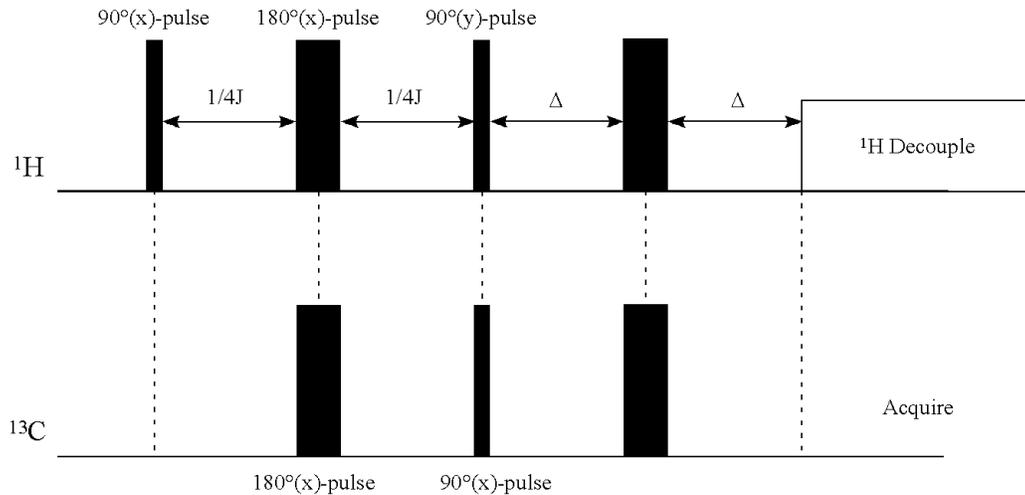


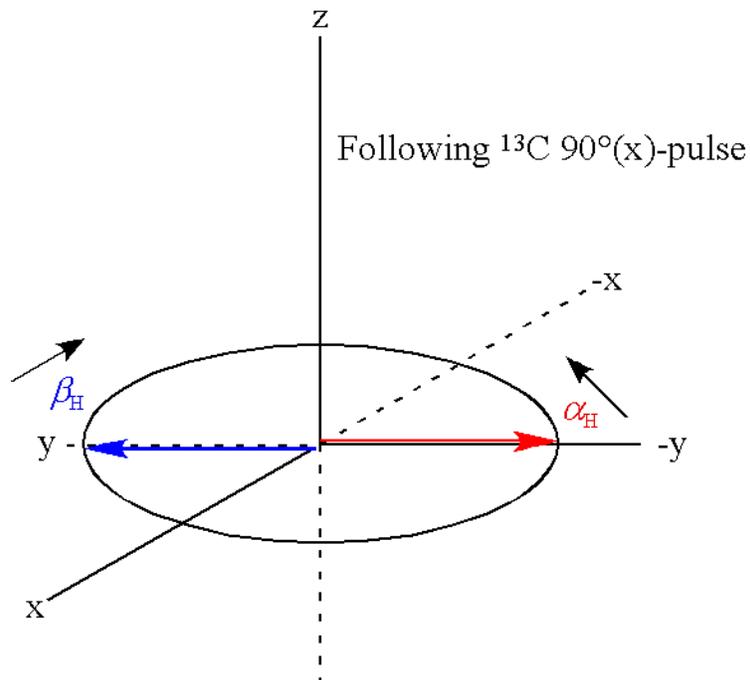
Spectral Editing

We begin with a review of the refocused INEPT experiment:

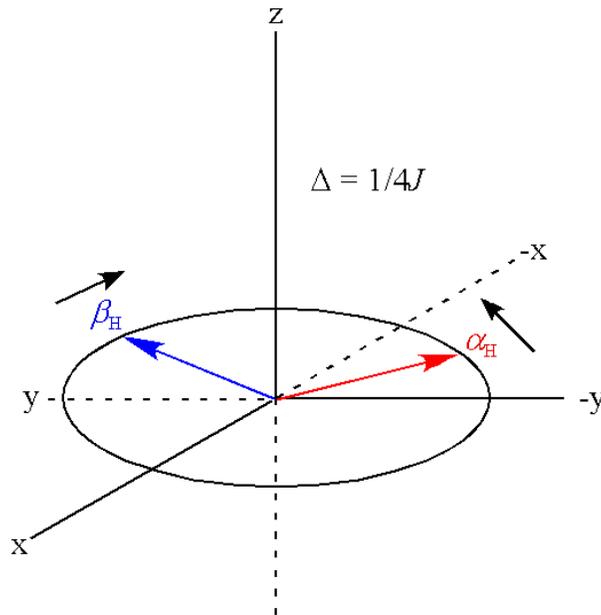


The claim was made in the previous lecture that for a ^1H - ^{13}C pair that the optimal value for Δ was $1/4J$ - let's see how this works.

We commence the analysis right after the ^{13}C $90^\circ(x)$ -pulse:

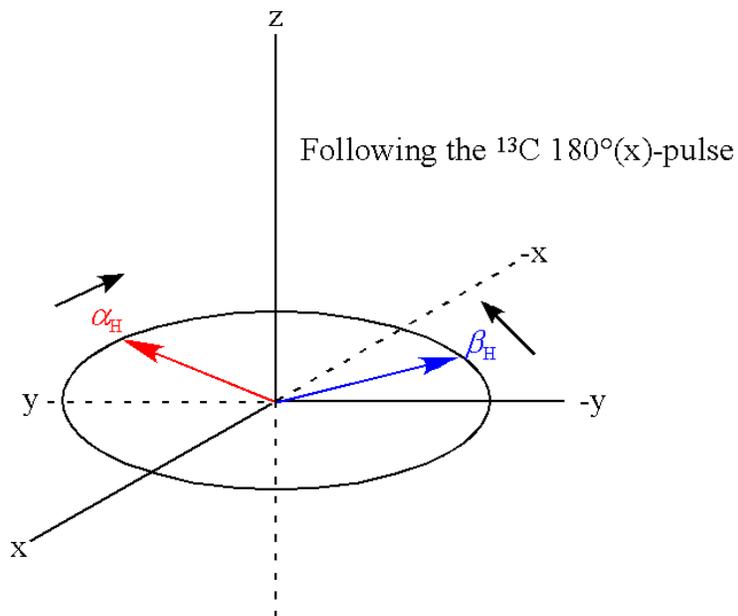


After an interval equal to $1/4J$ we would observe the following:

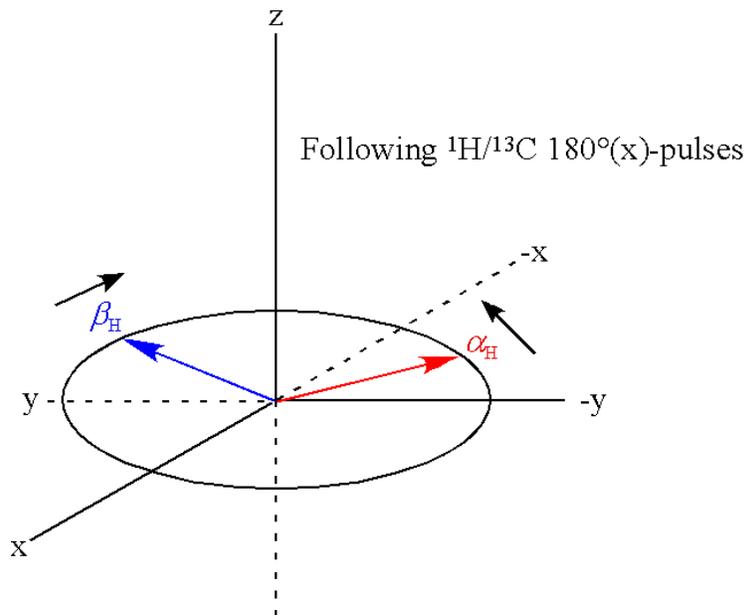


Let's look carefully at the $^1\text{H}/^{13}\text{C}$ $180^\circ(x)$ -pulse pair. We can decompose the pulse into two effects.

