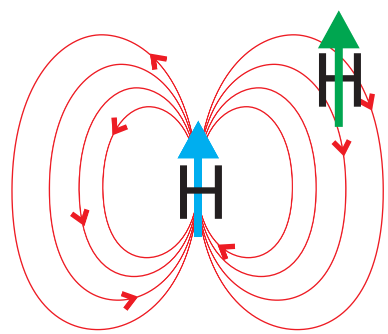
Relaxation and the Nuclear Overhauser Effect

- Relaxation rates depends 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.

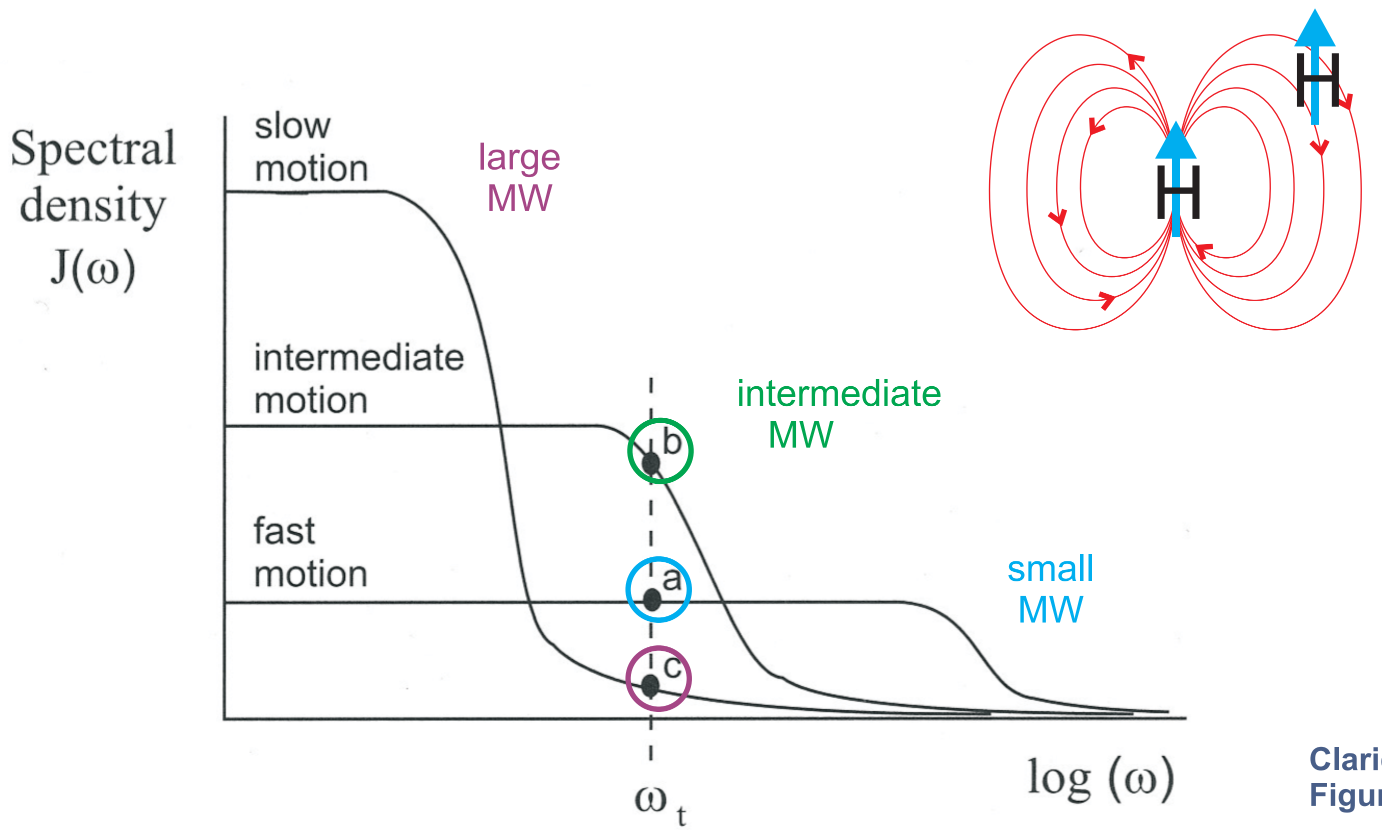
$$\Delta E_{dd} \propto \gamma_i^2 \gamma_j^2 \left\langle \sum_j \frac{\tau_c}{r_{ij}^6} \right\rangle_t$$



- 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.*

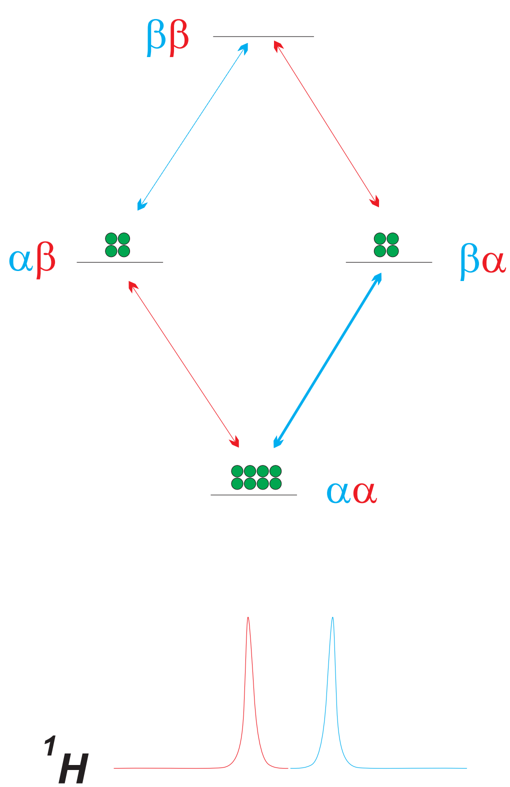


Claridge Figure 8.6

The Nuclear Overhauser Effect (2-spin system)

In NMR: excess population $\propto$ 1 - exp(-ΔE/RT) ~ ΔE

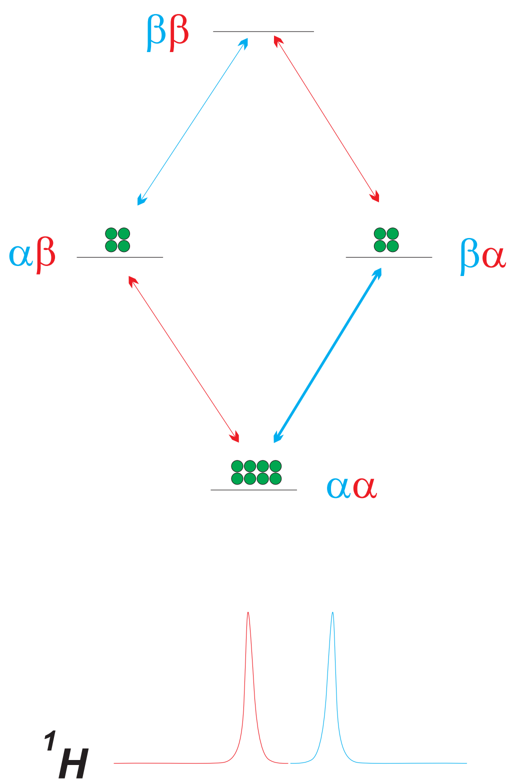
(see Claridge or Sanders&Hunter)



The Nuclear Overhauser Effect

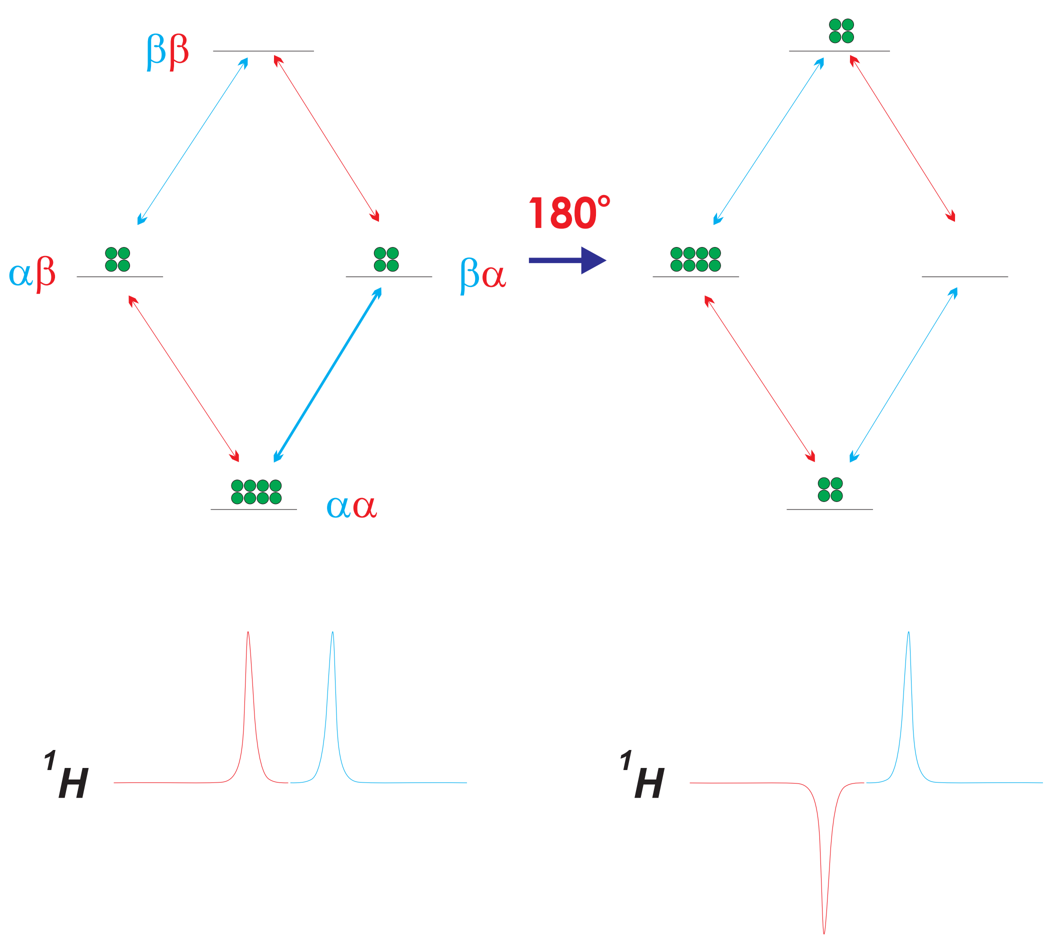
- **Through-space dipole-dipole interactions!**

