Spin Relaxation

BCMB/CHEM 8190
Feb 2, 2005
T₁, T₂, NOE (reminder)

T₁ is the time constant for longitudinal relaxation - the process of re-establishing the Boltzmann distribution of the energy level populations of the system following perturbation.

T₂ is the time constant for transverse relaxation - loss of phase coherences of the nuclear dipoles in the transverse plane.

The Nuclear Overhauser Effect is the change in intensity for a signal (resonance) when the equilibrium spin populations of a different spin are perturbed.
What are the origins of $T_1$ and $T_2$ relaxation and the Nuclear Overhauser Effect (NOE)?

Key: A fluctuating interaction is capable of causing a transition just like an rf pulse.

$$H(t) = -B_1(t) \gamma Ix$$

But, $B_1(t)$ is natural in origin (tumbling of molecules)

Some sources of interaction:

- chemical shift anisotropy
- dipole-dipole (nucleus-nucleus or nucleus-electron)
- nuclear quadrupole - electric field gradient
- others….
Chemical Shift Anisotropy (CSA)

Chemical shifts arise from electronic shielding of the nucleus

- shielding depends on orientation of the molecule with respect to \( B_0 \)
- the orientation dependent chemical shift differences or range is called the CSA
- in solution, rapid reorientation results in averaging of the chemical shift

Rapid molecular reorientation results in local, fluctuating magnetic fields (magnitude and direction)

- these local fluctuating fields lead to energy level transitions, just like applied fields
An Example for CSA Relaxation

The nuclear shielding can be described by a tensor, \( \sigma \), relating the induced field to the applied field. The average (isotropic) shielding is defined as \( \sigma_{\text{iso}} = (\sigma_{11} + \sigma_{22} + \sigma_{33})/3 \)

\[
\begin{bmatrix}
\sigma_{11} & \sigma_{12} & \sigma_{13} \\
\sigma_{21} & \sigma_{22} & \sigma_{23} \\
\sigma_{31} & \sigma_{32} & \sigma_{33}
\end{bmatrix}
\]

Orientation determines effective field: for instance, if \( \sigma_{33} \) is aligned with \( B_0 \), then

\[
B = (1-\sigma_{33}) B_0
\]

As a molecule rapidly reorients in solution, the effective field at a given nucleus fluctuates rapidly with time.

CSA can cause zero (\( W_0 \)) and one (\( W_1 \)) quantum transitions.
The Dipole-Dipole Interaction

The dipolar interaction depends on distance \((1/r^3)\) and orientation \((\theta)\)

A local fluctuating magnetic field is experienced at nucleus A as molecule tumbles and \(\theta\) changes

The fluctuating fields can cause zero \((W_0)\), single \((W_1)\), and two \((W_2)\) quantum transitions

The magnitude of \(\mu_B\) is important - an unpaired electron is about \((2000)^2\) more efficient than a proton at the same distance
The Nuclear Overhauser Effect (NOE)

-depends on competition between $W_0$ and $W_2$ processes:
Correlation Functions

The fluctuating local magnetic fields are time dependent and average to zero for long times.

Correlation / Autocorrelation Function, $G(\tau)$: defines the rate at which these fields fluctuate.

$$G(\tau) = \langle f(t + \tau) f(t) \rangle$$

$$G(0) = f^2(t)$$
Correlation Functions

Correlation function averages two points at increasing separation, $\tau$.

- for small $\tau$, $t$ and $t+\tau$ tend to be similar (and same sign), so for the ensemble, the average of $f(t)$ and $f(t+\tau)$ is high
- for large $\tau$, $t$ and $t+\tau$ are unrelated, and the ensemble average tends toward zero
Correlation Functions

Random processes give rise to exponential correlation functions:

\[ G(\tau) = G(0) \exp(-|\tau| / \tau_c), \]

where \( \tau_c \) is a “correlation time”, the time constant for decay of \( G(\tau) \)

\( \tau_c \) is a measure of the rotational correlation time of molecules in solution

Stoke’s Law relates \( \tau_c \) to molecular size, solvent viscosity and temp.:

\[ \tau_c = \frac{4\pi \eta a^3}{(3 \ k_b \ T)}: \text{ small molecule, high T, low } \eta \text{ means small } \tau_c \]
Correlation Functions

- slow fluctuations
- large molecules
- long $\tau_c$

- fast fluctuations
- small molecules
- short $\tau_c$

$G(\tau)$

$f(t)$
Power Spectral Densities

The Fourier transform of an exponential is a Lorentzian line. The Fourier transform of the correlation function exponential is called the spectral density, \( J(\omega) \)

\[
\text{exp}(- |\tau| / \tau_c) \xrightarrow{\text{FT}} 2\tau_c / (1 + \omega^2\tau_c^2) = J(\omega)
\]
Power Spectral Densities

