

# The Nuclear Overhauser Effect

Overhauser, A.W. (1953) Paramagnetic Relaxation in Metals. *Phys. Rev.* **89**, 689-700.

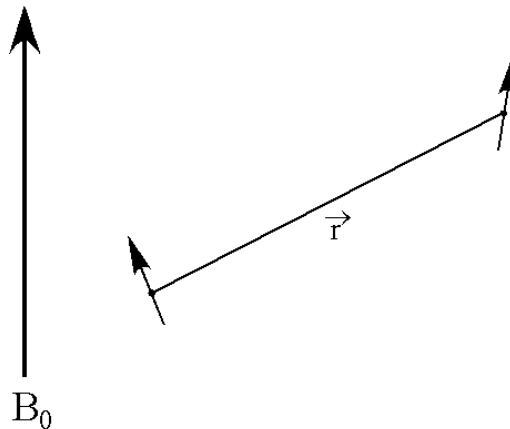
Solomon, I. (1955) Relaxation Processes in a System of Two Spins. *Phys. Rev.* **99**, 559-565.

Noggle, J.H. and Schirmer, R.E. (1971) *The Nuclear Overhauser Effect*. Academic Press, New York.

Neuhaus, D. and Williamson, M.P. (1989). *The Nuclear Overhauser Effect in Structural and Conformational Analysis*. VCH, New York.

## Relaxation Mechanisms

Recall that as molecule tumble in solution, the magnitude of the vector that between the two dipoles remains constant, but the orientation will clearly change, leading to a modulation of the dipole-dipole energy.



Recall also that the interaction energy depends upon the cube of the internuclear distance and the orientation between the two spins and the applied magnetic field.

The manner in which the orientation changes in time may be analyzed by considering the projection of a vector in the molecular frame over time. The expression that accounts for this behavior is called an *autocorrelation function* (or simply the correlation function), and is usually written as  $G(\tau)$ .

The exact form of the correlation depends upon the nature of the motion. For Brownian motion  $G(\tau)$  may be suitably modeled as an exponential decay. The correlation function may thus be written as:

$$G(\tau) = e^{-t/\tau} \quad \tau \approx \frac{\eta V_{eff}}{k_b T}$$

wherein  $\eta$  is the solution viscosity,  $V_{eff}$  is the effective molecular volume,  $k_b$  is Boltzmann's constant and  $T$  is the temperature in K.

The Fourier transform of the correlation function tells us about the frequency distribution the motions and is called the *spectral density function* and is usually written as  $J(\omega)$ :

$$J(\omega) = \frac{C\tau}{1 + \omega^2\tau^2}$$

Recall that the Fourier transform of an exponential function is a *Lorentzian function*.

The value of the spectral density thus depends both on the transition frequency and the correlation time:

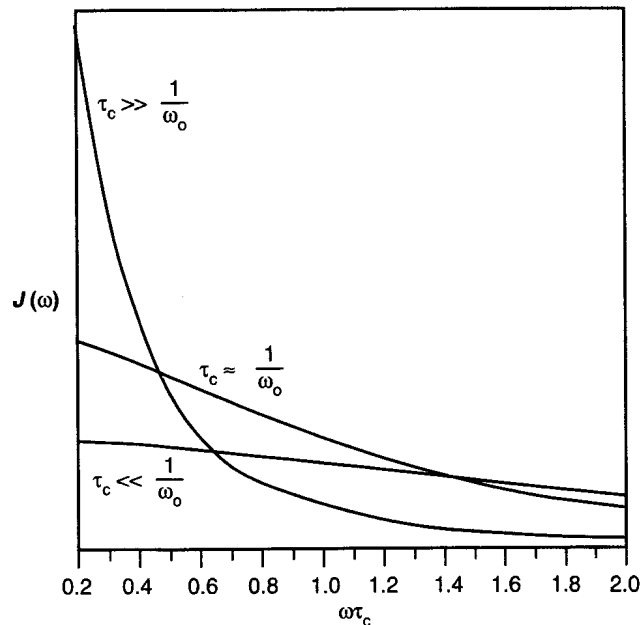


Figure 8.1 from Becker, E.D. (2000) High Resolution NMR. Academic Press, New York