First the ^{13}C $180^\circ(x)$ -pulse pair inverts the vectors:

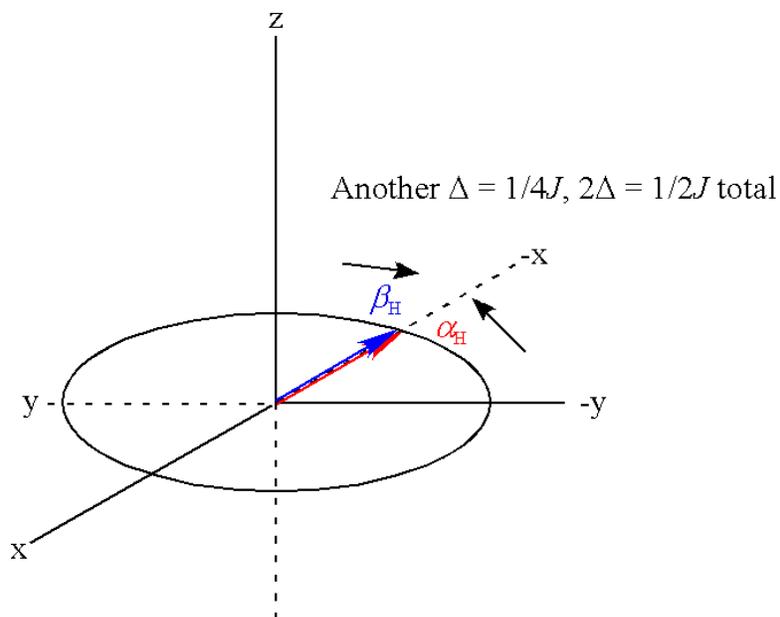


The ^1H $180^\circ(x)$ -pulse inverts the ^1H spin states (α_{H} and β_{H}):



This appears to produce a situation that is identical to the state of the system before the pulse-pair. If there had been a resonance offset, the pulse pair would prepare the system for chemical-shift refocusing as well.

During the final interval the ^{13}C vectors refocus:



Note that for the ^1H - ^{13}C pair, the angle, θ , between the angle between the α_{H} and β_{H} vectors is given by the following expression:

$$\theta = 2\pi\Delta J \text{ radians}$$

Thus at $\Delta = 1/4J$:

$$\theta = 2\pi\left(\frac{1}{4J}\right)J = \frac{\pi}{2} \text{ or } \theta = 360\left(\frac{1}{4J}\right)J = 90^\circ$$

which agrees with our previous vector analysis.

For CH_2 and CH_3 groups the dependence is different, and the intensity of the manifolds evolves according to the following expressions:

$$I_x^{\text{CH}}(\Delta) \propto \sin \theta = \frac{N\gamma_{\text{H}}\gamma_{\text{C}}B_0}{4k_bT} \sin \theta$$

$$I_x^{\text{CH}_2}(\Delta) \propto \sin 2\theta = \frac{N\gamma_{\text{H}}\gamma_{\text{C}}B_0}{4k_bT} \sin 2\theta$$

$$I_x^{\text{CH}_3}(\Delta) \propto 3\cos^2 \theta \sin \theta = \frac{N\gamma_{\text{H}}\gamma_{\text{C}}B_0}{4k_bT} 3\cos^2 \theta \sin \theta = \frac{N\gamma_{\text{H}}\gamma_{\text{C}}B_0}{4k_bT} \frac{3}{4}(\sin \theta + \sin 3\theta)$$

This dependence is shown graphically in the following figure (Note that $\theta = 360\Delta J_{\text{HC}}$):

