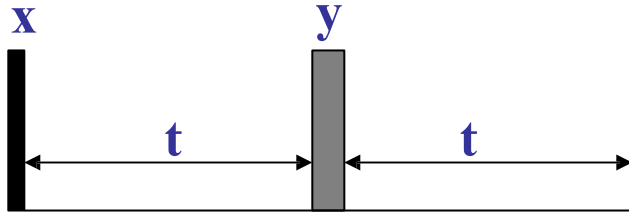# Spin echo


$$\tfrac{1}{2}\pi I_x - t - \pi I_y - t$$

t : as needed, not correlated with 1/J.

Functions: 1. refocusing; 2. decoupling.

- **Chemical shift evolution is refocused by the spin-echo.**
- **Heteronuclear J-couplings evolution are refocused by a spin-echo.** Because only one spin experiences a 180° pulse.
- **Homonuclear couplings evolution are not refocused by a spin-echo.** Because both spins experiences a 180° pulse.

- **It's also used for decoupling H^1-N^{15} by refocusing the H^1 magnetization.**

INEPT – Insensitive Nucleus Enhanced by Polarization Transfer
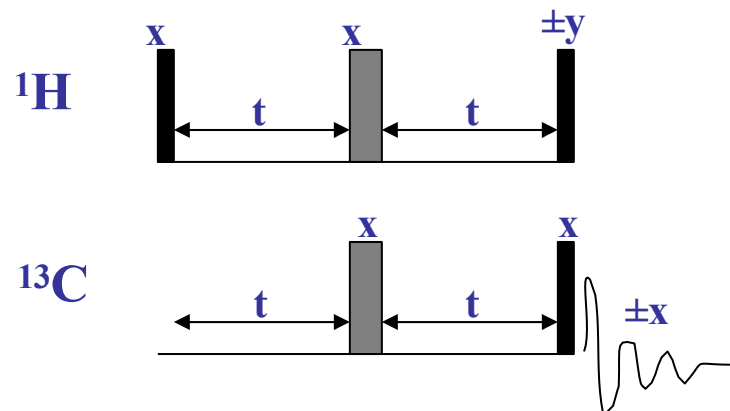
$$t = 1/(4*^1J_{CH}) = 1/(4*212Hz) = 1.18ms$$

INEPT sequence: transfer of population differences from ^1H to X (X: ^{13}C, ^{15}N etc. ^1H and X are J-coupling interaction), (by inversion of populations of proton, $\rightarrow$ changing populations of spin X). It can **enhance signal intensity** of X by γ_H/γ_X (^{13}C, ~4; ^{15}N, ~10), and is widely used in NMR experiments.

Refocused INEPT and Product operator analysis: — right hand rule
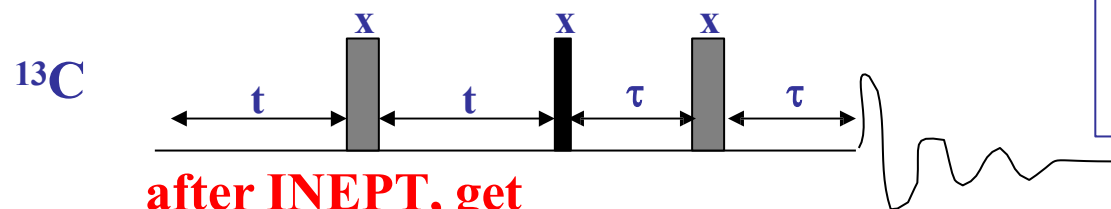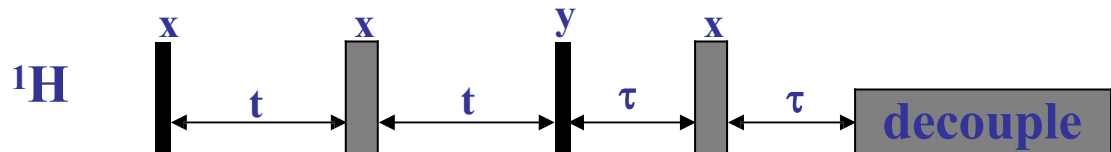
$\tfrac{1}{2}\pi I_x - t - \pi(I_x+S_x) - t - \tfrac{1}{2}\pi(I_y+S_x)$
$- \tau - \pi(I_x+S_x) - \tau$