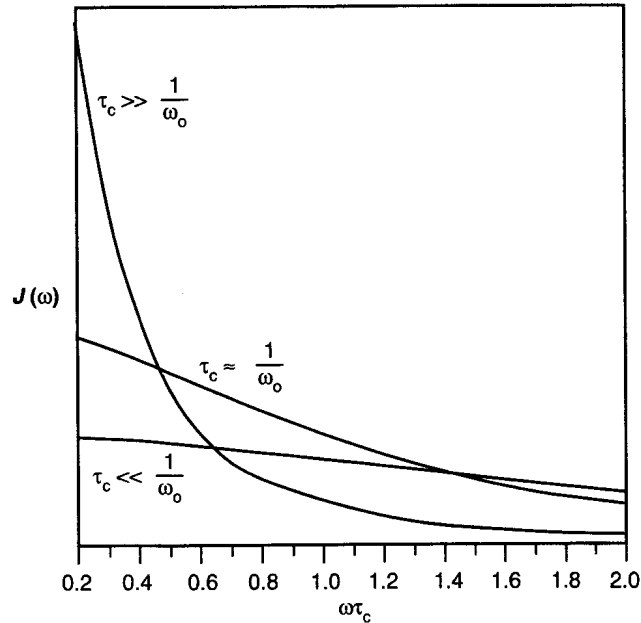


Figure 8.1 from Becker, E.D. (2000) High Resolution NMR. Academic Press, New York

Notice that for  $\tau \gg 1/\omega$  that the value of  $J(\omega)$  decreases rapidly as a function of  $\omega\tau$ , whereas for  $\tau \ll 1/\omega$  the opposite is true.

In fact, in the limit as  $\omega\tau \gg 1$ ,  $J(\omega)$  is directly proportional to  $\tau$ .

## Transition Rates

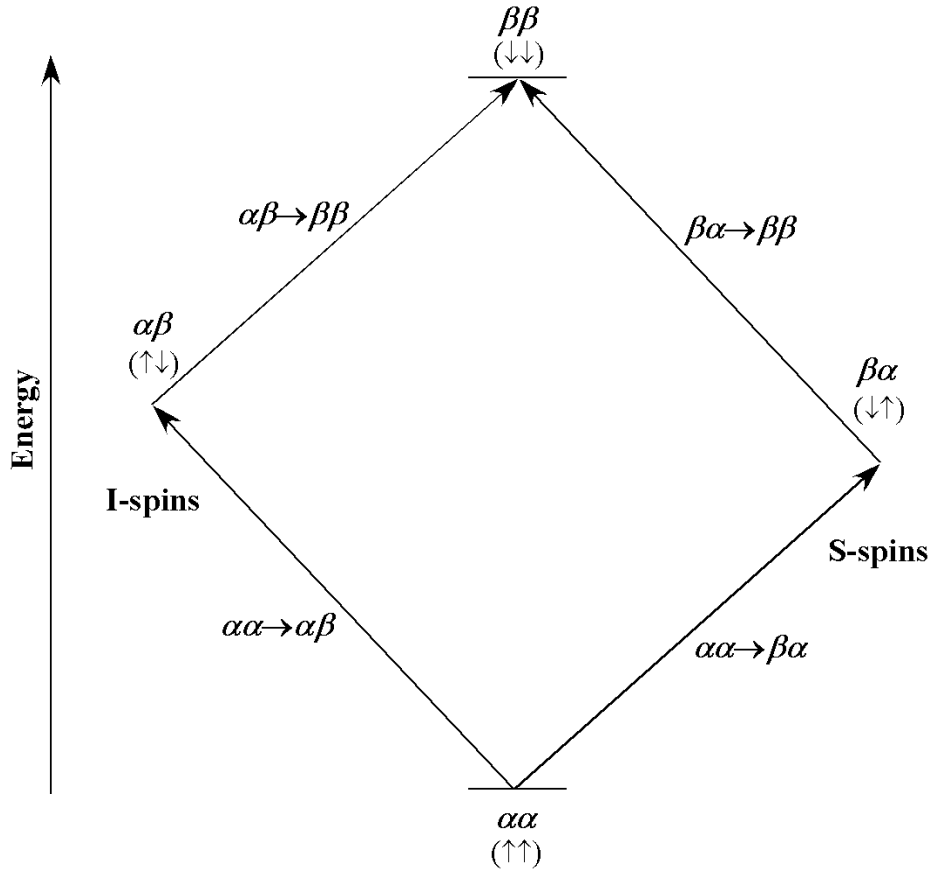
Perturbation theory provides us with an expression generally known as *Fermi's golden rule*:

$$\begin{aligned}
 W_{i \rightarrow j} &\equiv \text{transition probability per unit time} \\
 &= \frac{1}{\hbar^2 t} \int_0^t \int_0^t \langle i | H(t') | j \rangle \langle j | H(t'') | i \rangle e^{-i\omega_{ij}(t'-t'')} dt' dt''
 \end{aligned}$$

wherein  $\omega_{ij}$  is the transition frequency ( $\text{rad s}^{-1}$ ).

The expression is the Fourier transform of the Hamiltonian composed with itself and evaluated over time.

The  $W_{ij}$  are defined with respect to our energy level diagram:



If we substitute the dipolar coupling Hamiltonian into the transition rate expression, we will obtain the following:

$$W_0 = W(\alpha\beta \leftrightarrow \beta\alpha) \propto \frac{\gamma_I \gamma_S}{r_{IS}^6} J(\omega_I - \omega_S) \approx \frac{\gamma_I \gamma_S}{2r_{IS}^6} J(0) \text{ when } I=S$$

$$W_{1I} = W(\alpha\alpha \leftrightarrow \beta\alpha) = W(\alpha\beta \leftrightarrow \beta\beta) \propto \frac{\gamma_I \gamma_S}{r_{IS}^6} J(\omega_I)$$

$$W_{1S} = W(\alpha\alpha \leftrightarrow \alpha\beta) = W(\beta\alpha \leftrightarrow \beta\beta) \propto \frac{\gamma_I \gamma_S}{r_{IS}^6} J(\omega_S)$$

$$W_2 = W(\alpha\alpha \leftrightarrow \beta\beta) \propto \frac{\gamma_I \gamma_S}{r_{IS}^6} J(\omega_I + \omega_S)$$

We may write the rates of longitudinal and transverse relaxation in term of these transition rates to obtain the following expressions:

$$R_1^H(^1\text{H}, ^{13}\text{C}) = \frac{1}{T_1} = \frac{C\gamma_H^2\gamma_C^2}{r_{\text{HC}}^6} [J(\omega_H - \omega_C) + 2J(\omega_H) + J(\omega_H + \omega_C)]$$

$$R_1^C(^1\text{H}, ^{13}\text{C}) = \frac{1}{T_1} = \frac{C\gamma_H^2\gamma_C^2}{r_{\text{HC}}^6} [J(\omega_H - \omega_C) + 2J(\omega_C) + J(\omega_H + \omega_C)]$$

$$R_1^H(^1\text{H}, ^1\text{H}) = \frac{1}{T_1} = \frac{C\gamma_H^4}{r_{\text{HH}}^6} [J(\omega_H) + 4J(2\omega_H)]$$

$$R_2^H(^1\text{H}, ^{13}\text{C}) = \frac{1}{T_2} = \frac{C\gamma_H^2\gamma_C^2}{2r_{\text{HC}}^6} [4J(0) + J(\omega_H - \omega_C) + 3J(\omega_H) + 6J(\omega_C) + 6J(\omega_H + \omega_C)]$$

$$R_2^C(^1\text{H}, ^{13}\text{C}) = \frac{1}{T_2} = \frac{C\gamma_H^2\gamma_C^2}{2r_{\text{HC}}^6} [4J(0) + J(\omega_H - \omega_C) + 3J(\omega_C) + 6J(\omega_H) + 6J(\omega_H + \omega_C)]$$

$$R_2^H(^1\text{H}, ^1\text{H}) = \frac{1}{T_2} = \frac{C\gamma_H^4}{2r_{\text{HH}}^6} [3J(0) + 5J(\omega_H) + 2J(2\omega_H)]$$