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



The Nuclear Overhauser Effect

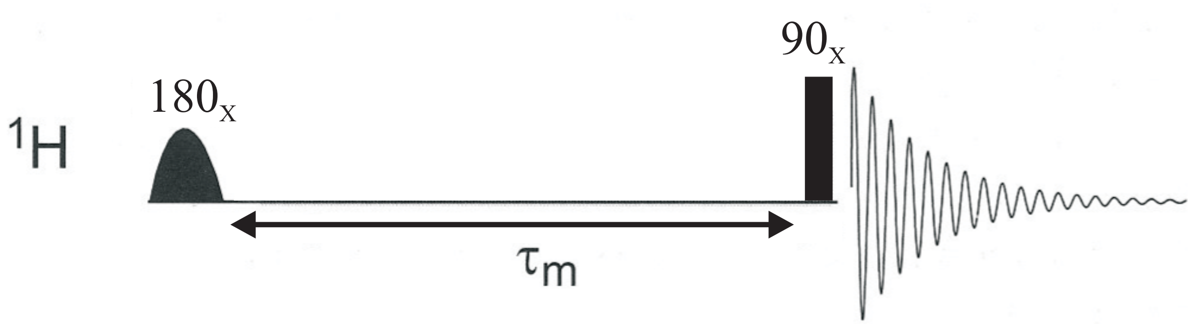
- Start with selective inversion:



NOEs in the Rotating Frame: NOEs

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

sel-NOESY



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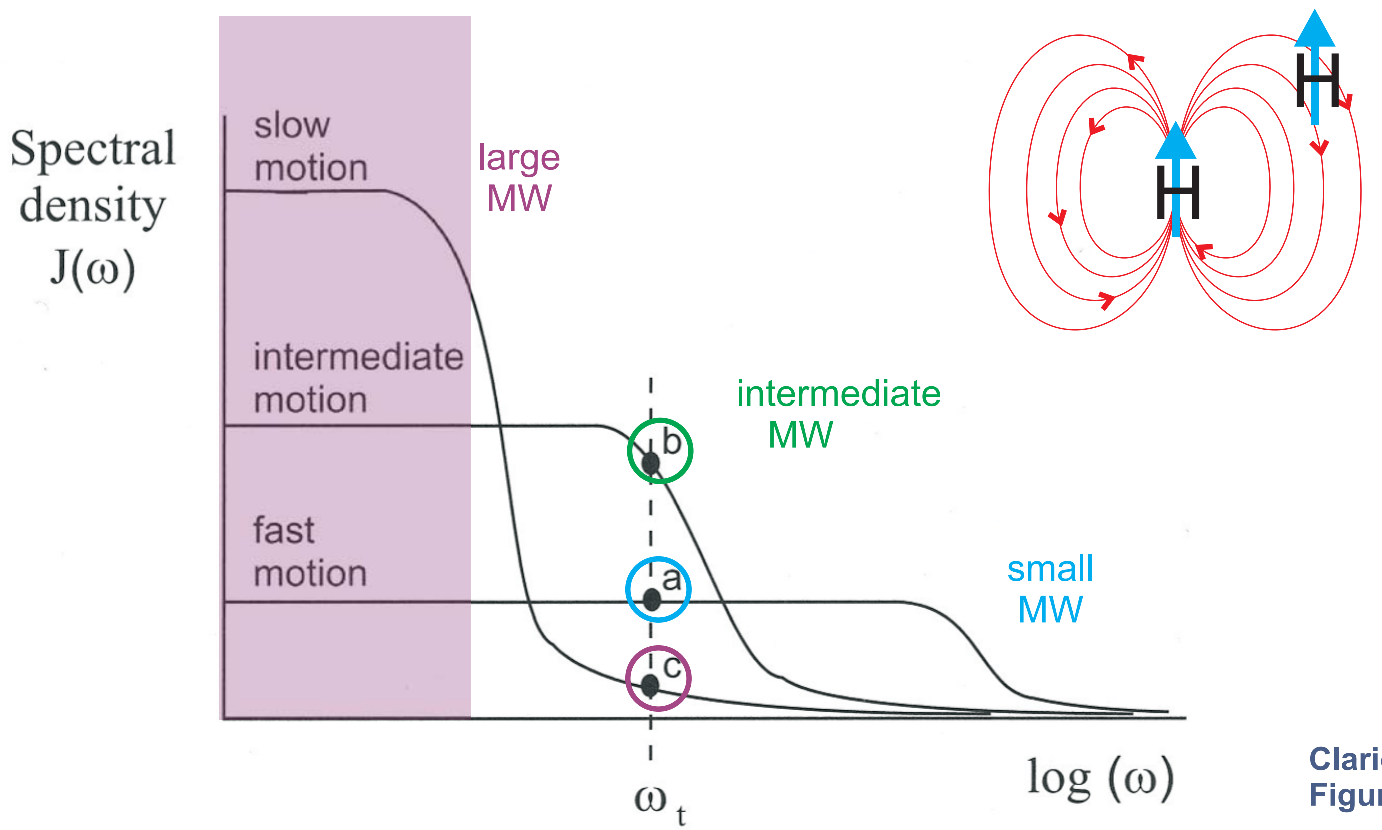
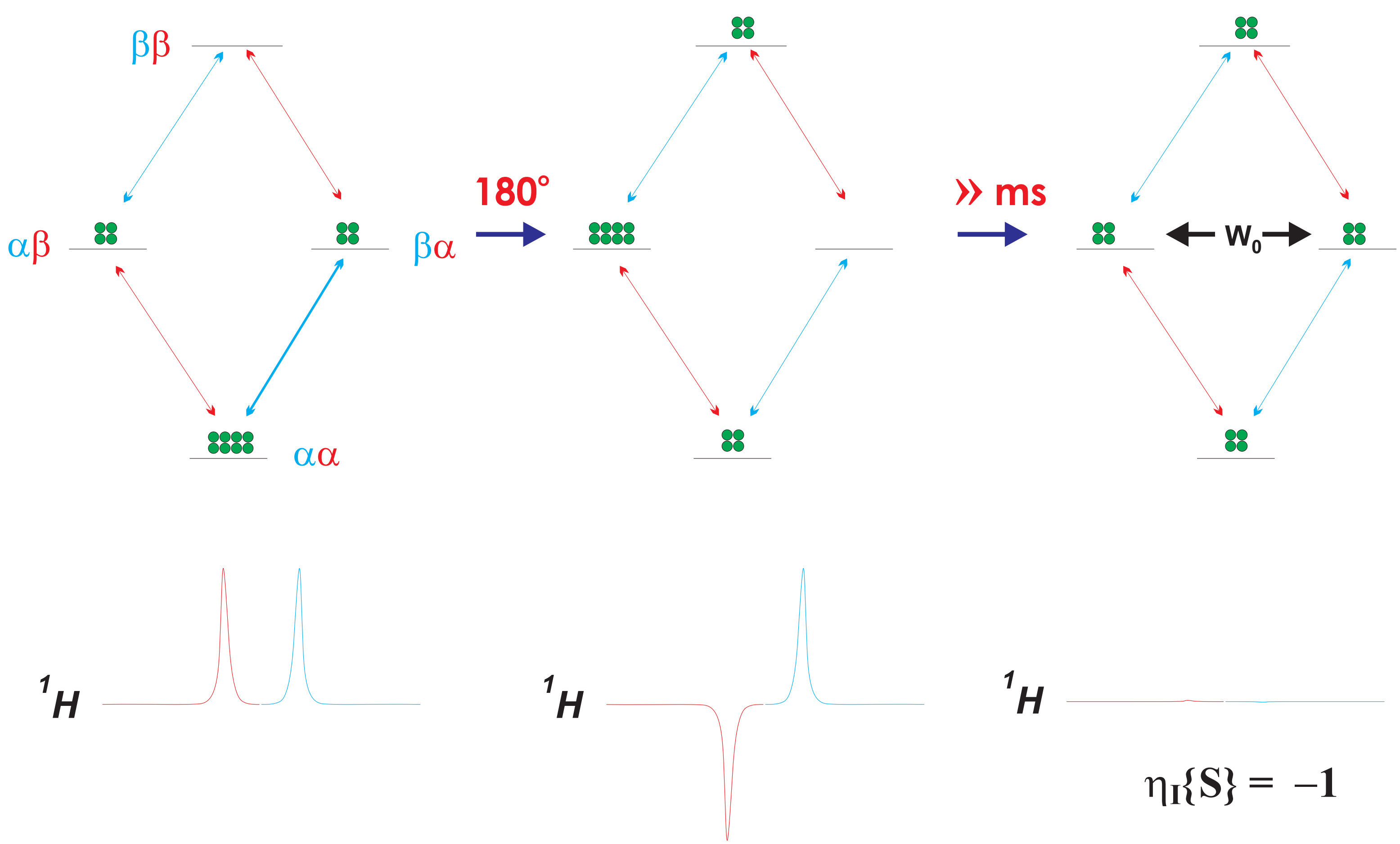