after INEPT, get anti-phase component

$-2I_zS_y$

$2I_zS_z$

$-2I_zS_y\cos(\pi J\tau)$

$+S_x\sin(\pi J\tau)$

πI_x

$+2I_zS_y\cos(\pi J\tau)$

$+S_x\sin(\pi J\tau)$

πS_x

$-2I_zS_y\cos(\pi J\tau)$

$+S_x\sin(\pi J\tau)$

$-2I_zS_y\cos(\pi J\tau)\bullet\cos(\pi J\tau)$

$+S_x\cos(\pi J\tau)\bullet\sin(\pi J\tau)$

$+S_x\sin(\pi J\tau)\bullet\cos(\pi J\tau)$

$+2I_zS_y\sin(\pi J\tau)\bullet\sin(\pi J\tau)$

If $\tau = 1/(4J)$,

$-2I_zS_y$

$+S_x$

$+2I_zS_y$

In-phase component

Reverse INEPT --- the reverse transfer is achieved.
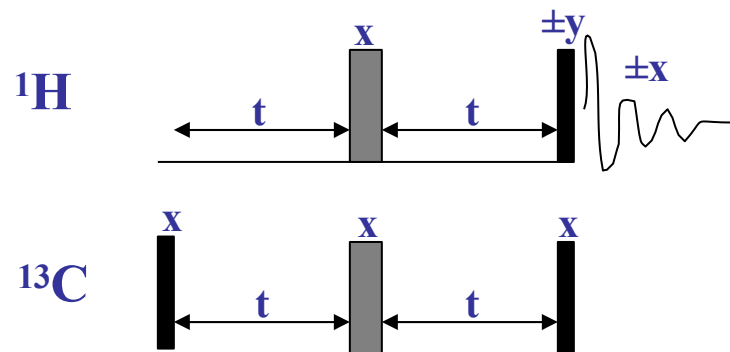
$$t = 1/(4*^1J_{CH}) = 1/(4*212Hz) = 1.18ms$$

In real 2D or 3D expt., the first 90 pulse on the ^{13}C is not needed because the antiphase magnetization is already present after t1 evolution, the reverse INEPT looks like the following in HSQC:
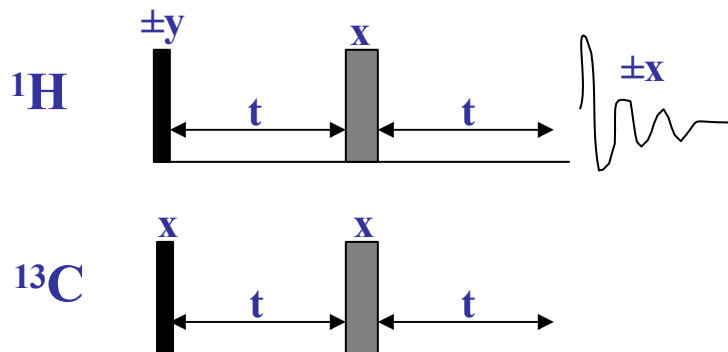
HSQC — Heteronuclear Single-Quantum Coherence

Refocusing of ^1H J & δ

Transfer of ^{15}N magnetization to ^1H

To remove J_{NH} doublet fine structure, and double S/N ratio

decouple

After INEPT, SQ coherence ($-2I_zS_y$) generates

Reverse INEPT reverses the anti-phase ^1H magnetization into in-phase ^1H magnetization