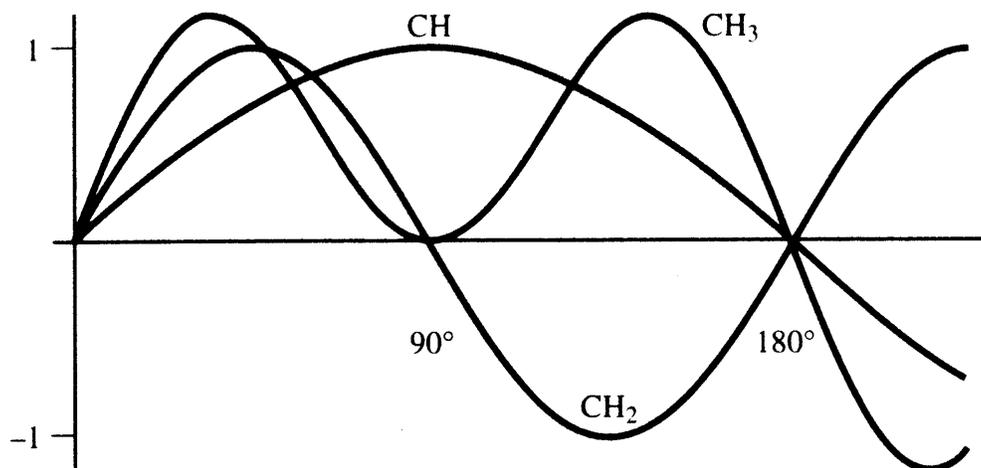


Figure 3 from Morris, G.A. (1995) INEPT. In *Encyclopedia of NMR Spectroscopy*. Academic Press, New York.

The following series of spectra illustrate results of the more common experimental variants of the INEPT experiment:

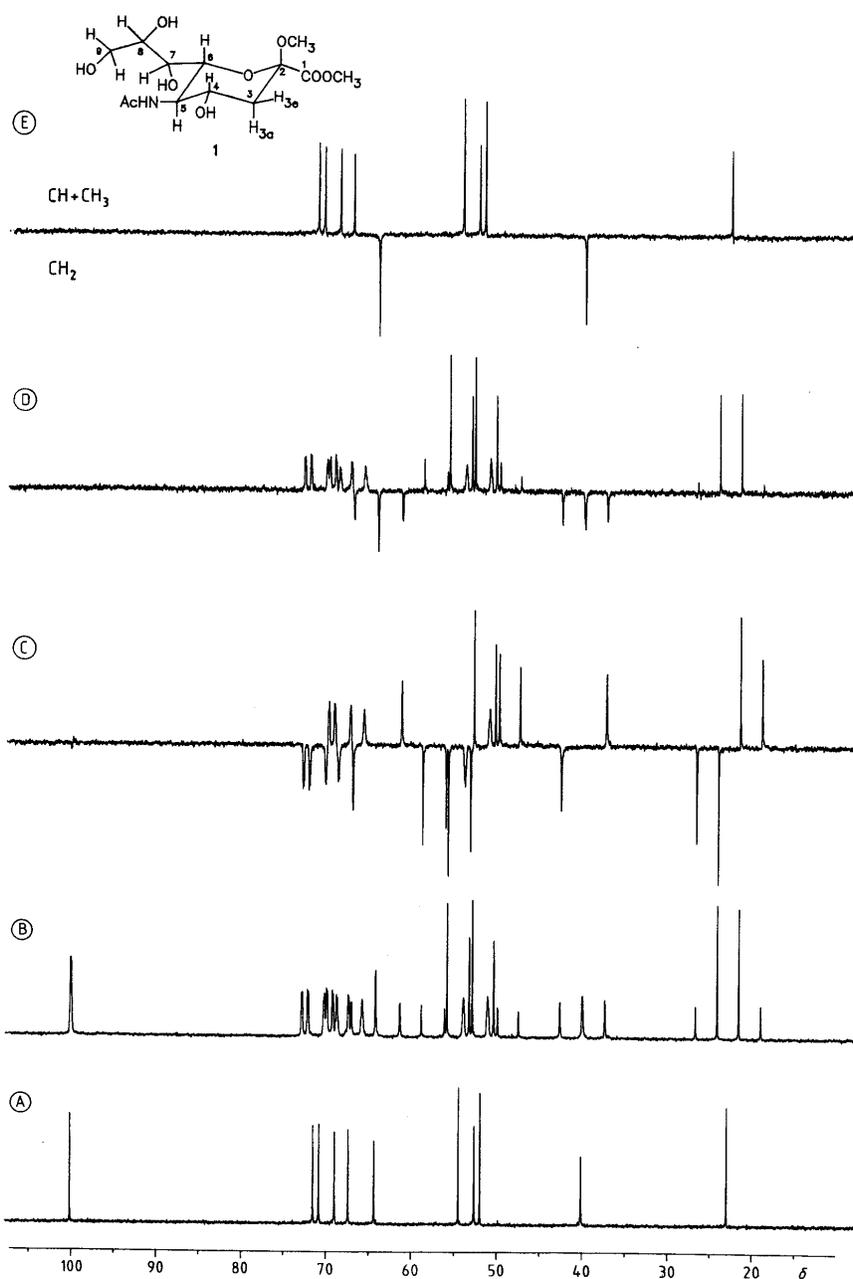
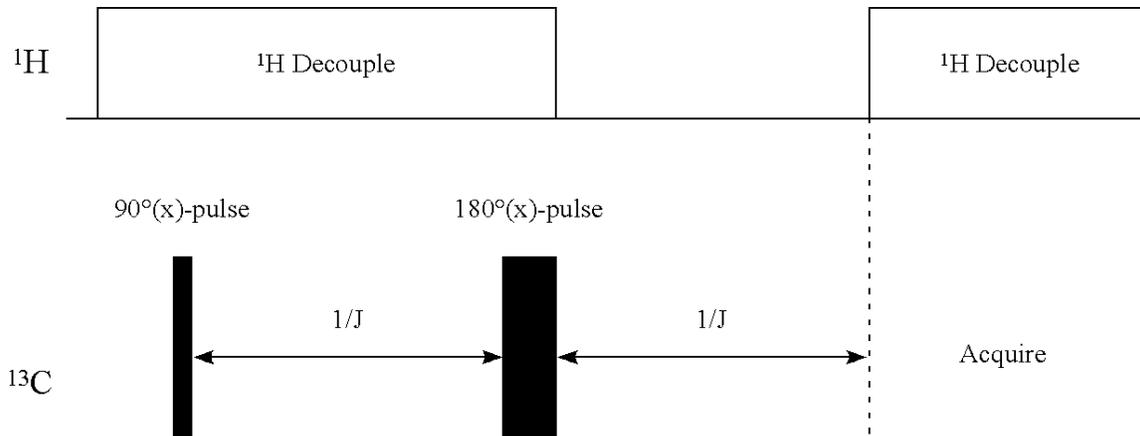