Claridge Figure 8.6

Negative NOEs: Large MW

- Slow motions are effective for small frequencies/energy differences:

  W_0 (ZQ) ⇨ negative NOE dominates for large MW



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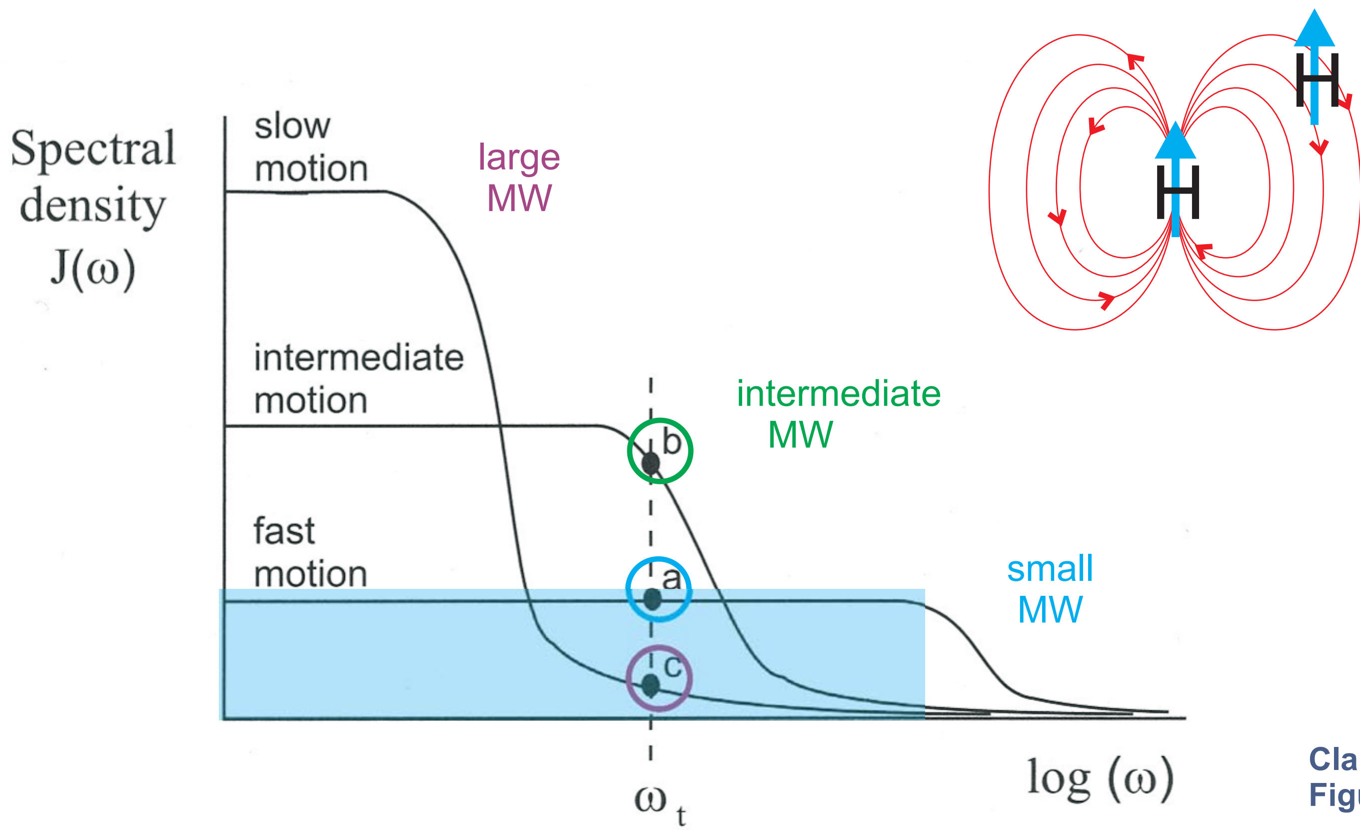
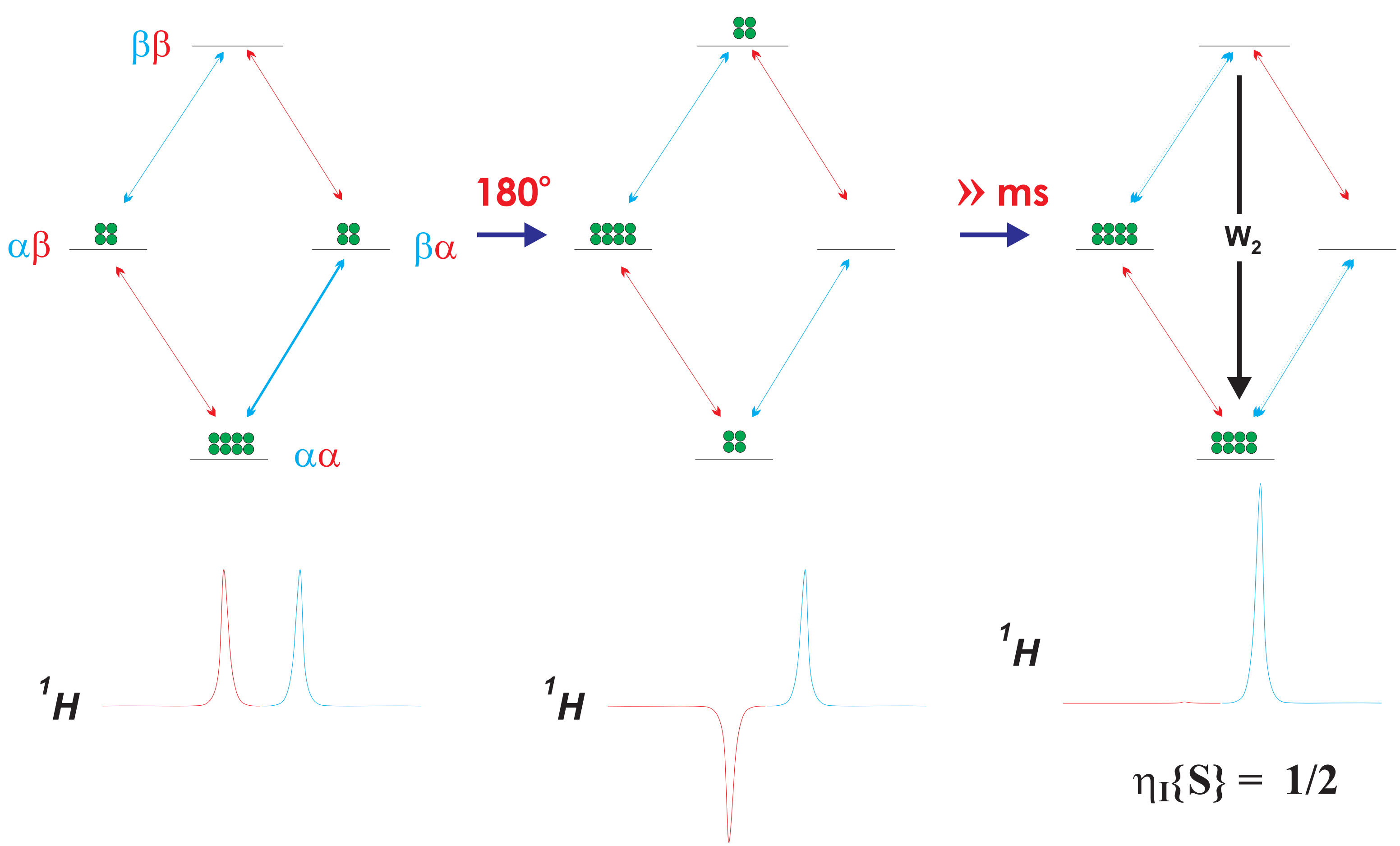