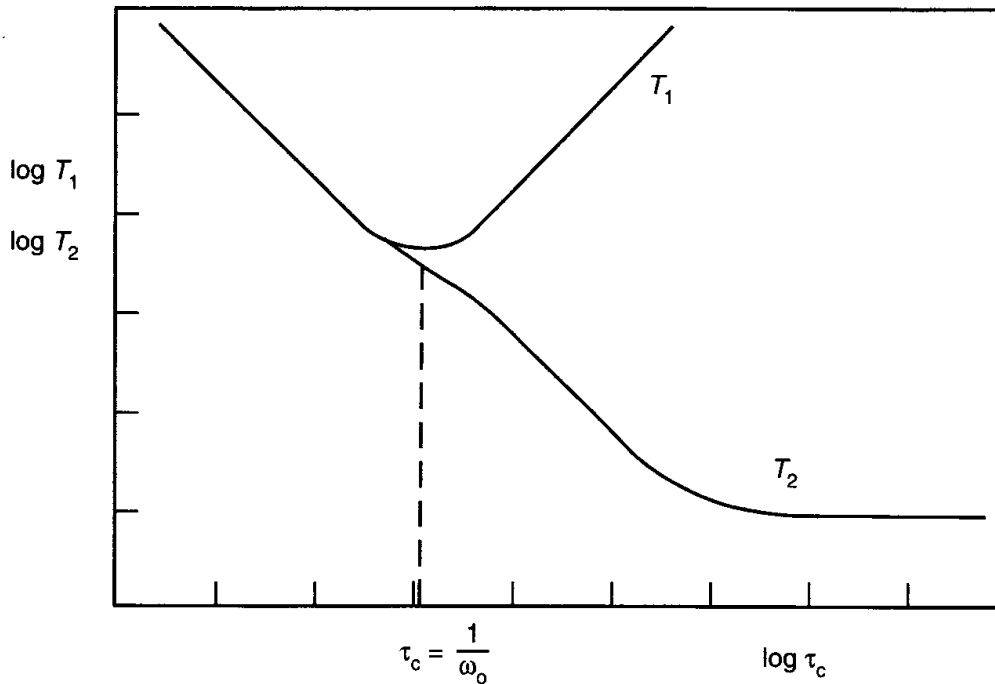
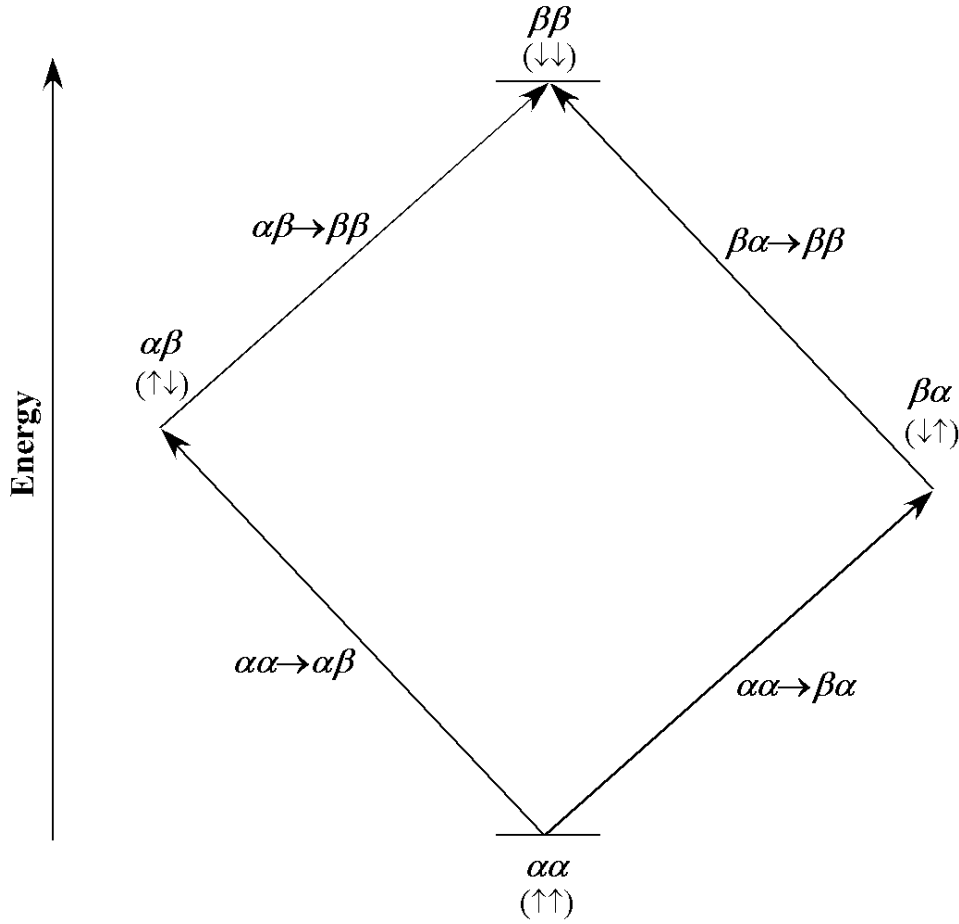


Figure 8.2 from Becker, E.D. (2000) High Resolution NMR. Academic Press, New York.