Figure 8-18 from the Friebolin text. A) 50.3 MHz ^{13}C NMR spectrum with ^1H decoupling. B) Coupled ^{13}C spectrum. C) INEPT spectrum. D) Refocused INEPT – no ^1H decoupling. E) Refocused INEPT with ^1H decoupling during acquisition. The τ delay is chose to be $1/4J$ and the Δ delay is set at $3/8J$

Attached Proton Test (APT)

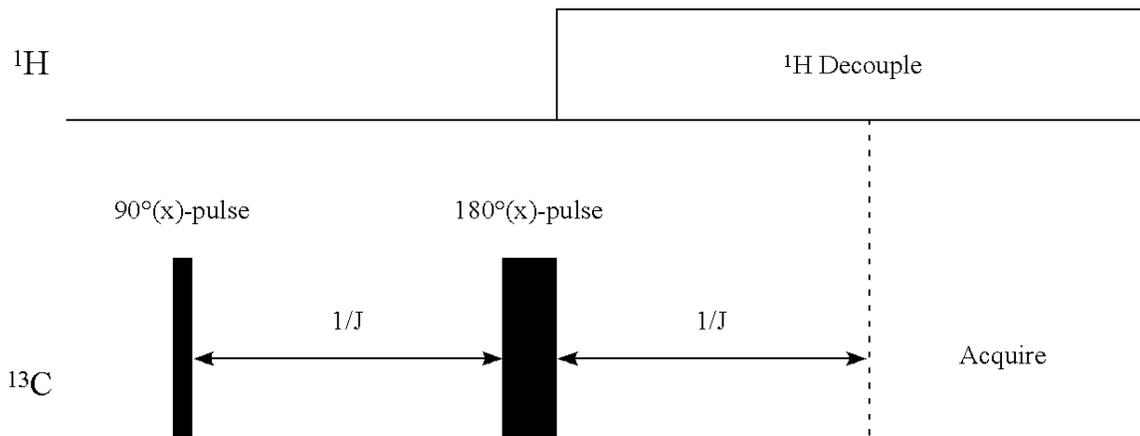
The precise experimental implementation of the APT experiment varies, but the overall goal is to distinguish from one another ^{13}C nuclei that are bonded to 0, 1, 2 or 3 ^1H nuclei.

