1946: Harvard (*F*(*ω*)) *vs.* Stanford (*S*(*t*)) experiment

Continuous Wave Experiment

A "CW" NMR spectrum is obtained by sweeping the rf frequency or the magnetic field through resonance slowly. In this experiment rf is on continuously. However, B_1 is small, so that only one resonance is excited at a time.

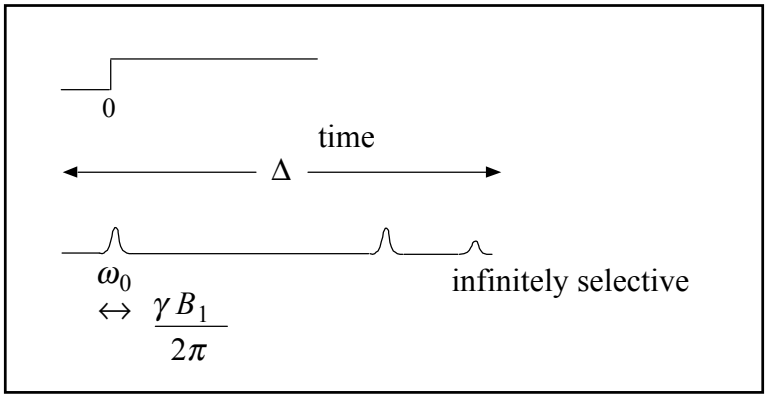


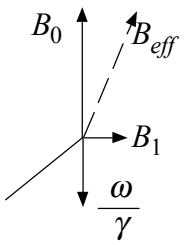
Figure 10-1

In a frame rotating with angular velocity ω, the "effective field" is given by

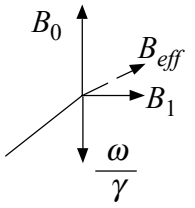
$$\vec{B}_{eff} = \left(B_0 + \frac{\omega}{\gamma} \right) \vec{k} + B_1 \vec{i}$$

Since $\left\| \vec{B}_0 \right\| >> \left\| \vec{B}_1 \right\|$, when ω is far from ω_0, $\vec{B}_{eff} \approx \vec{B}_0$ and M_z is unaffected, while when $\omega \to \omega_0$, $\vec{B}_{eff} \approx \vec{B}_1$ and absorption occurs.

Far from resonance $\vec{B}_{eff} = \left(B_0 + \frac{\omega}{\gamma} \right) \approx B_0$



At or near resonance $\vec{B}_{eff} = \left(B_0 + \frac{\omega}{\gamma} \right) + B_1 \approx B_1$



$F(\omega) \equiv$ NMR spectrum in frequency domain.

Fourier Transform NMR

Same information in $F(\omega)$ and $S(t)$.

In fact

$$F(\omega) = \frac{1}{\sqrt{2\pi}} \int_{-\infty}^{\infty} S(t) e^{-i\omega t} dt$$

$$S(t) = \left(\frac{1}{2\pi} \right)^{1/2} \int_{-\infty}^{\infty} F(\omega) e^{i\omega t} d\omega$$

Advantage of $S(t)$: it takes T_2 to acquire $T_2 \approx 20 - 100$ msec for small protons
T_1 per experiment $T_1 \approx 1$ sec