Claridge Figure 8.6

Positive NOEs: Small MW

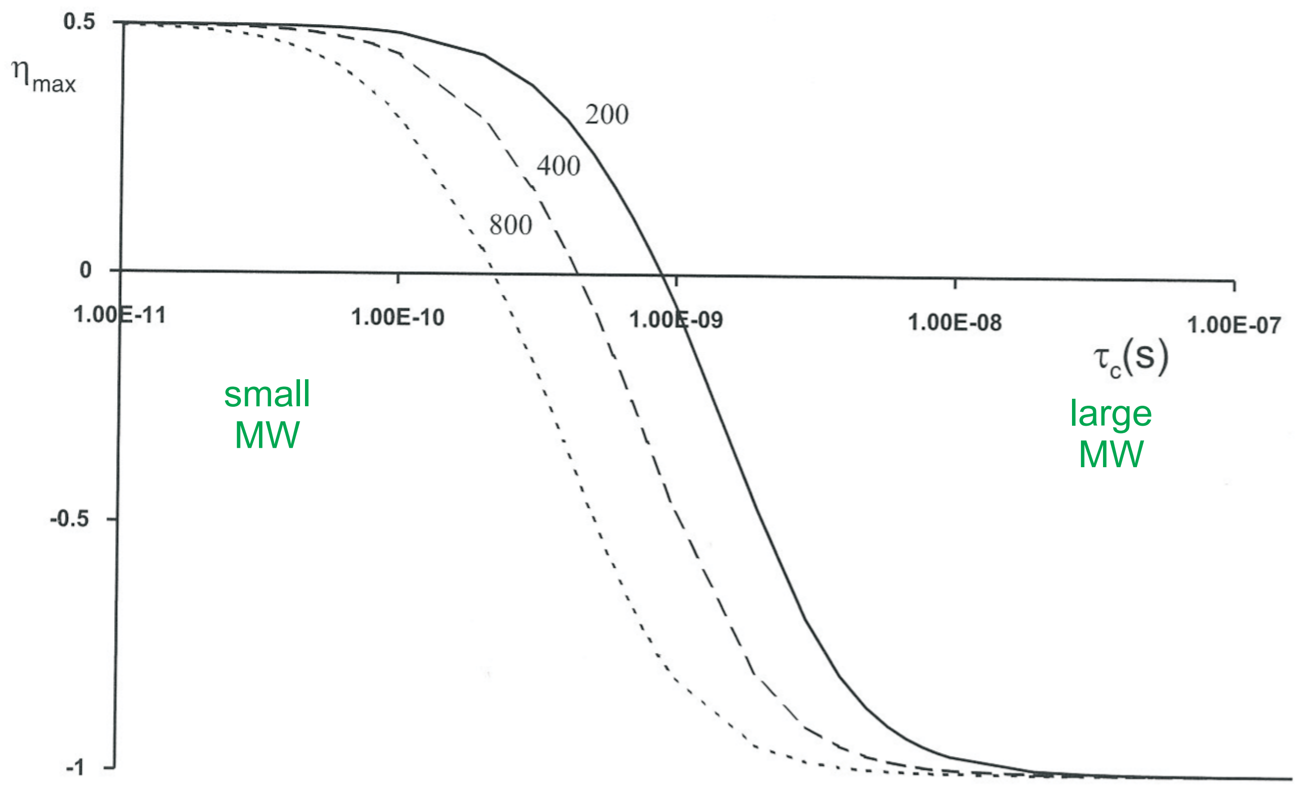
- Fast motions are effective for large frequencies/energy differences:

  W_2 (DQ) ⇨ positive NOE dominates for small MW



Zero NOEs: The Crossover Region

- Thus, NOEs go through a ***crossover region*** at intermediate MW, in the range 1000-5000 Da.



Claridge Figure 8.9

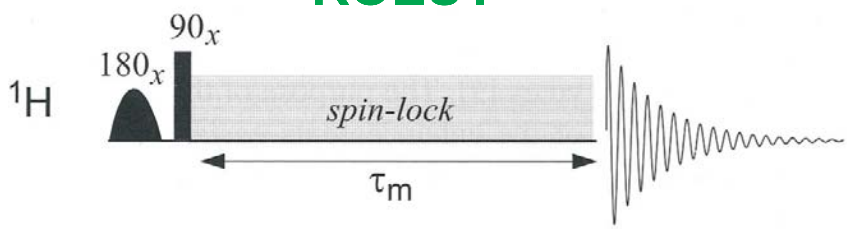
The ROESY Spin-Lock: Reducing the Effective Field

- 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.

Claridge Figure 8.24

ROESY

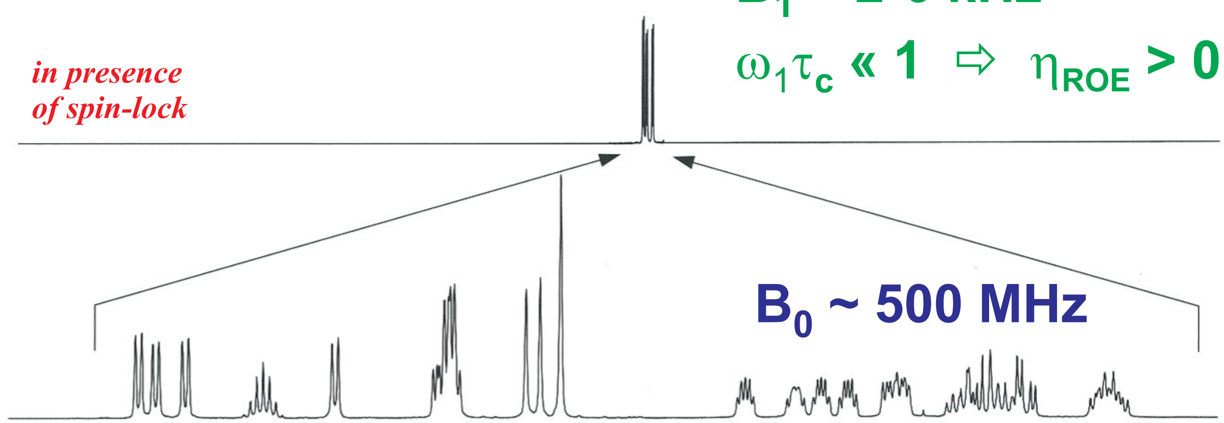


"All" molecules in solution tumble fast compared to 6 kHz!
ROEs are always positive.

B_1 ~ 2-6 kHz ⇨

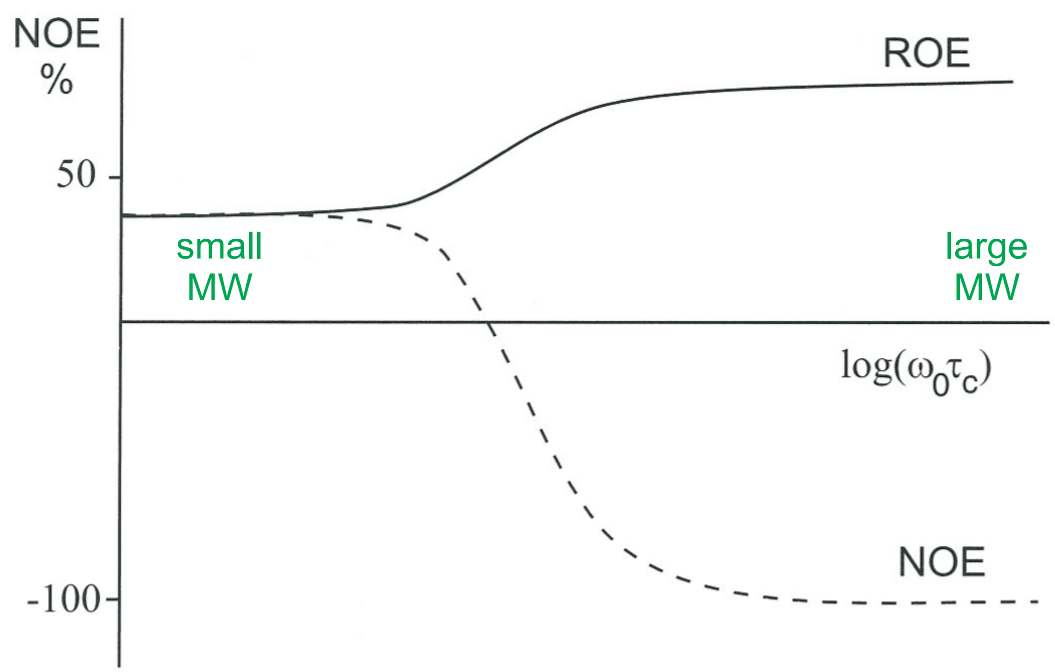
$\omega_1\tau_c$ « 1 ⇨ $\eta_{ROE} > 0$

Claridge 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).