One APT pulse sequence is shown below:



The ^{13}C $90^\circ(x)$ -pulse generates transverse magnetization that evolves during an interval equal to $1/J$. ^1H decoupling during this interval eliminates coupling and generates an NOE.

The version depicted in the Friebolin text (Figure 8-9) generates the same effect, but provides lower NOE enhancement:



The ^{13}C $180^\circ(x)$ -pulse inverts the ^{13}C spin states. During the subsequent $1/J$ interval, ^1H - ^{13}C J coupling develops – the components of the ^{13}C magnetization coupled to the α and β components of the ^1H spins will be coincident (inverted relative to ^{13}C and $^{13}\text{CH}_2$) for ^{13}CH and $^{13}\text{CH}_3$ groups and coincident (inverted relative to ^{13}CH and $^{13}\text{CH}_3$ groups) for ^{13}C and $^{13}\text{CH}_2$ moieties.

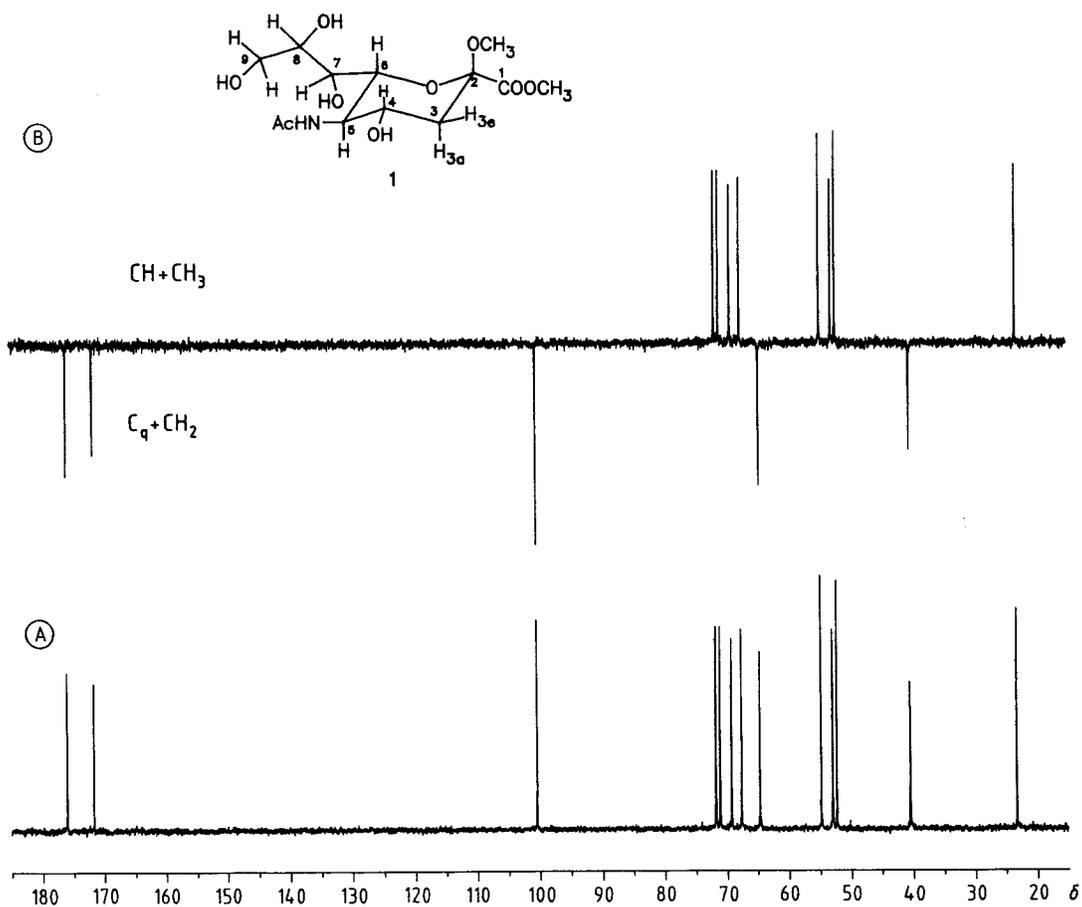


Figure 8-12.

Example of a J -modulated spin-echo experiment.