Evolution of ^{15}N chemical shift
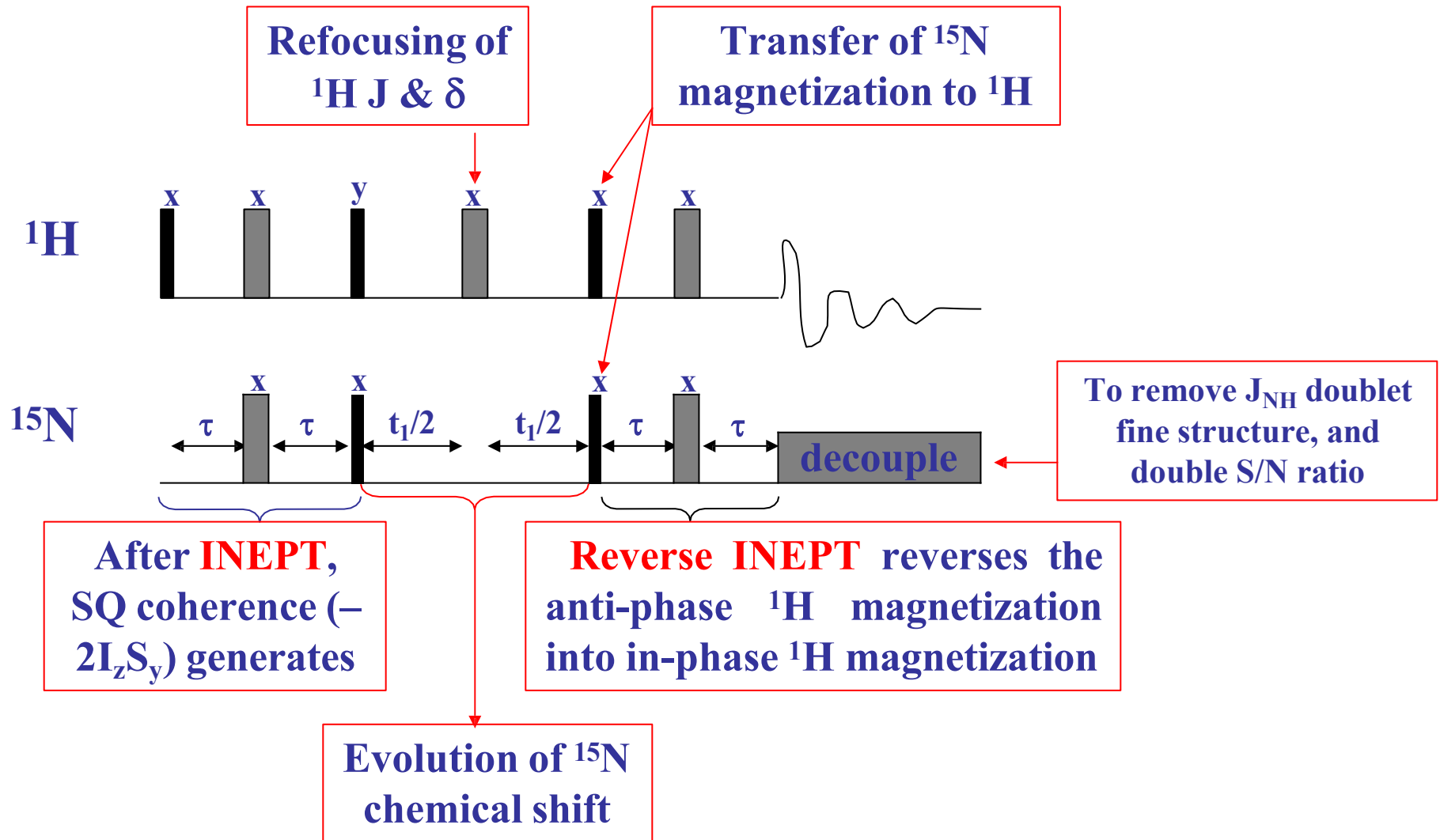
HSQC product operator analysis:

After INEPT sequence, we got:

- ½t₁ - πIₓ - ½t₁ -

$$-2I_zS_y$$

$$\text{- - - - - - - -} \leftarrow \text{Start of } t_1$$

½t₁ (left branch) ½t₁ (right branch)

$$-2I_zS_y\cos(\Omega_s\tfrac{1}{2}t_1) \qquad\qquad +2I_zS_x\sin(\Omega_s\tfrac{1}{2}t_1)$$

πIₓ πIₓ

$$+2I_zS_y\cos(\Omega_s\tfrac{1}{2}t_1) \qquad\qquad -2I_zS_x\sin(\Omega_s\tfrac{1}{2}t_1)$$

½t₁   ½t₁   ½t₁   ½t₁

$$+2I_zS_y\cos(\Omega_s\tfrac{1}{2}t_1)\bullet \qquad -2I_zS_x\cos(\Omega_s\tfrac{1}{2}t_1)\bullet \qquad -2I_zS_x\sin(\Omega_s\tfrac{1}{2}t_1)\bullet \qquad -2I_zS_y\sin(\Omega_s\tfrac{1}{2}t_1)\bullet$$
$$\cos(\Omega_s\tfrac{1}{2}t_1) \qquad\qquad \sin(\Omega_s\tfrac{1}{2}t_1) \qquad\qquad \cos(\Omega_s\tfrac{1}{2}t_1) \qquad\qquad \sin(\Omega_s\tfrac{1}{2}t_1)$$

$$\cos(2\alpha) = \cos^2\alpha - \sin^2\alpha;$$
$$\sin(2\alpha) = 2\sin(\alpha)\cos(\alpha) = \sin(\alpha)\cos(\alpha) + \cos(\alpha)\sin(\alpha)$$

$$+2I_zS_y[\cos^2(\Omega_s\tfrac{1}{2}t_1) - \sin^2(\Omega_s\tfrac{1}{2}t_1)] \qquad\qquad -2I_zS_x2\sin(\Omega_s\tfrac{1}{2}t_1)\cos(\Omega_s\tfrac{1}{2}t_1)$$
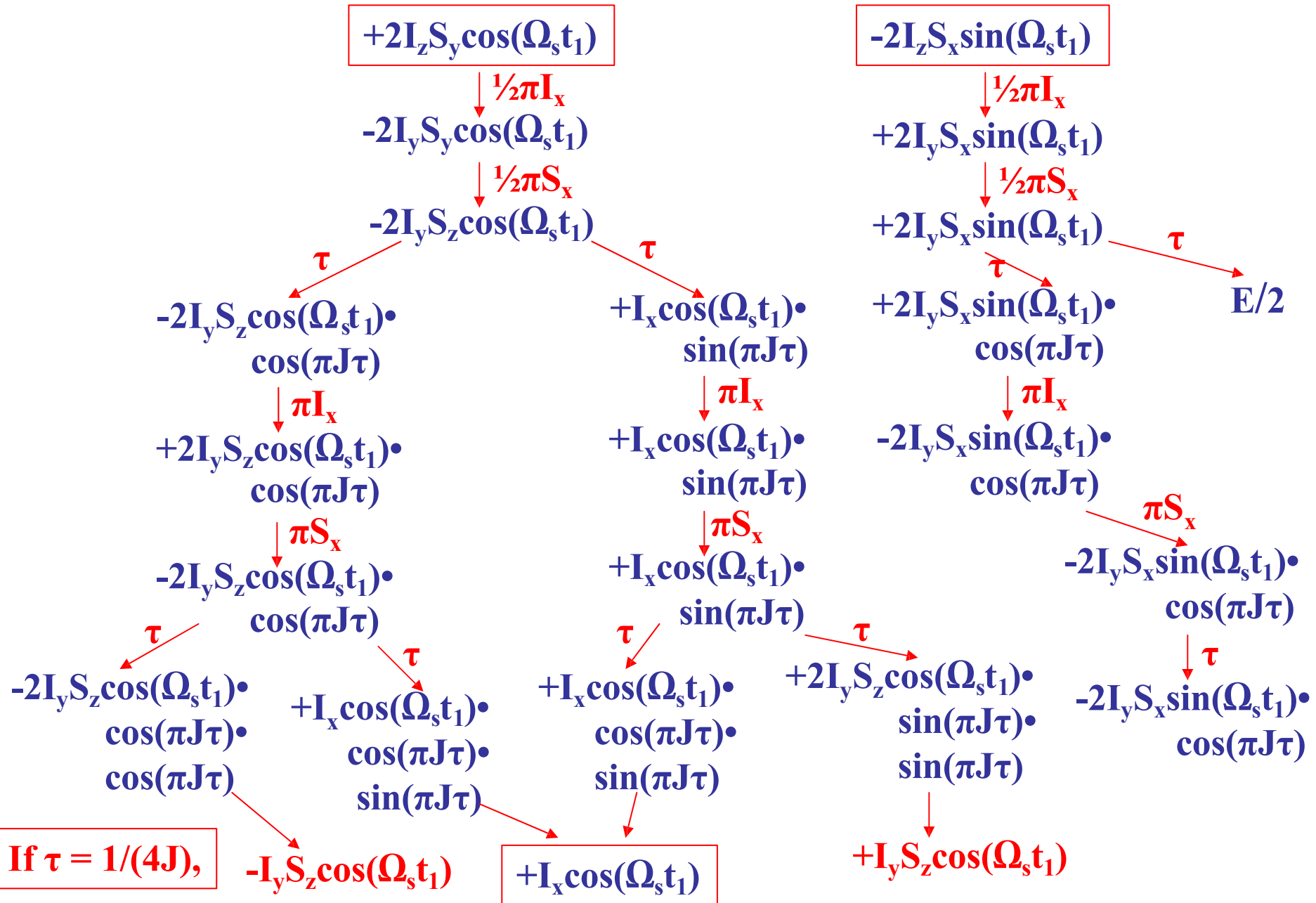
$$+2I_zS_y\cos(\Omega_st_1) \qquad\qquad\qquad -2I_zS_x\sin(\Omega_st_1)$$