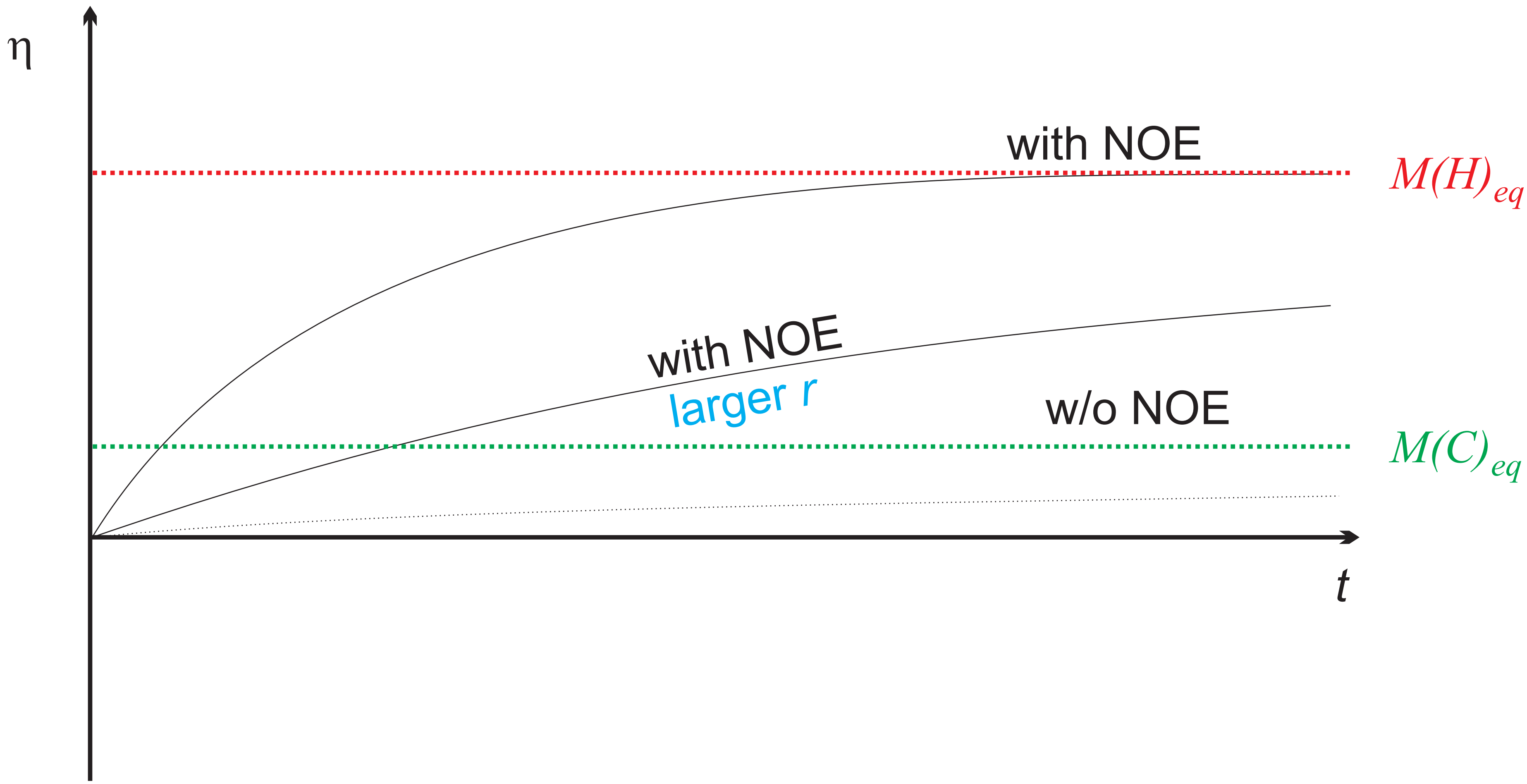
Review - ROESY

- 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.