The change in the populations of individual energy levels depend upon the transition rates as shown below:



$$\frac{d}{dt}n_{\alpha\alpha} = -(W_{1I} + W_{1S} + W_2)(n_{\alpha\alpha} - n_{\alpha\alpha}^0) + W_2(n_{\beta\beta} - n_{\beta\beta}^0) + W_{1I}(n_{\beta\alpha} - n_{\beta\alpha}^0) + W_{1S}(n_{\alpha\beta} - n_{\alpha\beta}^0)$$

$$\frac{d}{dt}n_{\alpha\beta} = W_{1S}(n_{\beta\alpha} - n_{\beta\alpha}^0) - (W_{1I} + W_{1S} + W_0)(n_{\alpha\beta} - n_{\alpha\beta}^0) + W_0(n_{\beta\alpha} - n_{\beta\alpha}^0) + W_{1I}(n_{\beta\beta} - n_{\beta\beta}^0)$$

$$\frac{d}{dt}n_{\beta\alpha} = W_{1I}(n_{\alpha\alpha} - n_{\alpha\alpha}^0) + W_0(n_{\alpha\beta} - n_{\alpha\beta}^0) - (W_{1I} + W_{1S} + W_0)(n_{\beta\alpha} - n_{\beta\alpha}^0) + W_{1S}(n_{\beta\beta} - n_{\beta\beta}^0)$$

$$\frac{d}{dt}n_{\beta\beta} = W_2(n_{\alpha\alpha} - n_{\alpha\alpha}^0) + W_{1I}(n_{\alpha\beta} - n_{\alpha\beta}^0) + W_{1S}(n_{\beta\alpha} - n_{\beta\alpha}^0) - (W_{1I} + W_{1S} + W_2)(n_{\beta\beta} - n_{\beta\beta}^0)$$

The macroscopic magnetic moments,  $I_I$  and  $I_S$  are what we actually observe:

$$I_z^I \propto (n_{\alpha\alpha} - n_{\beta\alpha}) + (n_{\alpha\beta} - n_{\beta\beta})$$

$$I_z^S \propto (n_{\alpha\alpha} - n_{\alpha\beta}) + (n_{\beta\alpha} - n_{\beta\beta})$$

And by substituting the expression for  $I_I$  and  $I_S$  into the one above it we obtain:

$$\begin{aligned}\frac{d}{dt} I_z^I &= -(W_0 + 2W_{1I} + W_2)(I_z^I - I_{z0}^I) - (W_2 - W_0)(I_z^S - I_{z0}^S) \\ \frac{d}{dt} I_z^S &= -(W_0 + 2W_{1S} + W_2)(I_z^S - I_{z0}^S) - (W_2 - W_0)(I_z^I - I_{z0}^I)\end{aligned}$$

wherein  $I_{z0}^I$  and  $I_{z0}^S$  are the equilibrium values of  $I_z^I$  and  $I_z^S$ .

Solomon made the following substitution:

$$\begin{aligned}\rho &= W_0 + 2W_{1I} + W_2 \\ \rho' &= W_0 + 2W_{1S} + W_2 \\ \sigma &= W_2 - W_0\end{aligned}$$

and rewrote our kinetic expressions as shown below:

$$\begin{aligned}\frac{d}{dt} I_z^I &= -\rho(I_z^I - I_{z0}^I) - \sigma(I_z^S - I_{z0}^S) \\ \frac{d}{dt} I_z^S &= -\rho'(I_z^S - I_{z0}^S) - \sigma(I_z^I - I_{z0}^I)\end{aligned}$$

Thus, a nonzero value of  $\sigma$  implies the presence of relaxation processes that link  $I_z^I$  and  $I_z^S$ .

The *steady-state* is defined as the condition under which the system comes into equilibrium. This state can be generated in either the absence or presence of perturbation and simply implies that no *net* change occurs in the system over time.