Here is the end of t₁.

PEP-HSQC keep this term too,
increase sensitivity by up to √2.

$\frac{1}{2}\pi(I_x + S_x) - \tau - \pi(I_x + S_x) - \tau$

$$\boxed{+2I_zS_y\cos(\Omega_st_1)}$$

$\downarrow \frac{1}{2}\pi I_x$

$-2I_yS_y\cos(\Omega_st_1)$

$\downarrow \frac{1}{2}\pi S_x$

$-2I_yS_z\cos(\Omega_st_1)$

$\swarrow \tau \qquad\qquad \searrow \tau$

$-2I_yS_z\cos(\Omega_st_1)\bullet$
$\cos(\pi J\tau)$

$\downarrow \pi I_x$

$+2I_yS_z\cos(\Omega_st_1)\bullet$
$\cos(\pi J\tau)$

$\downarrow \pi S_x$

$-2I_yS_z\cos(\Omega_st_1)\bullet$
$\cos(\pi J\tau)$

$\swarrow \tau \qquad\qquad \searrow \tau$

$-2I_yS_z\cos(\Omega_st_1)\bullet$
$\cos(\pi J\tau)\bullet$
$\cos(\pi J\tau)$

$+I_x\cos(\Omega_st_1)\bullet$
$\cos(\pi J\tau)\bullet$
$\sin(\pi J\tau)$

$\boxed{\text{If } \tau = 1/(4J),}$

$-I_yS_z\cos(\Omega_st_1)$

$+I_x\cos(\Omega_st_1)\bullet$
$\sin(\pi J\tau)$

$\downarrow \pi I_x$

$+I_x\cos(\Omega_st_1)\bullet$
$\sin(\pi J\tau)$

$\downarrow \pi S_x$

$+I_x\cos(\Omega_st_1)\bullet$
$\sin(\pi J\tau)$

$\swarrow \tau \qquad\qquad \searrow \tau$

$+I_x\cos(\Omega_st_1)\bullet$
$\cos(\pi J\tau)\bullet$
$\sin(\pi J\tau)$

$+2I_yS_z\cos(\Omega_st_1)\bullet$
$\sin(\pi J\tau)\bullet$
$\sin(\pi J\tau)$

$\boxed{+I_x\cos(\Omega_st_1)}$

$+I_yS_z\cos(\Omega_st_1)$

$$\boxed{-2I_zS_x\sin(\Omega_st_1)}$$

$\downarrow \frac{1}{2}\pi I_x$

$+2I_yS_x\sin(\Omega_st_1)$

$\downarrow \frac{1}{2}\pi S_x$

$+2I_yS_x\sin(\Omega_st_1)$

$\qquad \searrow \tau$

$\downarrow \tau$

$+2I_yS_x\sin(\Omega_st_1)\bullet$
$\cos(\pi J\tau)$

$E/2$

$\downarrow \pi I_x$

$-2I_yS_x\sin(\Omega_st_1)\bullet$
$\cos(\pi J\tau)$

$\searrow \pi S_x$

$-2I_yS_x\sin(\Omega_st_1)\bullet$
$\cos(\pi J\tau)$

$\downarrow \tau$

$-2I_yS_x\sin(\Omega_st_1)\bullet$
$\cos(\pi J\tau)$

$+I_x\cos(\Omega_s t_1)$

**The chemical shift of spin S cosine modulates the amplitude of peak I.**

1. HSQC and HMQC provide **single-bond heteronuclear shift correlations**, the correlation data are equivalent for both.
2. Historically, HSQC is favored by biological community, and presents ^{1}H-^{15}N correlations in protein molecules; HMQC is favored by chemical community, and presents ^{1}H-^{13}C correlations in small organic molecules.
3. Both HSQC and HMQC have the following 3 feathers:
   - **From known proton assignments, get to know the correlated heteronucleus assignments.**
   - **Proton peaks disperse according to the heteronucleus shift.**
   - **Can identify diastereotopic geminal pairs.**
4. Only difference between HSQC and HMQC is during t_1 period:
   - HSQC, only heteronulear transverse **SQ magnetization ($-2I_zS_y$) evolves,**
   - HMQC, ^{1}H-^{13}C **MQ coherence ($-2I_xS_y$) evolves.**
5. In HSQC, homonuclear ^{1}H-^{1}H couplings do not influence heteronuclear X (S_y) magnetization evolution $\rightarrow$ signals do not contain homonuclear ^{1}H-^{1}H couplings along f_1 $\rightarrow$ **improve resolution in f_1** $\rightarrow$ this is the principle advantage of HSQC over HMQC for small organic molecules.
   But HSQC use more pulses, especially 180° pulses on heteronuclears $\rightarrow$ promoting **intensity losses** from pulse miscalibration, rf inhomogeneity…