The random fluctuating fields produce a function composed of a range of frequencies (not discrete frequencies)
- spectral density curve represents power versus frequency, or the concentration of fluctuating fields present at a given frequency, or probability that motion of a given frequency exists, etc.
- the area under the curve is conserved

\[ \omega_0 \]
\[ \tau_c \text{ long, low power, long } T_1 \]
\[ \tau_c \text{ moderate, higher power, short } T_1 \]
\[ \tau_c \text{ short, low power, long } T_1 \]
Spectral Density and Relaxation

In order to cause the transitions necessary to promote relaxation, the spectral density must have frequency components at the Larmor frequency

- $T_1$ has a complex $\tau_c$ dependence
- $T_2$ depends on $J(\omega)$ at $\omega = 0$, and it decreases monotonically with $\tau_c$

\[ \tau_c \text{ long, low power, long } T_1 \]
\[ \tau_c \text{ moderate, higher power, short } T_1 \]
\[ \tau_c \text{ short, low power, long } T_1 \]
Heteronuclear dipolar relaxation

\[ ^{13}\text{C}-^{1}\text{H} \quad \text{– for example} \]

\[ R_{1S} = 1/T_1 = (\mu_0 h^2 \gamma_I^2 \gamma_S^2 \tau_{IS}^{-6} / 64\pi^3) (J(\omega_I - \omega_S) + 3J(\omega_S) + 6J(\omega_I + \omega_S)) \]

\[ R_{2S} = 1/T_2 = \]

\[ (\mu_0 h^2 \gamma_I^2 \gamma_S^2 \tau_{IS}^{-6} / 128\pi^3) (4J(0) + J(\omega_I - \omega_S) + 3J(\omega_S) + 6J(\omega_I) + 6J(\omega_I + \omega_S)) \]

\[ J(\omega) = (2/5)\tau_c/(1 + \omega^2 \tau_c^2) \]

Note: if \( \tau_c \) is small (small molecule), \( R_{1S} = R_{2S} \).

if \( \tau_c \) is large, \( R_{2S} \gg R_{1S} \)

Note: if given \( R_{1S} \) and \( R_{2S} \) for a molecule with a spin pair at a known distance (0.11 nm for C-H) can use Stoke’s formula to calculate molecule size
TROSY
Transverse Relaxation Optimized Spectroscopy

Relaxation by $T_2$ limits the size of macromolecules that can be studied by NMR.

- large molecule, long $\tau_c$, large $J(\omega) \rightarrow$ large $R_2$/short $T_2 \rightarrow$
  very broad line widths and poor S/N

- the mechanism for $T_2$ relaxation includes contributions from both dipole-dipole coupling and chemical shift anisotropy

- sometimes, constructive interference of the dipole-dipole coupling and CSA contributions can be effected, thus increasing $T_2$ and dramatically improving S/N
TROSY, Example

In a decoupled $^1$H, $^{15}$N HSQC spectrum, each peak is an average of the four multiplet components.

The S/N and line widths of the individual multiplet components are very different: each has different contributions from CSA and dipole-dipole coupling to $T_2$.

TROSY selects for one of the components - for this component, the CSA and dipole-dipole contributions to $T_2$ cancel one another (highest S/N).

R\textsubscript{2\alpha} and R\textsubscript{2\beta} are the transverse relaxation rates of the narrow and broad components of the $^{15}\text{N}$ doublet, respectively

$$R_{2\alpha} = (p - \delta_N)^2(4J(0) + 3J(\omega_N)) + p^2(J(\omega_{H-\omega_N}) + 3J(\omega_H) + 6J(\omega_{H+\omega_N})) + 3\delta_H^2J(\omega_H)$$

$$R_{2\beta} = (p + \delta_N)^2(4J(0) + 3J(\omega_N)) + p^2(J(\omega_{H-\omega_N}) + 3J(\omega_H) + 6J(\omega_{H+\omega_N})) + 3\delta_H^2J(\omega_H)$$

$p =$ dipole-dipole contribution

$\delta_N =$ CSA contribution $\propto B_0$

-as $\delta_N \rightarrow p$, the dipole-dipole and CSA contributions to $R_{2\alpha}$ cancel, $T_2$ increases, and the line width decreases
$^{15}\text{N} \text{ CSA and } ^1\text{H}-^{15}\text{N} \text{ Dipole Interactions Interfere}$

$^{15}\text{N}$

$^1\text{H}$

$B'$

$\alpha$ - fields interfere

$\nu - ^{15}\text{N}$

$\beta$ - fields reinforce

width

$\nu_0$

$1 \text{ GHz}$
α-Methyl Mannose Bound to Mannose Binding Protein
HSQC and TROSY of $^2$H, $^{15}$N-labeled mannose binding protein