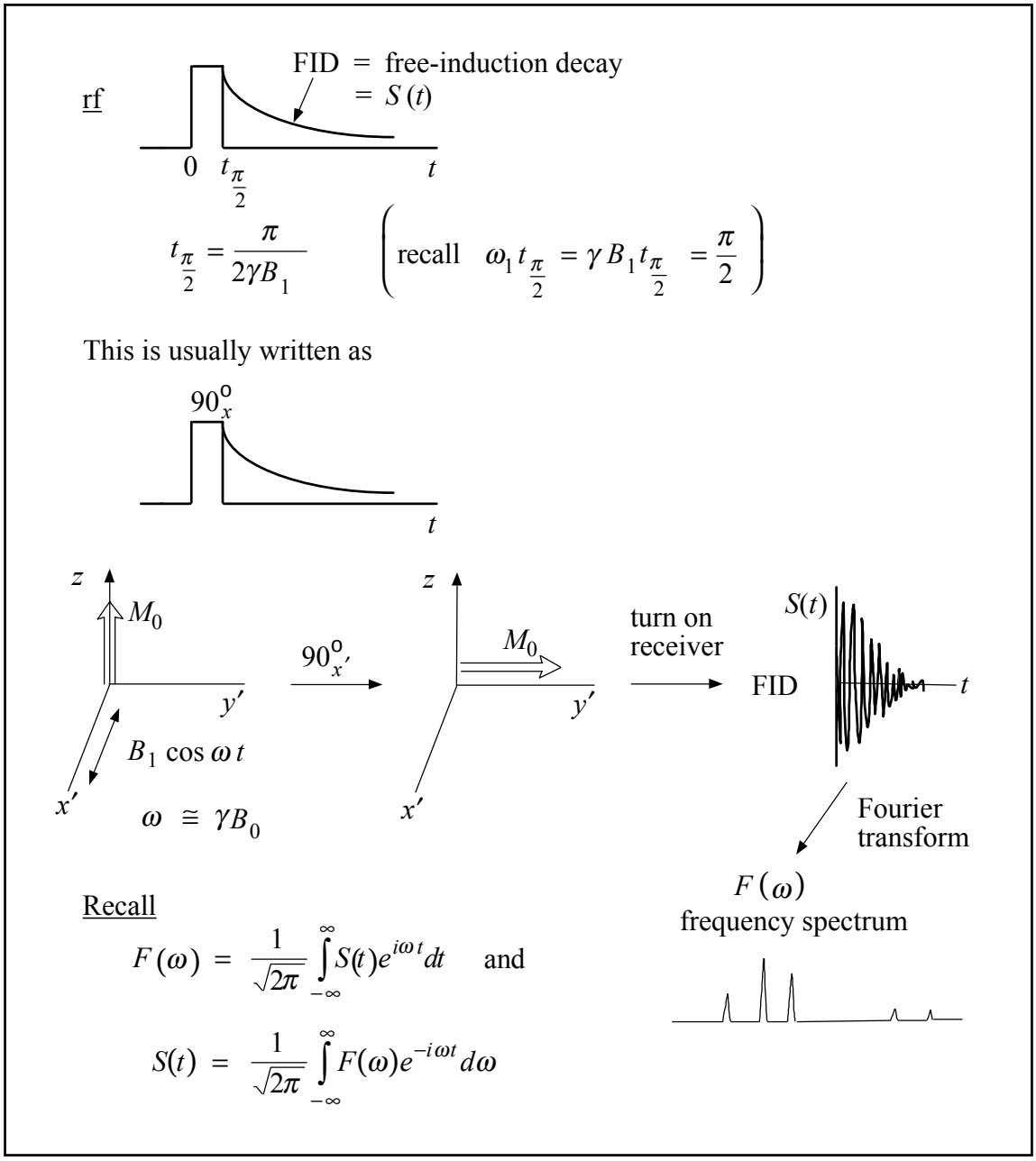
so in the time it takes to do a CW experiment, say 250 sec, you can do 250 Fourier transform experiments.

$\frac{S}{N}$ improvement $N^{1/2} = (250)^{1/2} = 16$

Another important difference is that in FT NMR experiments, all spins are affected simultaneously by rf, and all contribute to detected signal simultaneously.

FT NMR experiments are typically shown as timing diagrams or "pulse sequences", *e.g.* the simplest one-pulse experiment:

Single-pulse Experiment



$$t_{\frac{\pi}{2}} = \frac{\pi}{2\gamma B_1} \qquad \left(\text{recall} \quad \omega_1 t_{\frac{\pi}{2}} = \gamma B_1 t_{\frac{\pi}{2}} = \frac{\pi}{2} \right)$$

$$F(\omega) = \frac{1}{\sqrt{2\pi}} \int_{-\infty}^{\infty} S(t) e^{i\omega t} dt$$

$$S(t) = \frac{1}{\sqrt{2\pi}} \int_{-\infty}^{\infty} F(\omega) e^{-i\omega t} d\omega$$

Figure 10-2

Constraints on rf pulse

1) Must be << T_2

2) Must be short enough to evenly excite all resonances in spectrum. Pulse of length t contains these Fourier components:

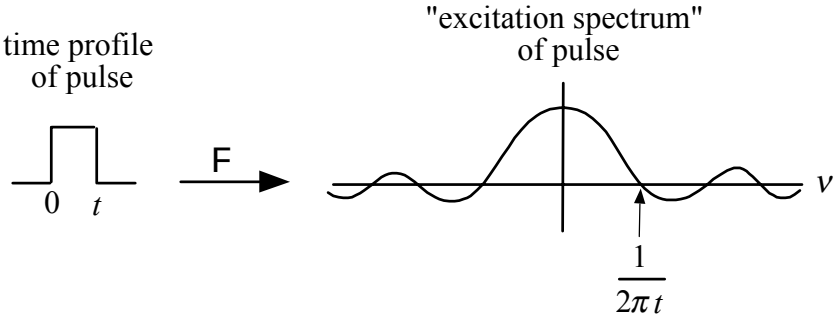


Figure 10-3

Recall for a $\frac{\pi}{2}$ pulse, $t = \frac{\pi}{2\gamma B_1}$. We require for uniform excitation that

$$\gamma B_1 >> 2\pi\Delta \quad \text{where} \quad \Delta \quad \text{is the spectral width}$$

$$\text{So} \quad t_{\frac{\pi}{2}} << \frac{\pi}{2\,(2\pi\Delta)} = \frac{1}{4\Delta}$$

Representative numbers:

$$t_{\frac{\pi}{2}} \approx 5\mu s \qquad \Delta \approx 5-10 \text{ kHz for } {}^{1}\text{H}$$

$$\Delta \approx 20 \text{ kHz for } {}^{13}\text{C}$$

Free-induction Decay

After $\frac{\pi}{2}$ pulse, $M_{xy} \neq 0$, nonequilibrium, so M_{xy} must decay with time. The rate of decay $(1/T_2)$ depends on how the individual spins that make up the magnetization can get out of phase, and this decay thus depends on

(a) different frequencies of precession of various spins that are excited by rf:

$$\Delta\omega_0 : \omega_1^0, \omega_2^0 \text{ K K}$$

(b) magnetic field inhomogeneity over the sample

(c) variations in the precession frequencies due to fluctuations in the local magnetic field at the nucleus: $\Delta\overline{(\omega - \omega_0)^2}^{1/2}$

In any case, free-induction decay (FID) ≡ NMR spectrum in the time domain. Long T_2's transform to sharp lines in the frequency domain; short T_2's yield broad lines.

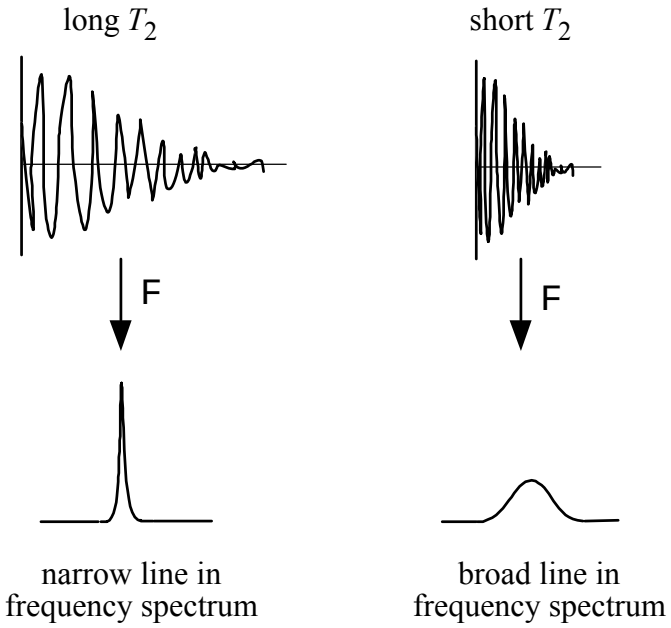


Figure 10-4

Spin Lattice Relaxation

After a single pulse $\neq 2n\pi$, M_z not at thermal equilibrium. Bloch postulated exponential recovery of M_z (justified for random isotropic rotational motion) with time constant T_1 … also called spin-lattice relaxation time (T_2 processes are adiabatic … spin-spin

magnetization transfer without loss of energy to surroundings *vs.* T_1 processes … spin system loses energy to surrounding "lattice" as relaxation proceeds). So from Bloch, after a single pulse

$$\frac{d}{dt}M_z = -\frac{(M_z - M_0)}{T_1}$$

General solution: $M_z(t) - M_0 = Ae^{-t/T_1}$. For a π pulse: $M_z(t) = (1 - 2e^{-t/T_1})M_0$

