

Nuclear Overhauser Effect (NOE)

The interaction involved is the direct magnetic coupling (the dipolar coupling) between nuclei, which does not usually have any observable effect on spectra recorded in solution.

NOE \Rightarrow Dipolar coupling \Rightarrow Intermolecular distances and molecular motion

NOE — a change in intensity of one resonance when the transitions of another are perturbed.

A physical process behind the NOE — nuclear relaxation

How?

Via saturation of some of the signals (elimination of the population difference across some transitions by irradiating them with a weak rf field) while observing signals from others. NOE is then a manifestation of the attempt of the system to stay at equilibrium.

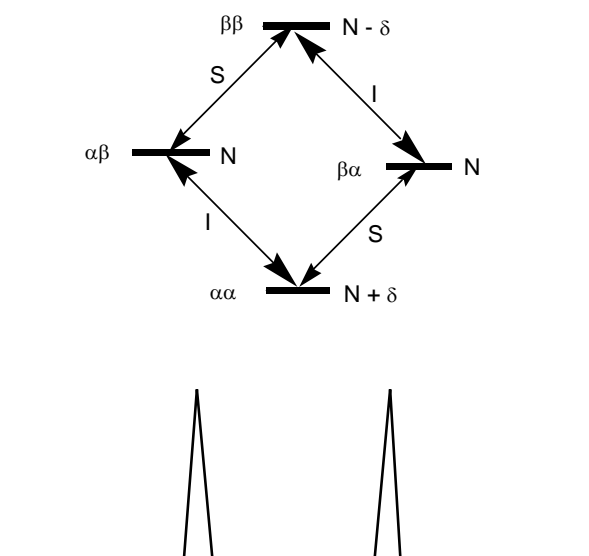
Quantitative definition:

$$\eta_i(s) = (I - I_0) / I_0$$

where I_0 is the normal intensity of a resonance and I is the intensity observed while saturating some other related resonance. $\eta_i(s)$ indicates that this is the NOE at nucleus i when nucleus s is saturated.

Two-spin homonuclear system

Two spin-1/2 nuclei i and s , in the same molecule, $J_{is} = 0$

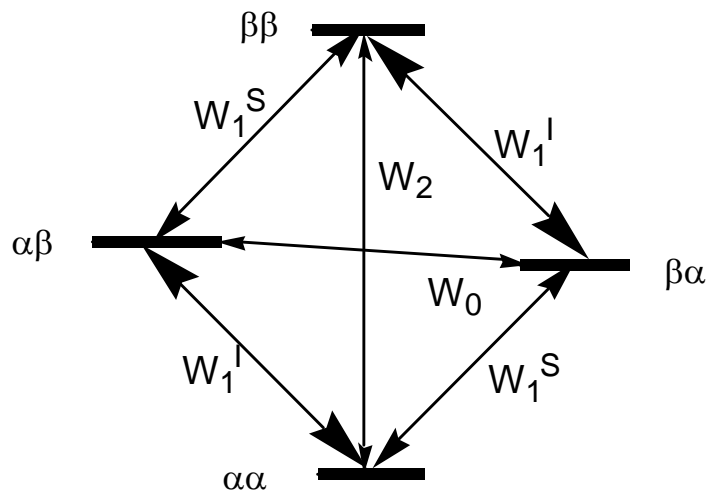


Population differences between sites

i transitions: $\alpha\alpha-\alpha\beta$ and $\beta\alpha-\beta\beta \Rightarrow$	δ
s transitions: $\alpha\alpha-\beta\alpha$ and $\alpha\beta-\beta\beta \Rightarrow$	δ
$\Delta M = 0$ transition: $\beta\alpha-\alpha\beta \Rightarrow$	0
$\Delta M = 2$ transition: $\alpha\alpha-\beta\beta \Rightarrow$	2δ

Transitions corresponding to $\Delta M = 0$ and $\Delta M = 2$ are not observed in NMR spectra under normal conditions, but they may be involved in the relaxation: if these population differences are disturbed the system will attempt to restore them.