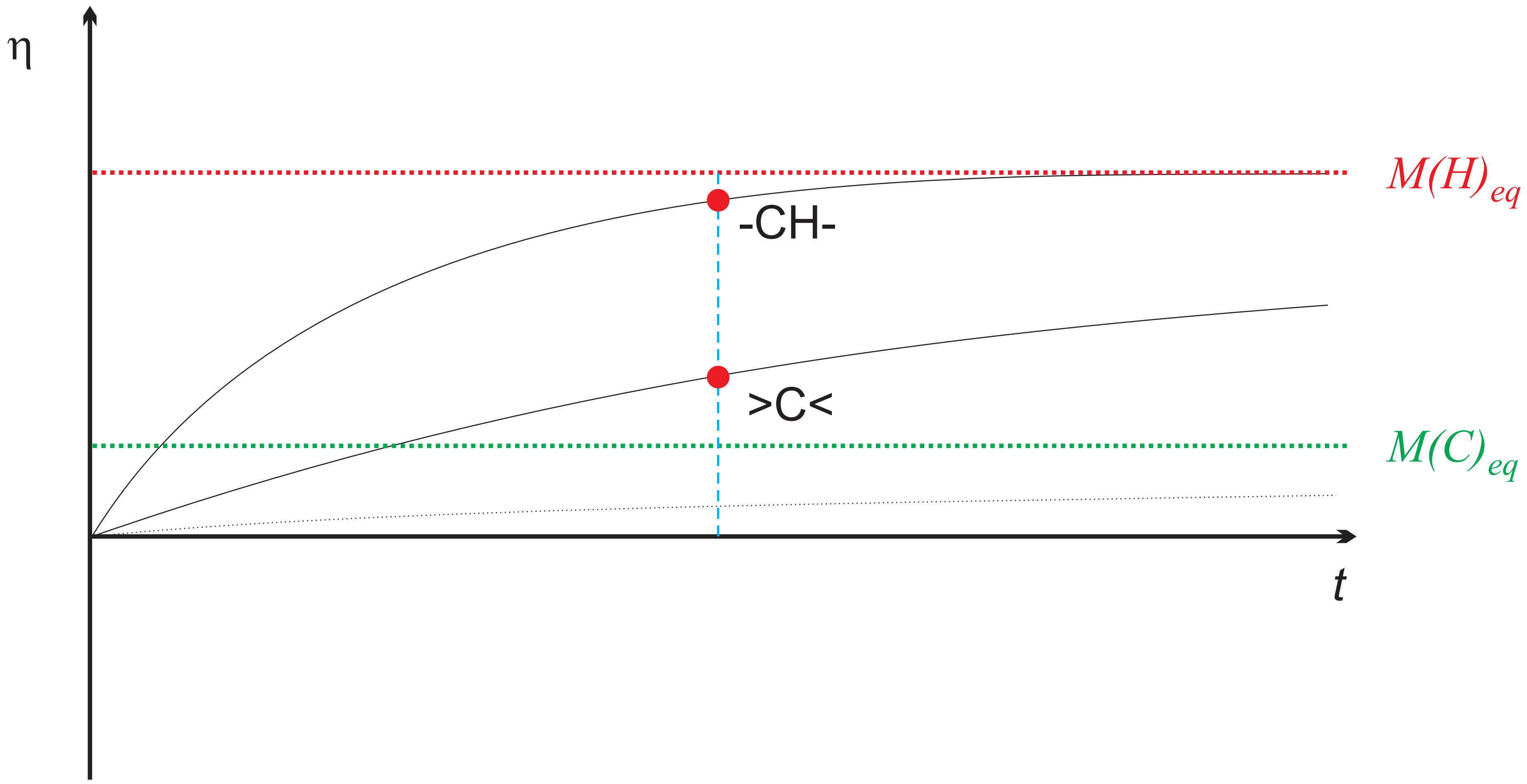


Claridge
Figure 8.23

Heteronuclear NOEs: Positive γ Nuclei

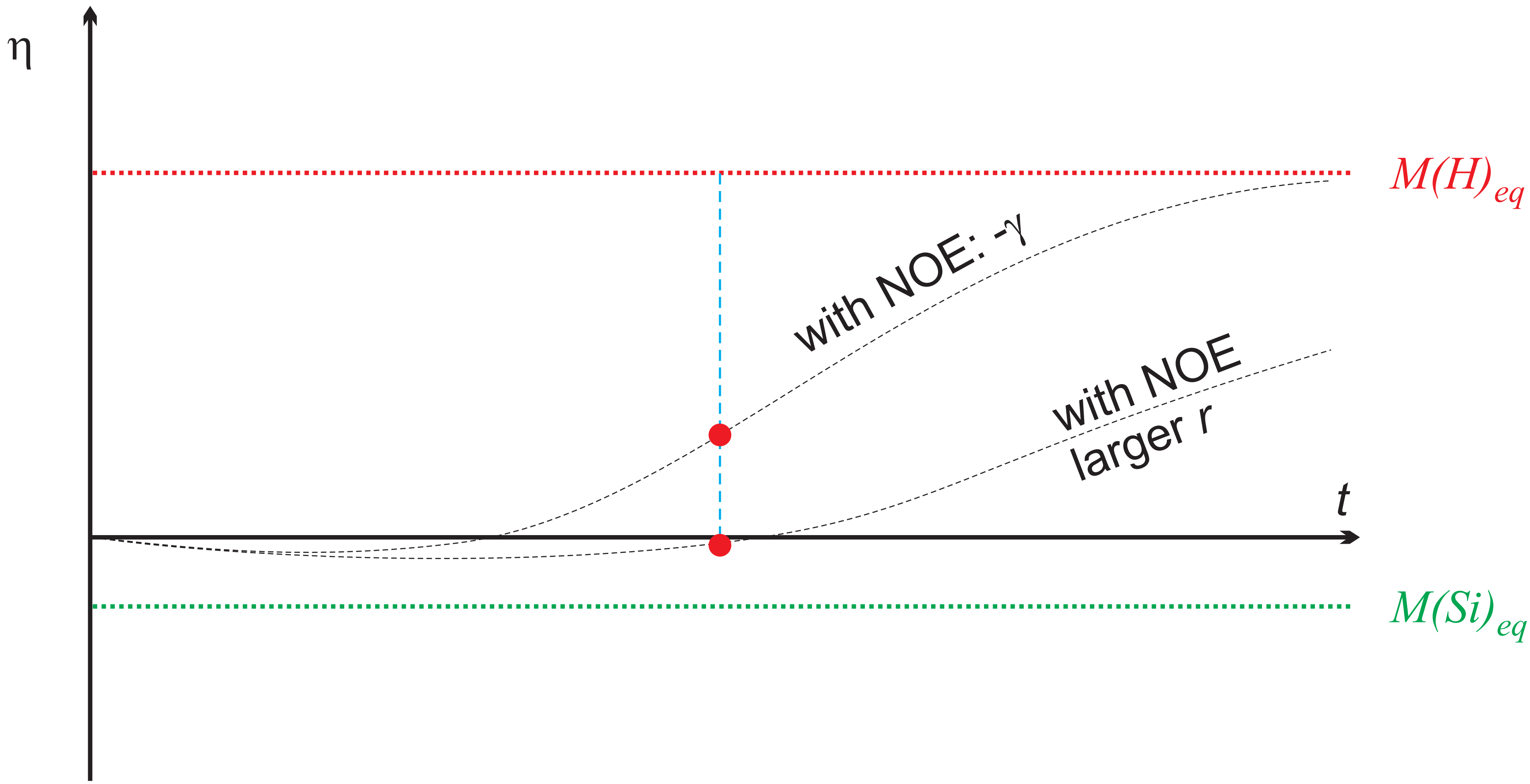


Heteronuclear NOEs: Positive γ Nuclei



Heteronuclear NOEs: Negative γ Nuclei

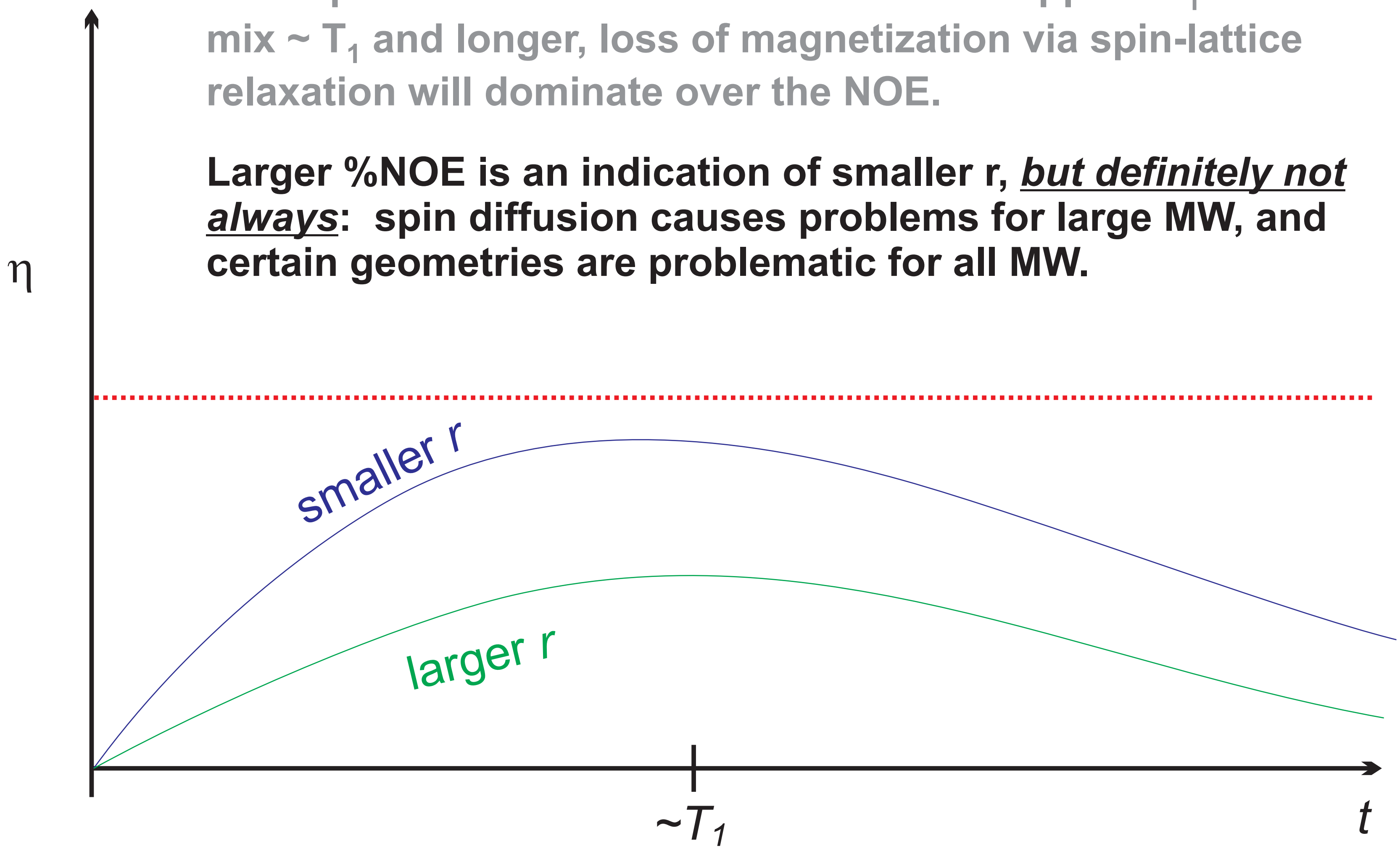
^{29}Si, ^{15}N, ^{119}Sn



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 ~ 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.