Kinetically, we may write that

$$\begin{aligned}\frac{d}{dt} I_z^I &= -\rho(I_z^I - I_{z0}^I) - \sigma(I_z^S - I_{z0}^S) = 0 \\ \frac{d}{dt} I_z^S &= -\rho'(I_z^S - I_{z0}^S) - \sigma(I_z^I - I_{z0}^I) = 0\end{aligned}$$

$$0 = -\rho(I_z^I - I_{z0}^I) - \sigma(I_z^S - I_{z0}^S)$$

$$0 = -\rho'(I_z^S - I_{z0}^S) - \sigma(I_z^I - I_{z0}^I)$$

Consider the S-spins and imagine what will happen if we supply RF irradiation at the I-spin frequency.

We have seen that such irradiation causes the number of spins in the  $\alpha$  and  $\beta$  states of the I-spins to become equal. This means that  $I_z^I$  is equal to zero and we write that

$$0 = -\rho'I_z^S + \rho'I_{z0}^S + \sigma(I_{z0}^I)$$

or equivalently that

$$I_z^S = I_{z0}^S + \left(\frac{\sigma}{\rho'}\right)I_{z0}^I$$

This result is known as the *nuclear Overhauser effect*, NOE.

The ratio of  $\sigma$  and  $\rho$  is known as the *Overhauser enhancement* factor and is symbolized with  $\eta$ :

$$\eta = \frac{\gamma_I}{\gamma_S} \left(\frac{\sigma}{\rho'}\right)$$

The Overhauser enhancement thus depends upon the transition rates among the energy levels:

$$\eta = \left(\frac{\sigma}{\rho'}\right) = \frac{\gamma_I}{\gamma_S} \frac{W_2 - W_0}{W_0 + 2W_{1S} + W_2}$$

$$W_0 = W(\alpha\beta \leftrightarrow \beta\alpha) = W(\beta\alpha \leftrightarrow \alpha\beta) = \frac{\hbar^2 \gamma_I \gamma_S}{10r_{IS}^6} \left( \frac{\tau}{1 + (\omega_I - \omega_S)^2 \tau^2} \right)$$

$$W_{1I} = W(\alpha\alpha \leftrightarrow \beta\alpha) = W(\alpha\beta \leftrightarrow \beta\beta) = \frac{3\hbar^2 \gamma_I \gamma_S}{20r_{IS}^6} \left( \frac{\tau}{1 + \omega_I^2 \tau^2} \right)$$



$$W_{1s} = W(\alpha\alpha \leftrightarrow \alpha\beta) = W(\beta\alpha \leftrightarrow \beta\beta) = \frac{3\hbar^2\gamma_I\gamma_S}{20r_{IS}^6} \left( \frac{\tau}{1 + \omega_S^2\tau^2} \right)$$

$$W_2 = W(\alpha\alpha \leftrightarrow \beta\beta) = \frac{6\hbar^2\gamma_I\gamma_S}{10r_{IS}^6} \left( \frac{\tau}{1 + (\omega_I + \omega_S)^2\tau^2} \right)$$

## Fast Motion Limit

For small molecules, in simple aqueous or organic solution the *fast motion limit* applies:

$$\omega^2\tau^2 \ll 1$$

In which case our expression become:

$$W_0 = W(\alpha\beta \leftrightarrow \beta\alpha) = W(\beta\alpha \leftrightarrow \alpha\beta) = \frac{\hbar^2\gamma_I\gamma_S}{10r_{IS}^6} \tau$$

$$W_{1I} = W(\alpha\alpha \leftrightarrow \beta\alpha) = W(\alpha\beta \leftrightarrow \beta\beta) = \frac{3\hbar^2\gamma_I\gamma_S}{20r_{IS}^6} \tau$$

$$W_{1S} = W(\alpha\alpha \leftrightarrow \alpha\beta) = W(\beta\alpha \leftrightarrow \beta\beta) = \frac{3\hbar^2\gamma_I\gamma_S}{20r_{IS}^6} \tau$$

$$W_2 = W(\alpha\alpha \leftrightarrow \beta\beta) = \frac{6\hbar^2\gamma_I\gamma_S}{10r_{IS}^6} \tau$$