Possible relaxation pathways:



W - the rate constants for various processes

Assumption made:

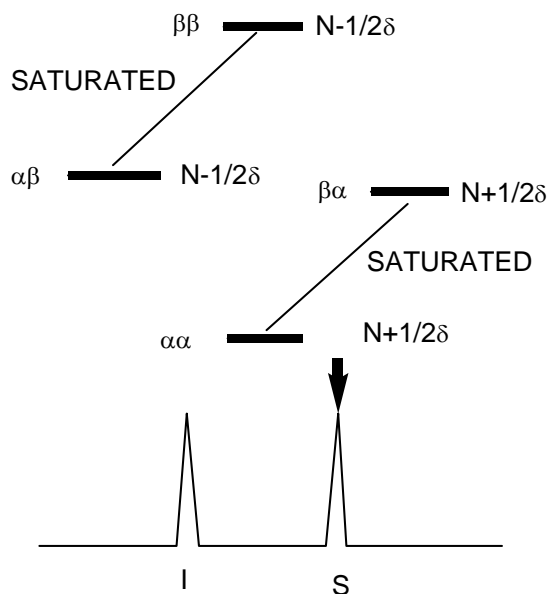
the relaxation is a 1st order process, i.e., it is proportional to the extent to which the population difference across the transition differs from its equilibrium value.

If the $\Delta M = 0$ and $\Delta M = 2$ transitions did not occur, then

$$T_1^i = 1 / (2W_1^i)$$

Otherwise the total relaxation rate for nucleus i must involve the population differences of nucleus s .

The NOE experiment



	<u>The new population differences</u>	<u>At equilibrium</u>
<i>i</i> transitions: $\alpha\alpha-\alpha\beta$ and $\beta\alpha-\beta\beta \Rightarrow$	δ	δ
<i>s</i> transitions: $\alpha\alpha-\beta\alpha$ and $\alpha\beta-\beta\beta \Rightarrow$	0	δ
$\Delta M = 0$ transition: $\beta\alpha-\alpha\beta \Rightarrow$	δ	0
$\Delta M = 2$ transition: $\alpha\alpha-\beta\beta \Rightarrow$	δ	2δ

If only single quantum transitions are active as relaxation pathways, saturating *s* does not affect the intensity of *i*. If W_0 is the dominant relaxation pathway, saturating *s* decreases the intensity of signals due to *i* (negative NOE).

If W_2 is the dominant relaxation pathway, saturating *s* increases the intensity of signals due to *i* (positive NOE).

$$\eta_i(s) = \frac{W_2 - W_0}{W_0 + 2W_I^i + W_2}$$

$\eta_i(s) > 0$ for small molecules, $\eta_i(s) < 0$ for macromolecules

Heteronuclear case:

$$\eta_i(s) = \frac{W_2 - W_0}{W_0 + 2W_I^i + W_2} \frac{\gamma_s}{\gamma_i}$$

The total relaxation rate $\rho_i = W_0 + 2W_I^i + W_2$

The cross relaxation rate $\sigma = W_2 - W_0$

$$\eta_i(s) = \frac{\sigma}{\rho_i} \frac{\gamma_s}{\gamma_i}$$

Dipolar relaxation

The strength of the dipolar interaction depends on the internuclear distance:

$$D_{is} \sim \frac{\gamma_i \gamma_s \hbar}{4\pi^2 r_{is}^3}$$

In solution rapid molecular motion averages the dipolar interaction over all possible orientations of the molecule. The rapid reorientation of the dipolar interaction can be considered as a source of fluctuating fields. Of the common mechanisms only the dipolar relaxation has cross-relaxation terms (W_0 and W_2 processes) and the observation of NOE is diagnostic of this type of relaxation.

Random motions can be characterized by a single parameter - the correlation time τ_c . If the molecule moves from one orientation to another and each move occurs instantaneously, then there will be a random distribution of "waiting times" at each orientation: τ_c is chosen such that waiting times shorter than τ_c seldom occur.

