI (or TPPI)


;use gradient ratio: gp 1 : gp 2 : gp 3 : gp 4
; 24 : -60 : 40 : 57.6

;for z-only gradients:
;gpz1: 24%
;gpz2: -60%
;gpz3: 40%
;gpz4: 57.6%

;use gradient files:
;gpnam1: SMSQ10.100
;gpnam2: SMSQ10.100
;gpnam3: SMSQ10.100
;gpnam4: SMSQ10.100



;$Id: trosyf3gpph19,v 1.6 2009/07/02 16:40:47 ber Exp $
```

Previous pp:**trosyetf3gpsi2**
Next pp:**trosyf3gpidphwg**

Pulse Programs

NMRSIM Pulse Diagram
Relevant parameters: **ased**
More info on **trosyetf3gpsi.2**

Pulse Diagram



Pulse Program

```
;trosyetf3gpsi.2
;avance-version (10/02/12)
;2D H-1/X correlation via TROSY
; using sensitivity improvement
;phase sensitive using Echo/Antiecho gradient selection
;using f3 - channel
;(use parameterset )
;
;T. Schulte-Herbrueggen & O.W. Sorensen, J. Magn. Reson. 144,
; 123 - 128 (2000)
;(M. Czisch & R. Boelens, J. Magn. Reson. 134, 158-160 (1998) )
;(K. Pervushin, G. Wider & K. Wuethrich, J. Biomol. NMR 12,
; 345-348 (1998) )
;(A. Meissner, T. Schulte-Herbrueggen, J. Briand & O.W. Sorensen, Mol. Phys. 96,
; 1137-1142 (1998) )
;(J. Weigelt, J. Am. Chem. Soc. 120, 10778-10779 (1998) )
;(M. Rance, J.P. Loria & A.G. Palmer III, J. Magn. Reson. 136, 91-101 (1999) )
;(G. Zhu, X.M. Kong & K.H. Sze, J. Biomol. NMR 13, 77-81 (1999) )
;
;$CLASS=HighRes
;$DIM=2D
;$TYPE=
;$SUBTYPE=
;$COMMENT=


prosol relations=<triple>
```

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


define list<gradient> EA2 = { 0.8750 1.0000}
define list<gradient> EA4 = { 1.0000 0.6667}
define list<gradient> EA6 = { 0.6595 1.0000}


"p2=p1*2"
"p22=p21*2"
"d11=30m"
"d26=1s/(cnst4*4)"


"in0=inf1/2"

"d0=6u"


"DELTA1=d26-p16-d16"
"DELTA2=d25-p16-d16"
"DELTA3=d26-p11-p16-d16-8u"

# ifdef LABEL_CN
"DELTA=d0*2+p8-p21*4/3.1416+8u"
# else
"DELTA=d0*2-p21*4/3.1416+6u"
# endif /*LABEL_CN*/


1 ze
2 d11
3 d1 pl1:f1
50u UNBLKGRAD

(p1 ph3)
p16:gp1
d16
DELTA1
(center (p2 ph2) (p22 ph1):f3 )
DELTA1
p16:gp1
d16
(p1 ph2)

p16:gp2*EA2
d16

(p21 ph5):f3
DELTA
(p22 ph1):f3

# ifdef LABEL_CN
d0 gron0
2u groff
(p8:sp13 ph1):f2
d0 gron0*-1
2u groff
# else
d0 gron0
d0 gron0*-1
```

```
2u groff
# endif /*LABEL_CN*/

4u
p16:gp2*-1*EA2
d16

(p1 ph6)
p16:gp3
d16
DELTA2
(center (p2 ph2) (p22 ph2):f3 )
DELTA2
p16:gp3
d16
(p1 ph1)

p16:gp4*EA4
d16

(p21 ph7):f3
p16:gp5
d16
DELTA3 pl0:f1
(p11:sp1 ph4:r):f1
4u
4u pl1:f1
(center (p2 ph2) (p22 ph8):f3 )
4u pl0:f1
(p11:sp1 ph4:r):f1
4u
DELTA3
p16:gp5
d16 pl1:f1
(p21 ph9:r):f3

p16:gp6*EA6
d16
4u BLKGRAD

go=2 ph31
d11 mc #0 to 2
F1EA(calgrad(EA2) & calgrad(EA4) & calgrad(EA6) & calph(ph6, +180) & calph(ph7, +180),
caldel(d0, +in0) & calph(ph5, +180) & calph(ph31, +180))
exit


ph1=0
ph2=1
ph3=2
ph4=3
ph5=0 2
ph6=3
ph7=0 0 2 2
ph8=1 1 3 3
ph9=3 3 1 1
ph31=0 2 2 0


;pl0 : 0W
;pl1 : f1 channel - power level for pulse (default)
;pl3 : f3 channel - power level for pulse (default)
;sp1: f1 channel - shaped pulse 90 degree (H2O on resonance)
```

;sp13: f2 channel - shaped pulse 180 degree (adiabatic)
;p1 : f1 channel - 90 degree high power pulse
;p2 : f1 channel - 180 degree high power pulse
;p8 : f2 channel - 180 degree shaped pulse for inversion (adiabatic)
;p11: f1 channel - 90 degree shaped pulse [1 msec]
;p16: homospoil/gradient pulse [1 msec]
;p21: f3 channel - 90 degree high power pulse
;p22: f3 channel - 180 degree high power pulse
;d0 : incremented delay (2D) [6 usec]
;d1 : relaxation delay; 1-5 * T1
;d11: delay for disk I/O [30 msec]
;d16: delay for homospoil/gradient recovery
;d25 : 1/(4J')NH,
; compensation delay for suppression of other Trosy peaks
;d26 : 1/(4J)NH
;cnst4: = J(NH)
;inf1: 1/SW(N) = 2 * DW(N)
;in0: 1/(2 * SW(N)) = DW(N)
;nd0: 2
;NS: 4 * n
;DS: 16
;td1: number of experiments
;FnMODE: echo-antiecho

;use gradient ratio: gp 0 : gp 1 : gp 2 : gp 3 : gp 4 : gp 5 : gp 6
; 3 : 3 : 80 : 5 : 30 : 7 :30.13

;for z-only gradients:
;gpz0: 3%
;gpz1: 3%
;gpz2: 80%
;gpz3: 5%
;gpz4: 30%
;gpz5: 7%