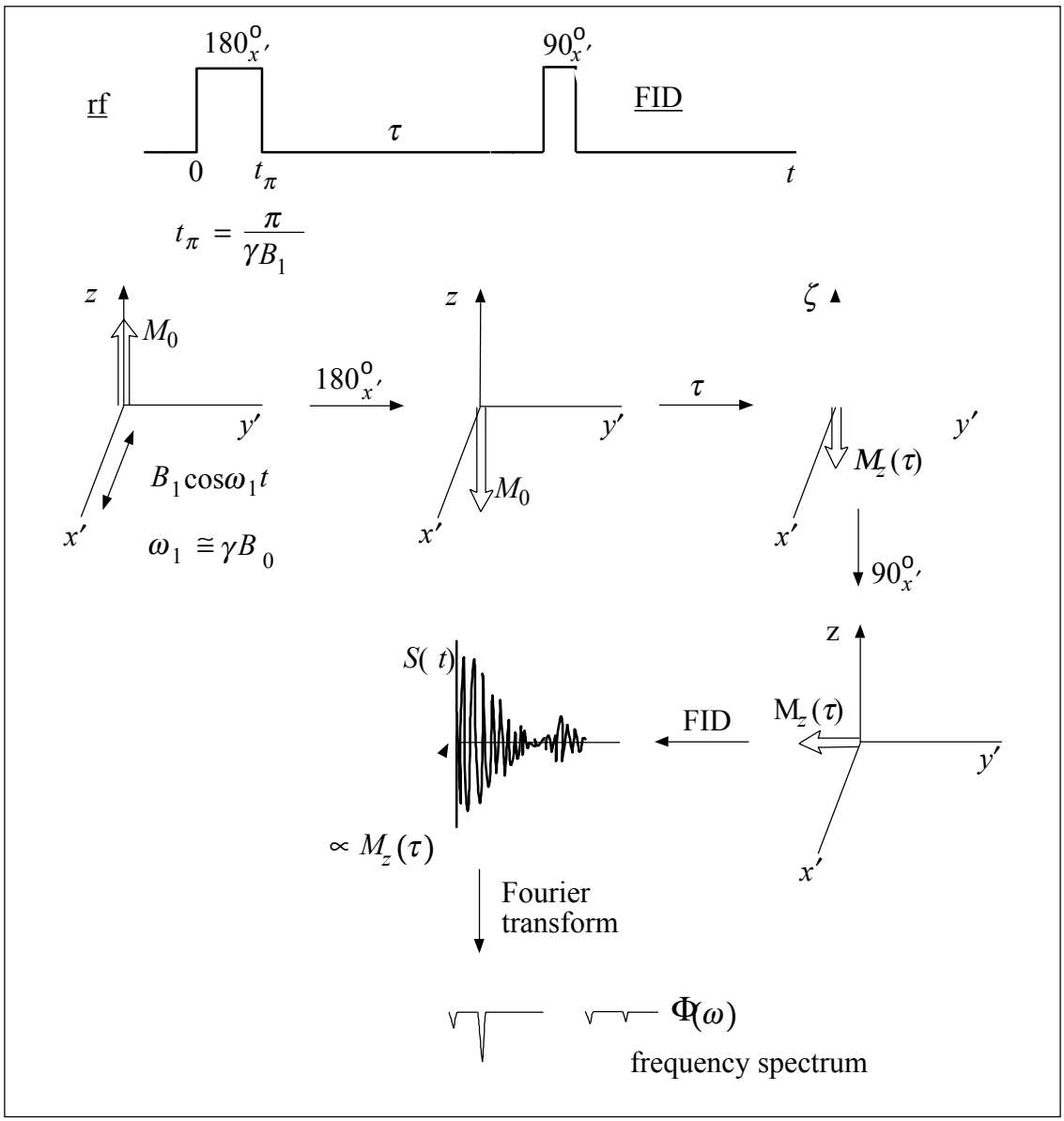


Figure 10-5

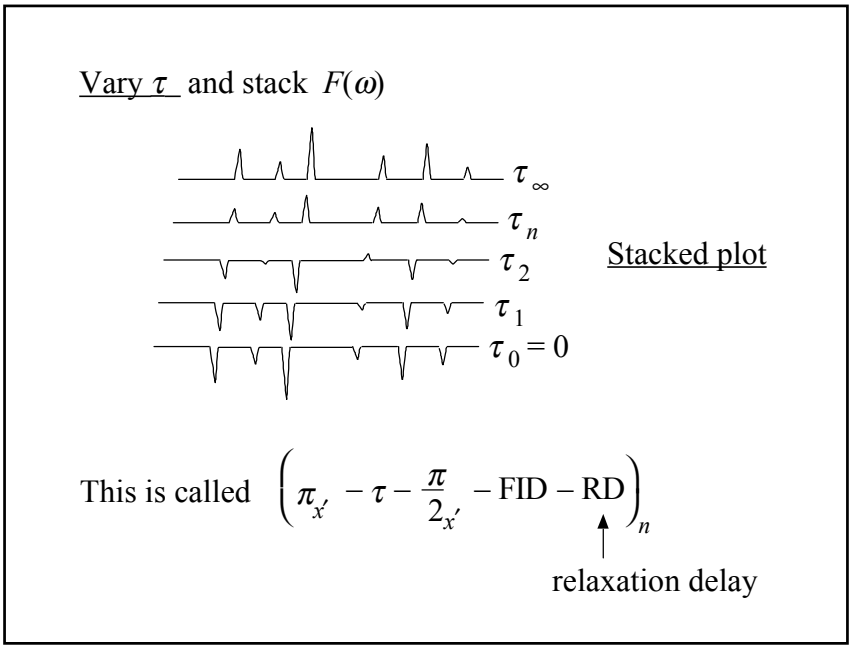


Figure 10-6

Note that different resonances have different T_1's! For each of the resonances, obtain a curve. Use nonlinear least squares analysis to extract T_1.