Under these circumstances  $\eta$  becomes:

$$\eta = \left( \frac{\sigma}{\rho'} \right) = \frac{W_2 - W_0}{W_0 + 2W_{1S} + W_2} = \frac{\gamma_I}{2\gamma_S}$$

If our *I*-spins are  $^1\text{H}$  nuclei and our *S*-spins are  $^{13}\text{C}$  nuclei, then the expression suggests that we may substantially enhance the intensity of the *S*-spin by irradiating the *I*-spins.

$$\eta = \left( \frac{\sigma}{\rho'} \right) = \frac{\gamma_I}{2\gamma_S} = \frac{\gamma_H}{2\gamma_C} = \frac{267.522 \times 10^{-6} \text{ rad s}^{-1} \text{ T}^{-1}}{2(67.283 \times 10^{-6} \text{ rad s}^{-1} \text{ T}^{-1})} = 1.988$$

## Slow Motion Limit

For larger molecules a new set of approximations apply. For a pair of dipolar-coupled  $^1\text{H}$  nuclei ( $\gamma_I = \gamma_S$ ) in the limit as

$$\omega^2 \tau^2 \gg 1$$

the transition rates assume the following form:

$$W_0 = W(\alpha\beta \leftrightarrow \beta\alpha) = W(\beta\alpha \leftrightarrow \alpha\beta) = \frac{\hbar^2 \gamma_I \gamma_S}{10r_{IS}^6} \left( \frac{\tau}{1 + (\omega_I - \omega_S)^2 \tau^2} \right) = \frac{\hbar^2 \gamma_I \gamma_S}{10r_{IS}^6} \tau$$

$$W_{II} = W(\alpha\alpha \leftrightarrow \beta\alpha) = W(\alpha\beta \leftrightarrow \beta\beta) = \frac{3\hbar^2 \gamma_I \gamma_S}{20r_{IS}^6} \left( \frac{\tau}{1 + \omega_I^2 \tau^2} \right) \approx 0$$

$$W_{IS} = W_{II} = W(\alpha\alpha \leftrightarrow \alpha\beta) = W(\beta\alpha \leftrightarrow \beta\beta) = \frac{3\hbar^2 \gamma_I \gamma_S}{20r_{IS}^6} \left( \frac{\tau}{1 + \omega_S^2 \tau^2} \right) \approx 0$$

$$W_2 = W(\alpha\alpha \leftrightarrow \beta\beta) = \frac{6\hbar^2 \gamma_I \gamma_S}{10r_{IS}^6} \left( \frac{\tau}{1 + (\omega_I + \omega_S)^2 \tau^2} \right) \approx 0$$

The NOE for  $^1\text{H}$  nuclei in the slow motion limits is thus given by:

$$\eta = \left( \frac{\sigma}{\rho'} \right) = \frac{W_2 - W_0}{W_0 + 2W_{IS} + W_2} = \frac{W_2 - W_0}{W_0 + 2W_{II} + W_2} = \frac{-W_0}{W_0} \frac{\gamma_I}{\gamma_I} = -1$$

i.e., the enhancement produces a negative value.

In practice, since the gyromagnetic ratio for  $^{13}\text{C}$  nuclei is lower than that of protons the maximum heteronuclear enhancement is only achieved for large biological macromolecules at high applied magnetic field strength, i.e., at 11.74 (500 MHz  $^1\text{H}$ ), the Larmor frequency of  $^{13}\text{C}$  is approximately 125 MHz and given a correlation time of  $10^{-8}$  seconds we observe that

$$\omega\tau = 2\pi(125 \times 10^6 \text{ s}^{-1})(1 \times 10^{-8} \text{ s}) \approx 8$$

Thus for dipolar-coupled  $^1\text{H}$  spins ( $\gamma_I = \gamma_S$ ) the enhancement is -1, while for a  $^1\text{H}$ - $^{13}\text{C}$  pair the maximum enhancement is -4.

We have already seen that the NOE can be an effective approach to sensitivity enhancement. The utility of the NOE as a structural tool far surpasses its value as a spectral enhancement tool.

If NOE effects can be monitored at a numbers of molecular sites, the information obtained,  $^1\text{H}$ - $^1\text{H}$  distances, can be used to reconstruct the molecule of interest.

The most effective applications of the NOE make use of multidimensional techniques and may also involve resolution of the heteronuclear chemical shifts.