HSQC
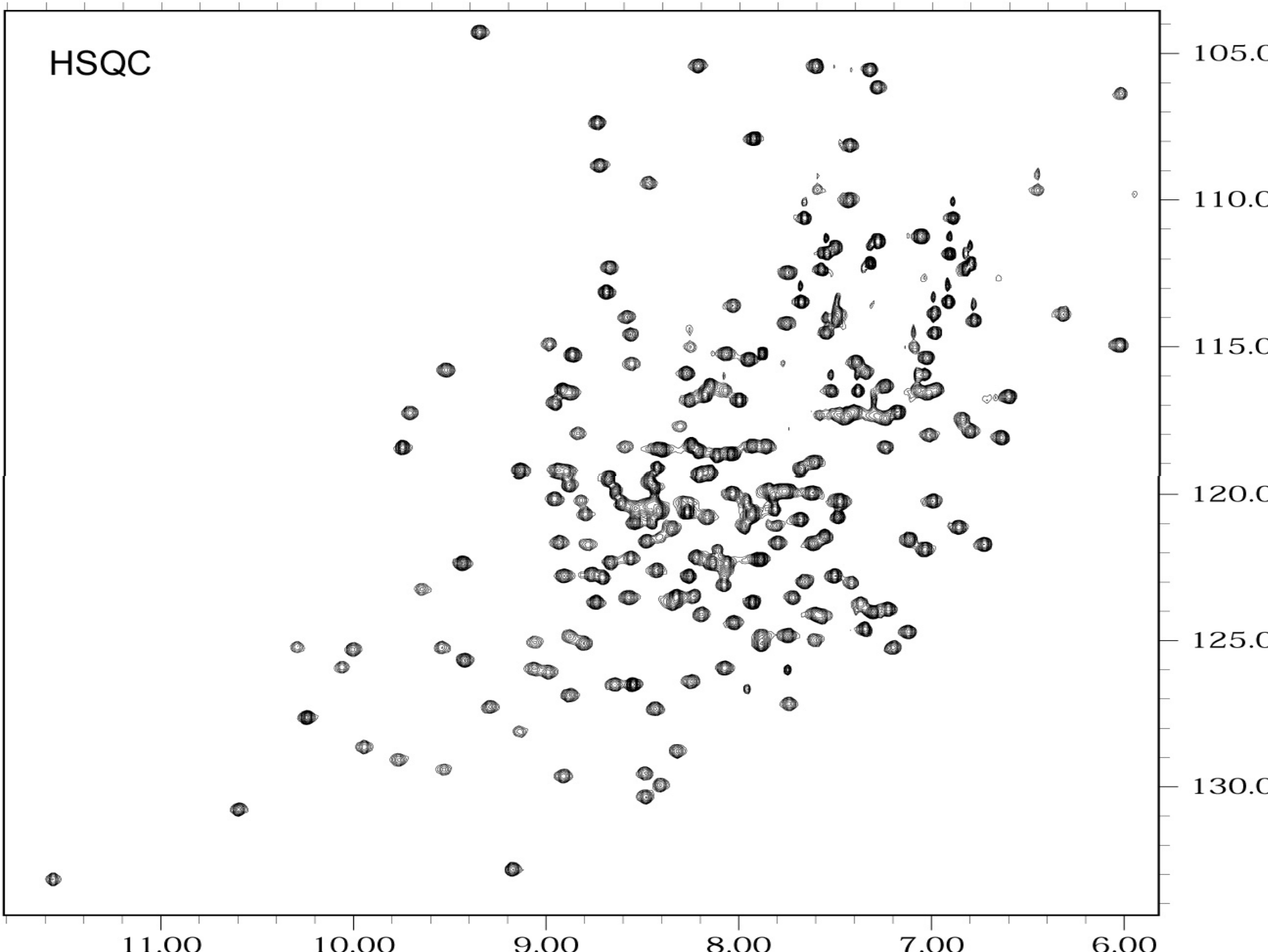
# PEP-HSQC --- "Preservation of Equivalent Pathways" developed by Rance and coworkers
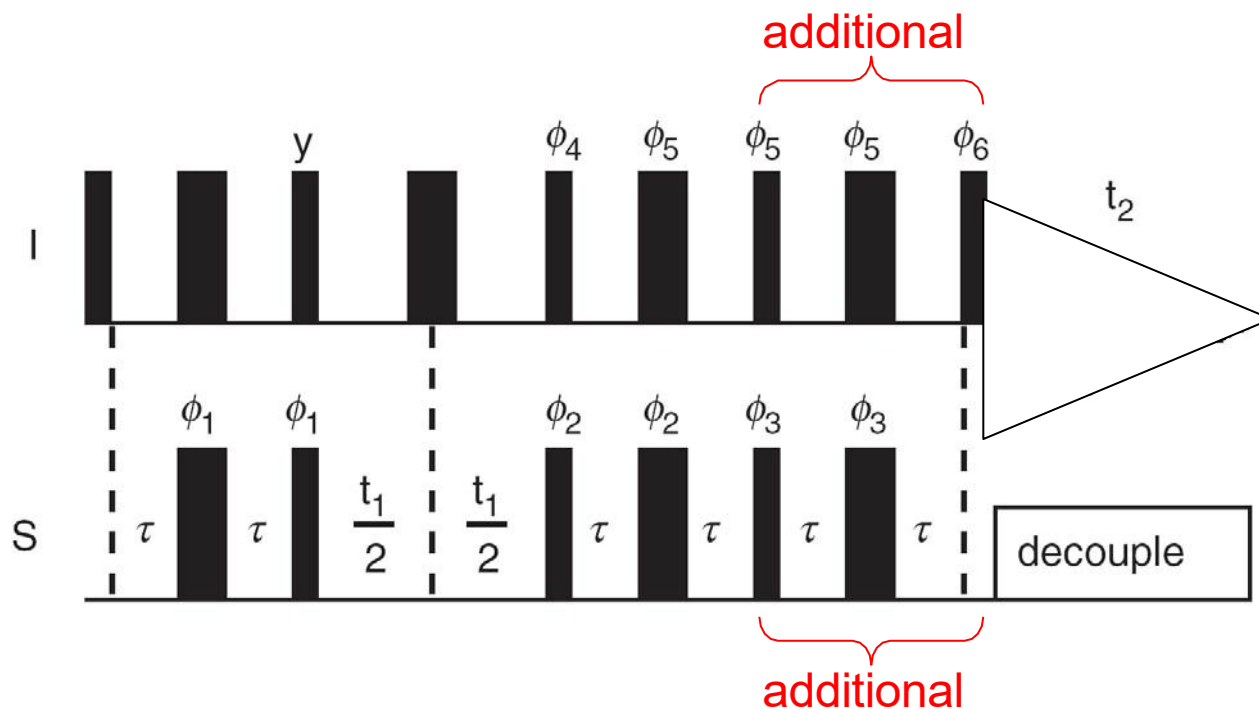
After the first INEPT, $I_z \rightarrow -2I_zS_y$, after t_1 evolution ($t_1/2 - \pi(I_x+K_x) - t_1/2$), $-2I_zS_y \rightarrow 2I_zS_y\cos(\Omega_s t_1) - 2I_zS_x\sin(\Omega_s t_1)$; these two orthogonal terms can be preserved, and sensitivity can be improved by a factor up to $\sqrt{2}$.