High molecular weight macromolecules — longer τ_c

The relaxation rates via dipolar coupling in a system of two spins:

$$W_{I^i} \sim \frac{3\tau_c}{r^6(1+\omega_i^2\tau_c^2)}$$

$$W_0 \sim \frac{2\tau_c}{r^6(1+(\omega_i-\omega_s)^2\tau_c^2)}$$

$$W_2 \sim \frac{12\tau_c}{r^6(1+(\omega_i+\omega_s)^2\tau_c^2)}$$

where ω_i and ω_s are the Larmor frequencies of the two nuclei.

Slowly tumbling molecules have little or no motion at 2ω ($\approx \omega_i + \omega_s$) and W_0 exceeds W_2 . This is the limit in which $\omega^2\tau_c^2 \gg 1$, called the spin diffusion limit (NOE is negative).

In the extreme narrowing limit ($\omega^2\tau_c^2 \ll 1$):

$$W_{I^i} \sim \frac{3\tau_c}{r^6} \quad W_0 \sim \frac{2\tau_c}{r^6} \quad W_2 \sim \frac{12\tau_c}{r^6}$$

Homonuclear case:
$$\eta_i(s) = \frac{W_2 - W_0}{W_0 + 2W_{I^i} + W_2} = \frac{(12-2)\tau_c/r^6}{(6+12+2)\tau_c/r^6} = \frac{1}{2}$$

Heteronuclear case:
$$\eta_i(s) = \frac{1}{2} \frac{\gamma_s}{\gamma_i}$$

The *maximum* positive homonuclear NOE we can ever obtain is 50%. For the $^{13}\text{C}\{^1\text{H}\}$ case, the maximum enhancement is about 200%. Due to this great increase in sensitivity, routine ^{13}C spectra are usually acquired using broadband proton irradiation. The γ dependence leads to another consideration for those nuclei with negative γ values (e.g., ^{15}N and ^{29}Si) as these will show *negative* enhancements. For example, $\frac{\gamma_{\text{H}}}{\gamma_{\text{Si}}} \approx -5$, the maximum NOE = -250%, i.e., if the full NOE is not realised enhancements close to -100% may lead to the loss of the signal.

The NOE enhancement is inversely proportional to the total relaxation rate of i spin (ρ_i). If the i spin is rapidly relaxing due to other relaxation mechanisms, then the total relaxation rate becomes large and the signal enhancement due to NOE is reduced. For example, the i spin can effectively interact with unpaired electrons. The magnetic moment of the electron is about 2000 times greater than that of the proton. In the presence of paramagnetic materials (e.g., paramagnetic oxygen molecules in solutions) intermolecular interactions are strong and lead to very short relaxation times. For nuclei with spin greater than 1/2 the quadrupolar relaxation mechanism becomes dominant and the NOE is not significant.

Transient NOE (TOE)

The NOE itself depends on the competing balance between various relaxation pathways, but the initial rate at which it grows depends only on the rate of cross-relaxation (σ): for constant τ_c this will simply depend on the internuclear distance. Since saturation cannot be brought about instantaneously, a selective inversion of the s spin is used:

$$(180^\circ)_{\text{SEL}} - \tau_m - 90^\circ - \text{Acquisition}$$

The build-up of the i spin's NOE as a function of time is followed. Since $\sigma \sim r^{-6}$ this can be used to estimate internuclear distances. Usually a known fixed distance within the same molecule is used to calibrate the build-up curve (TOE vs. τ_m).

Transient NOE in the rotating frame (ROE)

$$(180^\circ)_{\text{SEL}} - 90^\circ - \tau_m (\text{spin-locking}) - \text{Acquisition}$$

Two important consequences:

- (i) the ROE is always positive (38.5% for $\omega\tau_c \ll 1$ and 67.5% for $\omega\tau_c \gg 1$) and there is no "difficult region" of $\omega\tau_c$. For molecules of intermediate size (with $M_w \sim 1000$ Da) $\omega\tau_c$ is close to 1;
- (ii) multispin effects will be minor.