

Laboration 5

INEPT

KR

Theory

The **INEPT (Insensitive Nuclei Enhanced by Polarization Transfer) experiment** was designed to improve the sensitivity of NMR experiments on low-abundant and low magnetogyric ratio nuclei. The net effect is the non-selective polarization transfer from protons to X nuclei with the appropriate $^1\text{H-X}$ coupling. X is in this case ^{13}C .

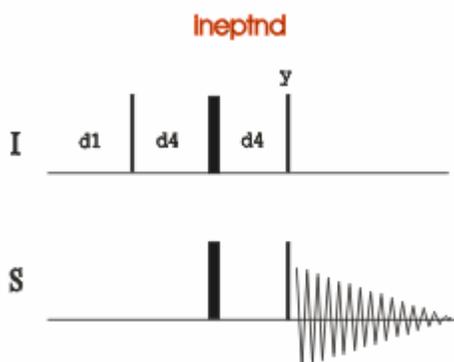


Fig. 1. The INEPT pulse sequence.

The classical INEPT experiment affords a ^1H -coupled X spectrum in which X resonances appear as antiphase multiplets with respect $^1\text{J}(\text{CH})$. The intensities of the multiplet lines depend of the nuclei being observed. Thus, for instance, a doublet with relative intensity 5:-3 is obtained for a $^1\text{H-}^{13}\text{C}$ pair compared to the 1:1 doublet obtained in a classical ^1H -coupled ^{13}C spectrum. Usually, the phase of the second 90° ^1H pulse in the INEPT sequence is inverted (+y and -y) on alternate scans and a relative 4:-4 (or 1:-1) ratio results for a doublet system. Triplets and quartets due to CH_2 and CH_3 systems are similarly modified according to 1:0:-1 and 1:1:-1:-1 ratios, respectively.

This is what we will do in this lab.

Refocused INEPT experiment: Addition of a refocusing period (delay- $180^\circ(^1\text{H},\text{X})$ -delay) in order to obtain in-phase X magnetization. Now, it is possible to apply broadband proton decoupling during acquisition in order to obtain a ^1H -decoupled X spectrum.

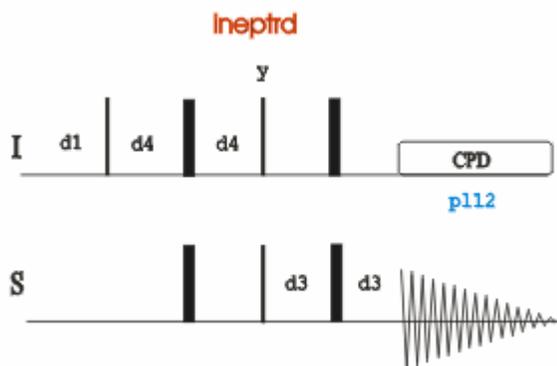


Fig. 2. The Refocused INEPT pulse sequence.

In the refocused version, in-phase X multiplets are obtained. This experiment is usually recorded with broadband proton decoupling, and therefore a ^1H -decoupled X spectrum with improved sensitivity is recorded.

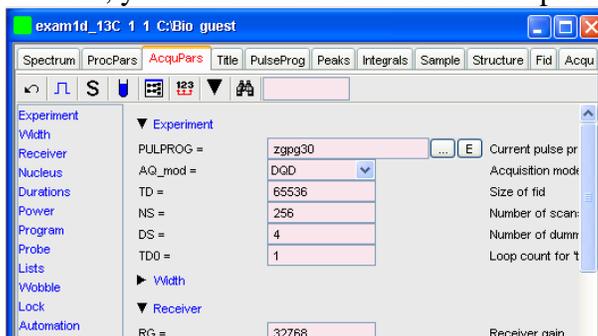
Refocused INEPT experiments were used to determine multiplicity of X nuclei, similarly as described for DEPT experiments, by optimizing the refocusing delay.

Practical

1. Run a *PROTON* according to "Bruker run manual for 500 MHz NMR".
2. Check the ^1H 90°-pulse (lab 3).

Experiment setup

1. **edc** or **new** and read the parameter set *A_13C*
2. *Tune and match* (new nucleus involved)
3. If required, any acquisition parameter can be modified manually or in the *AcquPars* section., you can see what is valid for the parameters in *PulseProg*.



- a. From the standard ^{13}C spectrum file, change the pulse program to **pulprog = ineptnd** in the *AcquPars*, write it in the menu or go in to rpar .
 - b. If required optimize the values of **o1p** and **sw** (the protonated carbons) from the conventional ^{13}C spectrum.
 - c. **o2p** must be set to the center of the proton spectrum.
 - d. Set an appropriate number of scans **ns**. Number of scans must be increased 3-5 times owing to a considerable deterioration in S/N ratio according to a standard ^{13}C spectrum, because of the splitting of signals.
 - e. Set **d1** to an appropriate value, you find the parameter under the  icon
2. **getprosol** (get the preset probe and solvent specific parameters from the prosol list) or if the ^1H 90°-pulse value need to be changed

Set the measured **p1** (^1H 90°-pulse)

getprosol 1H 'p1-value' 'p11-value' (get probe and solvent specific parameters and use your adjusted p1 value to calculate related pulses)

4. The expected experimental time is displayed with the **expt** command or 
5. **rga** find the receiver gain
6. **zg** start the acquisition

Processing recorded data

7. **ef** add a window function and Fourier transform the data.
8. **apk**, perform an automatic phase
9. **abs n** perform a baseline correction