Relaxation and the Nuclear Overhauser Effect

- Relaxation rates depend on matching transition frequencies to time-varying magnetic fields.
- A proton affects surrounding protons via dipole-dipole interactions. The dipole field can be visualized as a small bar magnet placed at the proton nucleus.

\[
\Delta E_{dd} \propto \gamma_i \gamma_j^2 \left\langle \sum_j \frac{\tau_c}{r_{ij}^6} \right\rangle_t
\]

- Note the distance and correlation time dependencies.
Molecular Motions and Relaxation

- Relaxation rates depend on matching transition frequencies to time-varying magnetic fields: $T_1$ relaxation is most efficient when protons experience molecular motions at the Larmor frequency.

Figure 8.6
The Nuclear Overhauser Effect (2-spin system)

In NMR: excess population $\propto 1 - \exp(-\Delta E/RT) \sim \Delta E$

(see Claridge or Sanders&Hunter)
The Nuclear Overhauser Effect

- **Through-space dipole-dipole interactions!**

[spins can be J-coupled, but nOe does not arise from J-coupling!]

\[ \begin{align*}
\beta\beta \\
\alpha\beta \\
\alpha\alpha \\
\beta\alpha \\
\end{align*} \]
The Nuclear Overhauser Effect

- Start with selective inversion:
NOEs in the Rotating Frame: NOEs

- Selective inversion can be pictured in a simplified manner as:

\[ \text{sel-NOESY} \]
Relaxation rates depend on matching transition frequencies to time-varying magnetic fields: $T_1$ relaxation is most efficient when protons experience molecular motions at the Larmor frequency.
**Negative NOEs: Large MW**

- Slow motions are effective for small frequencies/energy differences:
  \[ W_0 (ZQ) \Rightarrow \text{negative NOE dominates for large MW} \]
Molecular Motions and Relaxation

- Relaxation rates depend on matching transition frequencies to time-varying magnetic fields: $T_1$ relaxation is most efficient when protons experience molecular motions at the Larmor frequency.
Positive NOEs: Small MW

- Fast motions are effective for large frequencies/energy differences:

$$W_2 \ (DQ) \Rightarrow \text{positive NOE dominates for small MW}$$

\[\eta_H\{S\} = 1/2\]
Zero NOEs: The Crossover Region

- Thus, NOEs go through a *crossover region* at intermediate MW, in the range 1000-5000 Da.
The ROESY Spin-Lock: Reducing the Effective Field

Problems with the crossover region can be avoided by *spin-locking* the magnetization.

During a spin-lock, the *effective* magnetic field (on-resonance) is $B_1$.

\[ B \approx 2-6 \text{ kHz} \]

\[ B_0 \approx 500 \text{ MHz} \]

“*All*” molecules in solution tumble fast compared to 6 kHz!

ROEs are always positive.

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**Claridge Figure 8.24**

A schematic illustration of events during spin-lock mixing. All chemical shift differences between spins are eliminated yet all spin–spin couplings between them remain. This forces the *strong-coupling* condition on all spins (see text).
Review - ROESY

- Theoretical steady-state ROESY enhancements are shown below.
  - In practice, the spin-lock causes many problems, the worst being the possibility of TOCSY (J-coupling) transfers in the spectrum.
  - Avoid having coupled multiplets centered in the spectrum.
  - A number of variations of ROESY exist, with differing attributes.
  - Attempt NOESY first, and use ROESY only if required.
Heteronuclear NOEs: Positive $\gamma$ Nuclei

$\eta$

$M(H)_{eq}$

$M(C)_{eq}$

with NOE

with NOE, larger $r$

w/o NOE
Heteronuclear NOEs: Positive $\gamma$ Nuclei

\[ \text{\eta} \]

\[ M(H)_{eq} \]

\[ M(C)_{eq} \]

-CH-

>C<
Heteronuclear NOEs: Negative $\gamma$ Nuclei

$^{29}\text{Si}, ^{15}\text{N}, ^{119}\text{Sn}$
Homonuclear NOEs: Transient Experiments

Transient NOE experiments (e.g., NOESY1D, NOESY2D) will impose a limitation on the mix time of approx $T_1$. At mix $\sim T_1$ and longer, loss of magnetization via spin-lattice relaxation will dominate over the NOE.

Larger %NOE is an indication of smaller $r$, but definitely not always: spin diffusion causes problems for large MW, and certain geometries are problematic for all MW.
Homonuclear NOEs: Transient Experiments

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Larger %NOE is an indication of smaller $r$, but definitely not always: spin diffusion causes problems for large MW, and certain geometries are problematic for all MW. And $T_1$!
NOE - Qualitative and Quant Cautions

- Geometry can be very important to NOE interpretations.
  - E.g., three protons distributed in a near-equilateral triangle can produce zero NOE, independent of $r_{IS}$.
  - It is important to be aware that not observing an NOE is weak evidence. *Measure NOEs in all directions.*
Distance Information via NOE Measurements

- The enhancement maximizes at $1 + \gamma_H/2\gamma_X$ (fast limit) or 0 (slow limit).

Note lack of $r$ dependence!!
Distance Information via NOE Measurements

- The enhancement maximizes at $1 + \frac{\gamma_H}{2\gamma_X}$ (fast limit) or 0 (slow limit).

Note lack of r dependence!!

Claridge
Figs 8.14+8.15
• The build-up rate with mix time is $r$ dependent:

$$ R_v = K' \gamma_i^2 \gamma_S^2 \tau_c r_{IS}^{-6} $$

Plot NOE versus mix, and compare to known pair:

Claridge Fig. 8.21
Distance Information via NOE Measurements

- The build-up rate with mix time is $r$ dependent:

$$ R_v = K' \gamma_1^2 \gamma_S^2 \tau_c r_{IS}^{-6} $$

Plot NOE versus mix, and compare to known pair:

use mix times in the build-up regime

Claridge Fig. 8.21
Summary: NOE

- NOE’s occur via population transfers, and are slow to occur (taking times approaching \( \sim T_1 \)).
  - DQ relaxation leads to positive NOEs, but requires high-frequency modulations (low MW).
  - ZQ relaxation leads to negative NOEs, occurs for all molecules (0 frequency): dominates at high MW.

- NOESY-1D and NOESY-2D are *transient* experiments. These experiments have utility for \( \text{mix} \leq T_1 \).

- The spin-lock of a ROESY experiment reduces the effective magnetic field to 2-6 kHz. Thus, ROEs are *always* positive (i.e., motions in liquids are *always* fast compared to these kHz frequencies).
Spin-locking the magnetization scales the chemical shift to near-zero (in Hz), producing strong coupling.
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Strongly coupled protons exchange magnetization once every $\sim 1/2J$.

**Figure 8.24** TOCSY

**Figure 5.62** A schematic illustration of events during spin-lock mixing. All chemical shift differences between spins are eliminated yet all spin–spin couplings between them remain. This forces the strong-coupling condition on all spins (see text).