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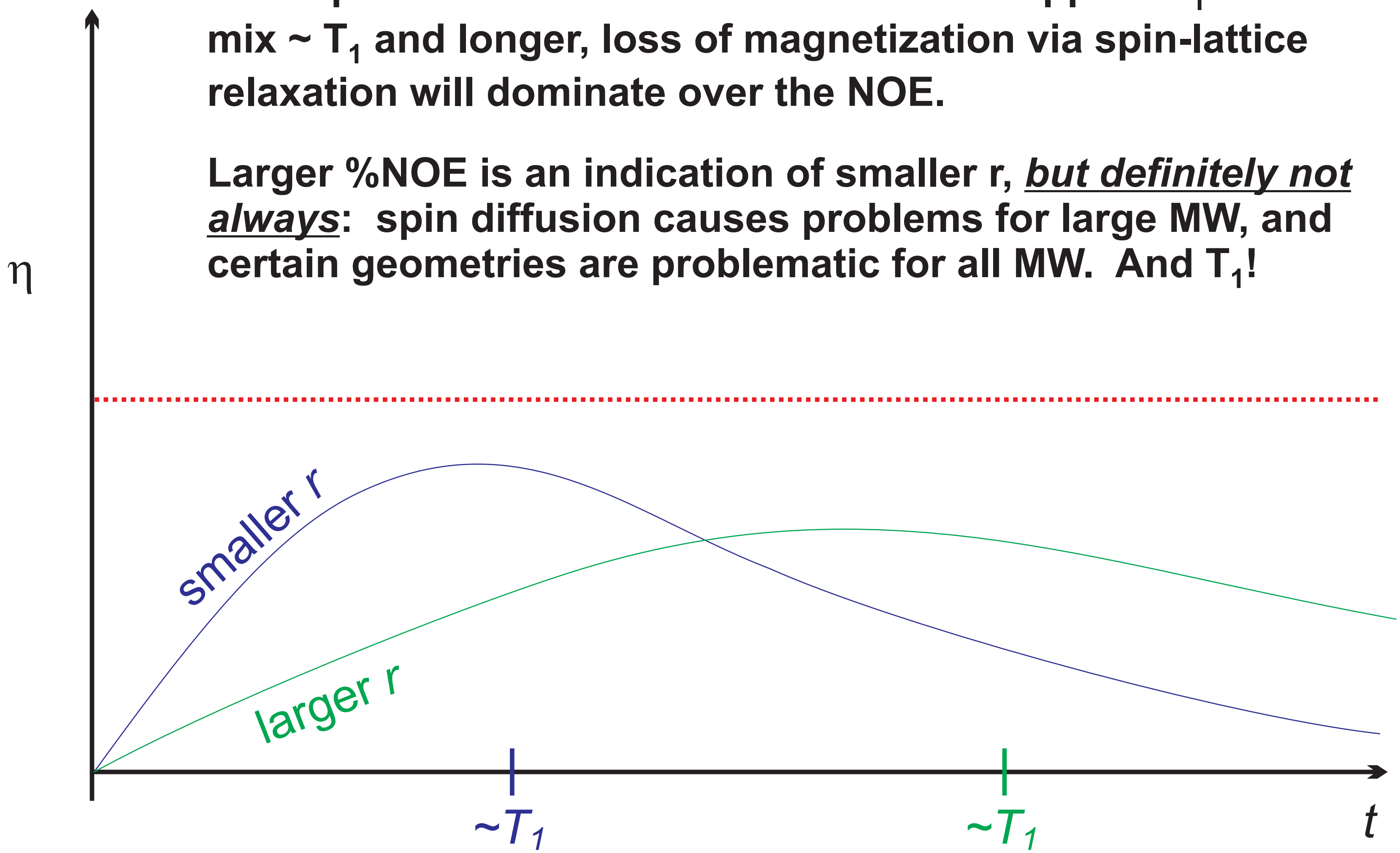
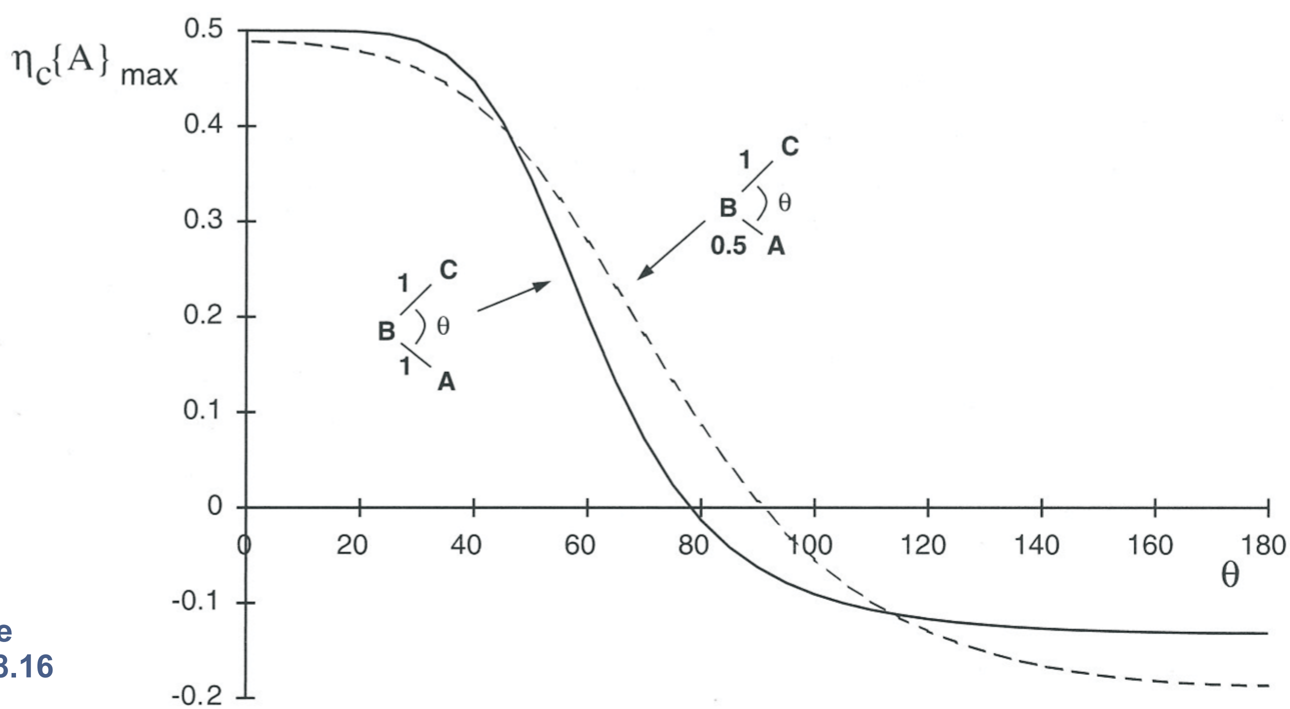
Larger %NOE is an indication of smaller r, ***<u>but definitely not always</u>***: spin diffusion causes problems for large MW, and certain geometries are problematic for all MW. And T_1!



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 $\boldsymbol{r_{IS}}$.
  - ⇨ It is important to be aware that *not* observing an NOE is weak evidence. ***Measure NOEs in all directions.***

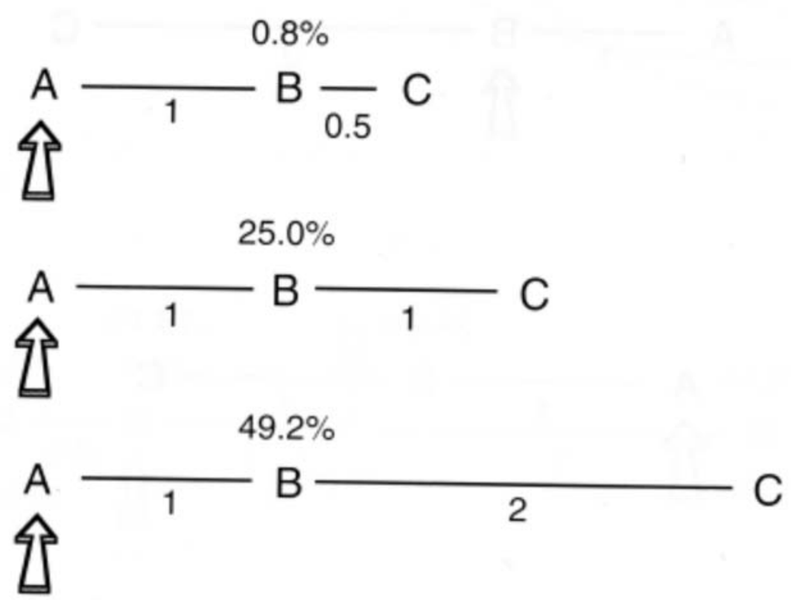


Claridge
Figure 8.16

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!!

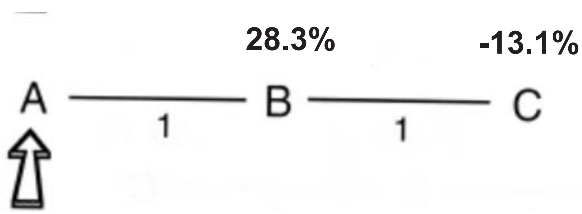


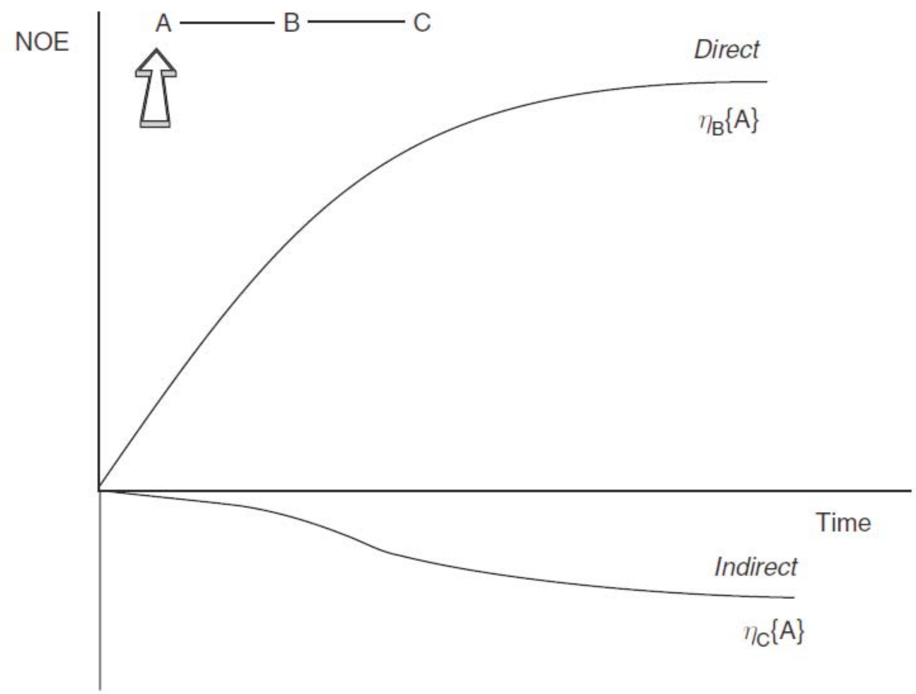
Claridge
Figure 8.12

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!!