When processing these kinds of 2D or 3D spectra, one should choose "Rance-Kay" as yMODE or zMODE in nmrPipe process macro. (Not for Bruker data.)

S^{3}CT : spin-state-selective coherence transfer

I spin:			180°$_x$		180°$_x$
S spin:	90°$_x$ – t – 180°$_x$ –t – 90°$_y$				

I spin:

                x   x
                                t

S spin:

        x   x   y
              t   t
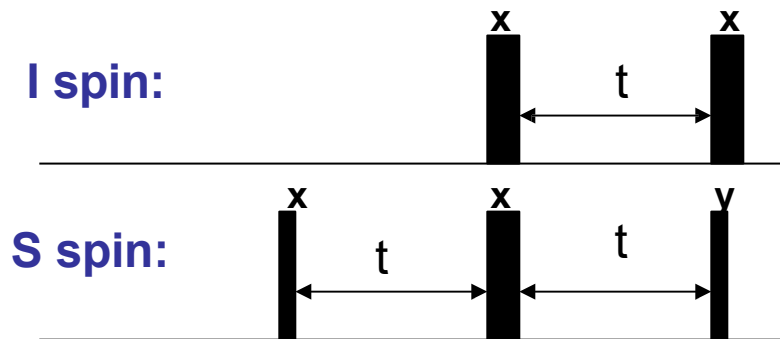
$t = 1/(4J_{IS})$

The S^{3}CT element can convert ZQ and DQ coherences to SQ coherence.