A: 50.3 MHz ^{13}C NMR spectrum of the neuraminic acid derivative **1** with ^1H BB decoupling.

B: 50.3 MHz ^{13}C NMR spectrum of the same sample, recorded using the J -modulated spin-echo pulse sequence (Fig. 8-9 A). Signals with positive amplitudes are assigned to CH or CH_3 groups, those with negative amplitudes to quaternary carbons or CH_2 groups.

(*Experimental conditions for B:*

20 mg of the compound in 0.5 ml D_2O ; 5 mm sample tube; 224 echoes recorded; 32 K data points; $\tau = 7.14$ ms; duration of experiment: approx. 5 min.)

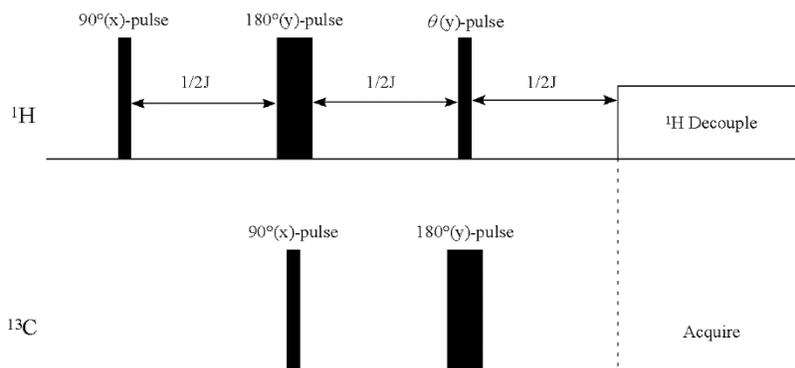
Figure 8-12 from Friebolin text.

Distortionless Enhancement by Polarization Transfer (DEPT)

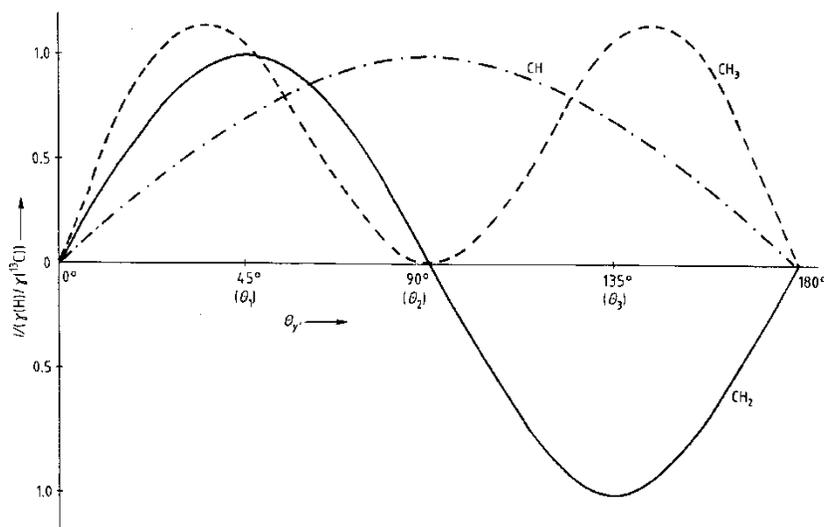
Pegg, D.T., Doddrell, D.M., Brooks, W.M. and Bendall, M.R. (1981) Proton Polarization Transfer Experiment for a Nucleus with Arbitrary Spin Quantum Number from N Scalar Coupled Protons for Arbitrary Preparation Times. *J. Magn. Reson.* **44**, 32-40.