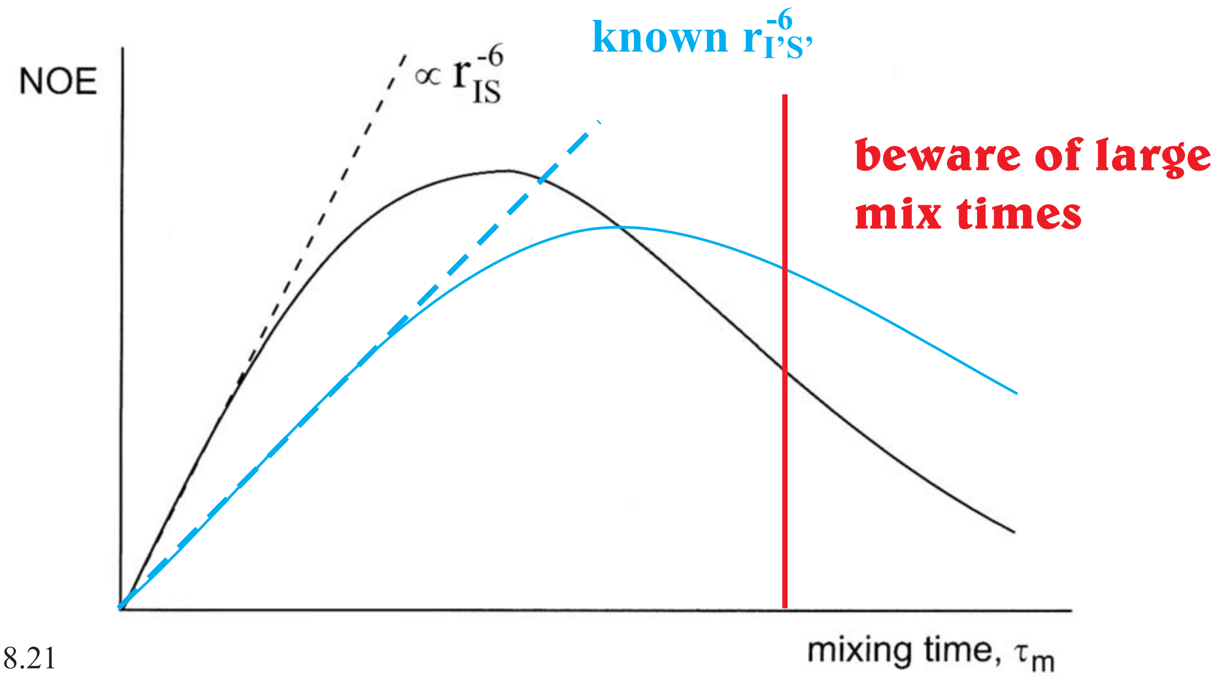
Claridge
Figs 8.14+8.15

Distance Information via NOE Measurements

- 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

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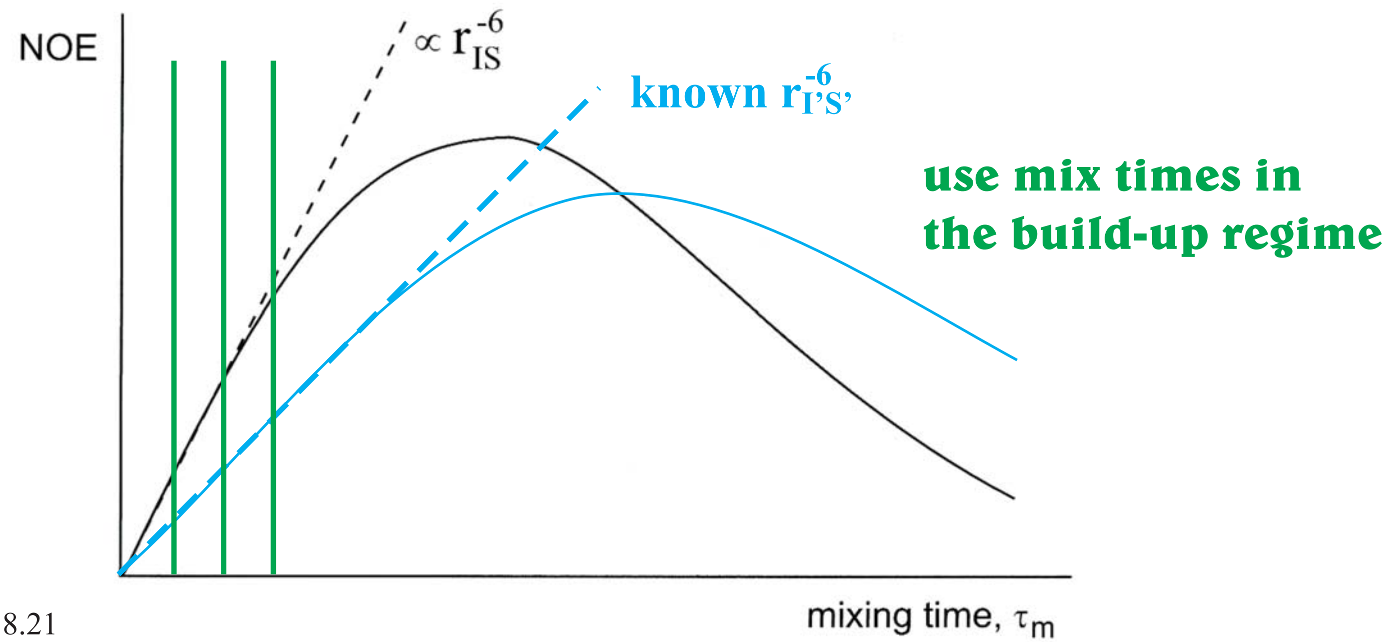


Claridge Fig. 8.21

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.

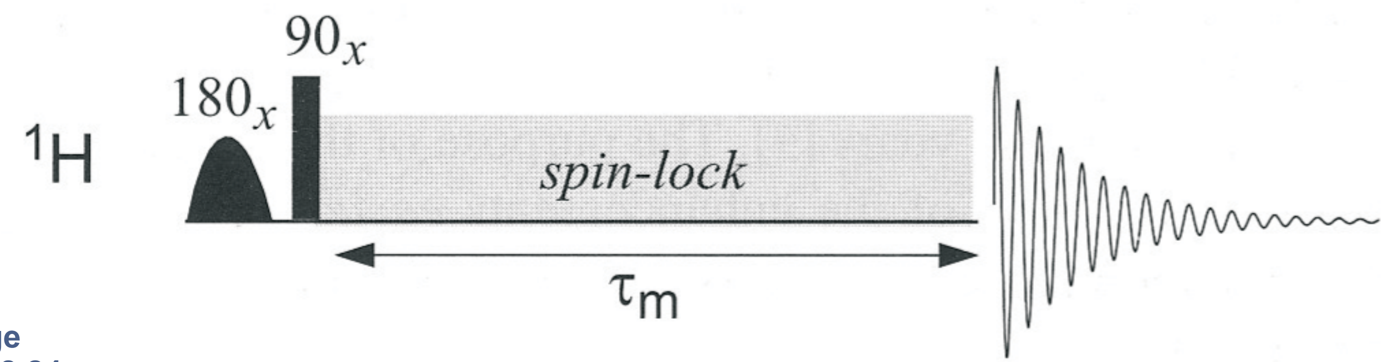
- NOESY-1D and NOESY-2D are *transient* experiments. These experiments have utility for **mix** $\leq$ $\mathbf{T_1}$**.**

- 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).

Review - TOCSY and Scalar Coupling

- ***Spin-locking*** the magnetization scales the chemical shift to near-zero (in Hz), producing strong coupling.

ROESY

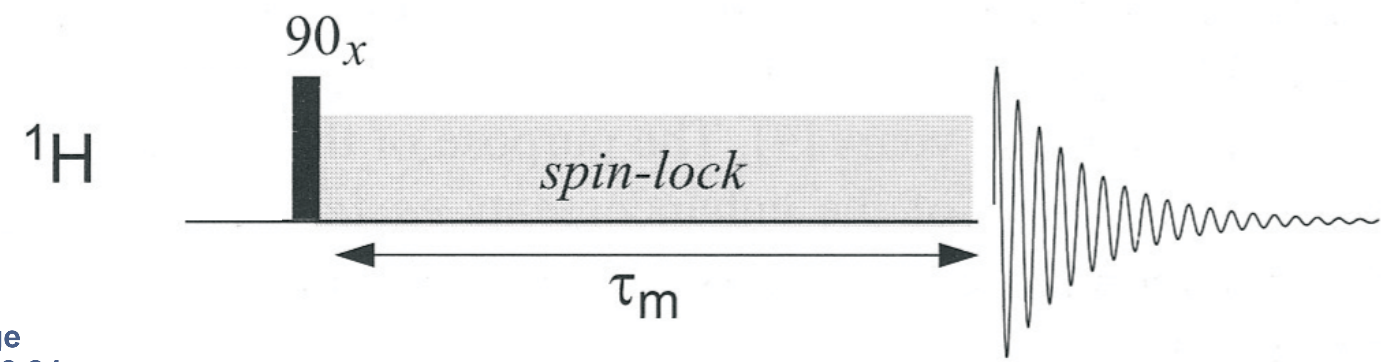


Claridge
Figure 8.24

Review - TOCSY and Scalar Coupling

- ***Spin-locking*** the magnetization scales the chemical shift to near-zero (in Hz), producing strong coupling.

TOCSY



Claridge
Figure 8.24

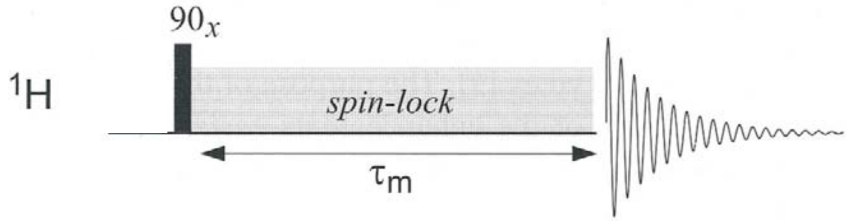
Review - TOCSY and Scalar Coupling

- ***Spin-locking*** the magnetization scales the chemical shift to near-zero (in Hz), producing strong coupling.

  Strongly coupled protons exchange magnetization once every ~1/2J.

Claridge Figure 8.24

TOCSY



Claridge 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).