The INEPT and APT experiments both provide reasonably effective spectral editing; however both suffer from the shortcoming that effective results are obtained only when the value of J is similar for all functional groups.

This liability was address in the DEPT sequence, which substitutes a pulse with a variable tip angle, θ_y , for the variable delay:



The selectivity of the DEPT experiment depends upon the nutation angle produced by the θ_y -pulse:



From Friebolin text, Figure 8-23

A remarkable feature of the DEPT experiment is the ability to produce so subspectra that have been edited to contain specific subsets of the full spectrum:

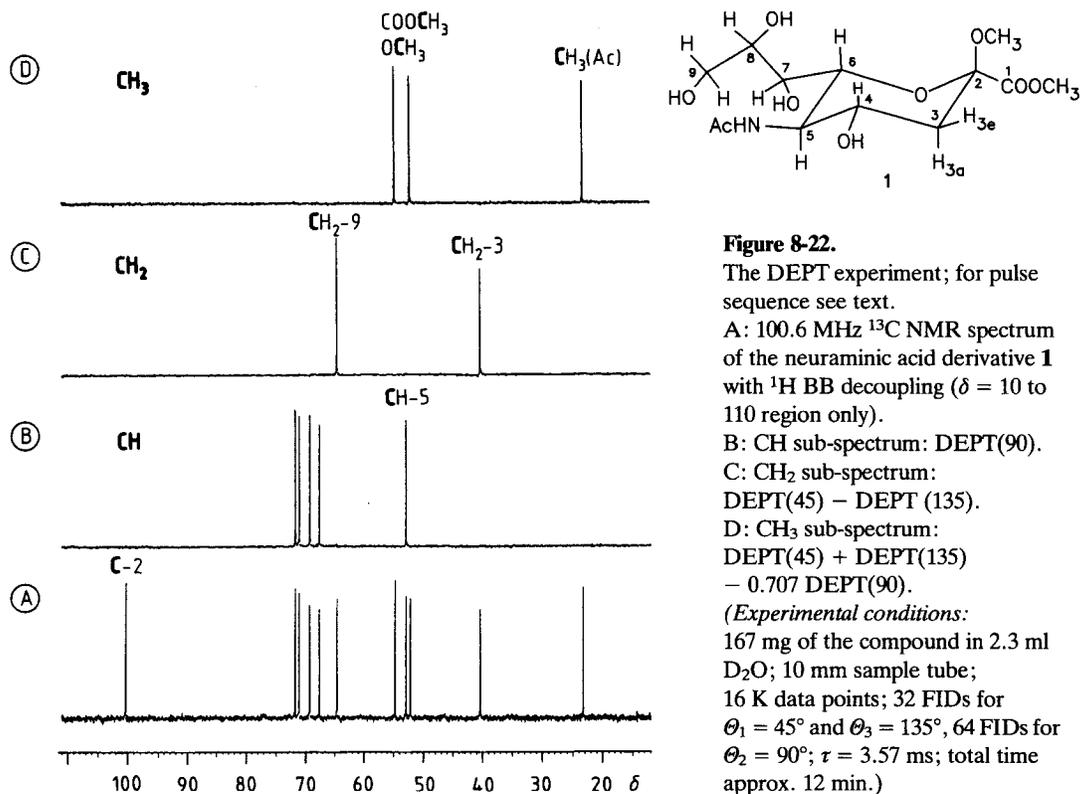


Figure 8-22 from the Friebolin text.