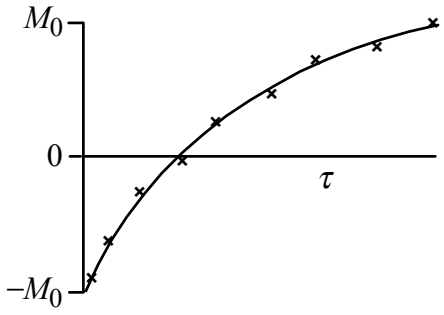
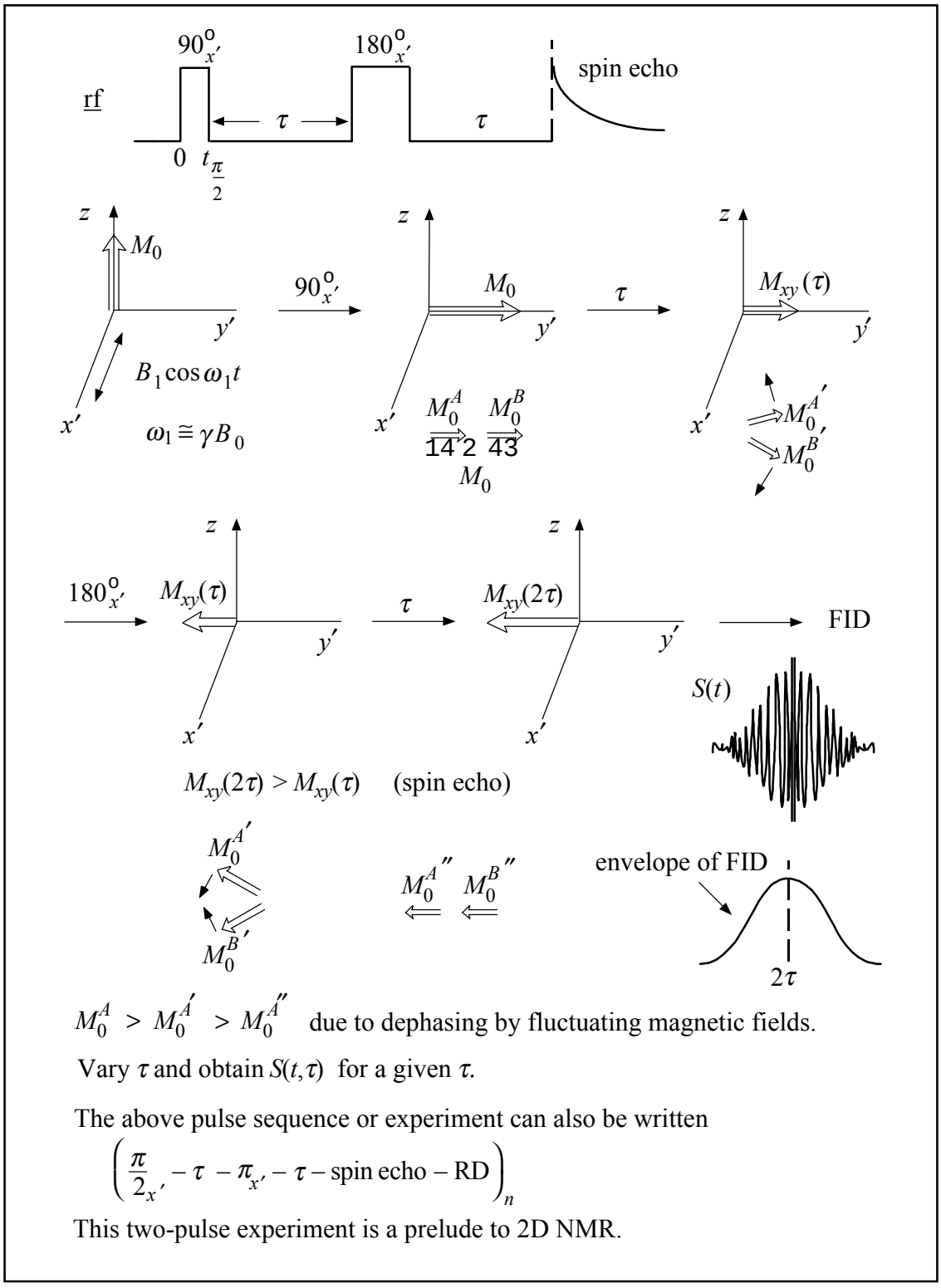


Figure 10-7

Another Two-pulse Experiment — "The Spin Echo"



$M_0^A > M_0^{A'} > M_0^{A''}$ due to dephasing by fluctuating magnetic fields.

Vary τ and obtain $S(t,\tau)$ for a given τ.

The above pulse sequence or experiment can also be written

$$\left(\frac{\pi}{2}_{x'} - \tau - \pi_{x'} - \tau - \text{spin echo} - \text{RD}\right)_n$$

This two-pulse experiment is a prelude to 2D NMR.

Figure 10-8

The spin echo is a component of <u>many</u> more complicated pulse sequences. By itself, it can be used to measure T_2 … by varying τ and looking at envelope intensity (or signal intensity in $F(\omega)$). Furthermore, Hahn showed an effect of diffusion on NMR spectra: in an inhomogeneous field, the spin echo amplitude diminishes according to:

$$M(2\tau) = M_0 \exp\left[\frac{-2\tau}{T_2}\right] \exp\left[-\gamma\left(\frac{\partial B}{\partial z}\right)^2 \frac{2}{3} D\tau^3\right] \quad \text{where } D = \text{diffusion constant}$$

Pulsed Field Gradients

Recently (past 5 years) great use has been made of short duration (*i.e.* "pulsed") field gradients. A linear gradient applied along the z axis modifies the Larmor frequency for different volume elements along the length of the sample tube:

$$\omega(z) = -\gamma\left[B_0 + B_g(z)\right]$$
$$= \omega_0 - \gamma z G \qquad \text{where } G \text{ is a constant} \propto \text{gradient strength}$$

If $M_{xy} \neq 0$ when the gradient is applied, the spins become "phase-encoded"… their phase $\theta(z)$ in the xy plane depends on their location along the z axis:

$$\phi(z) = \gamma z G t \qquad \text{where } t = \text{duration of gradient pulse}$$

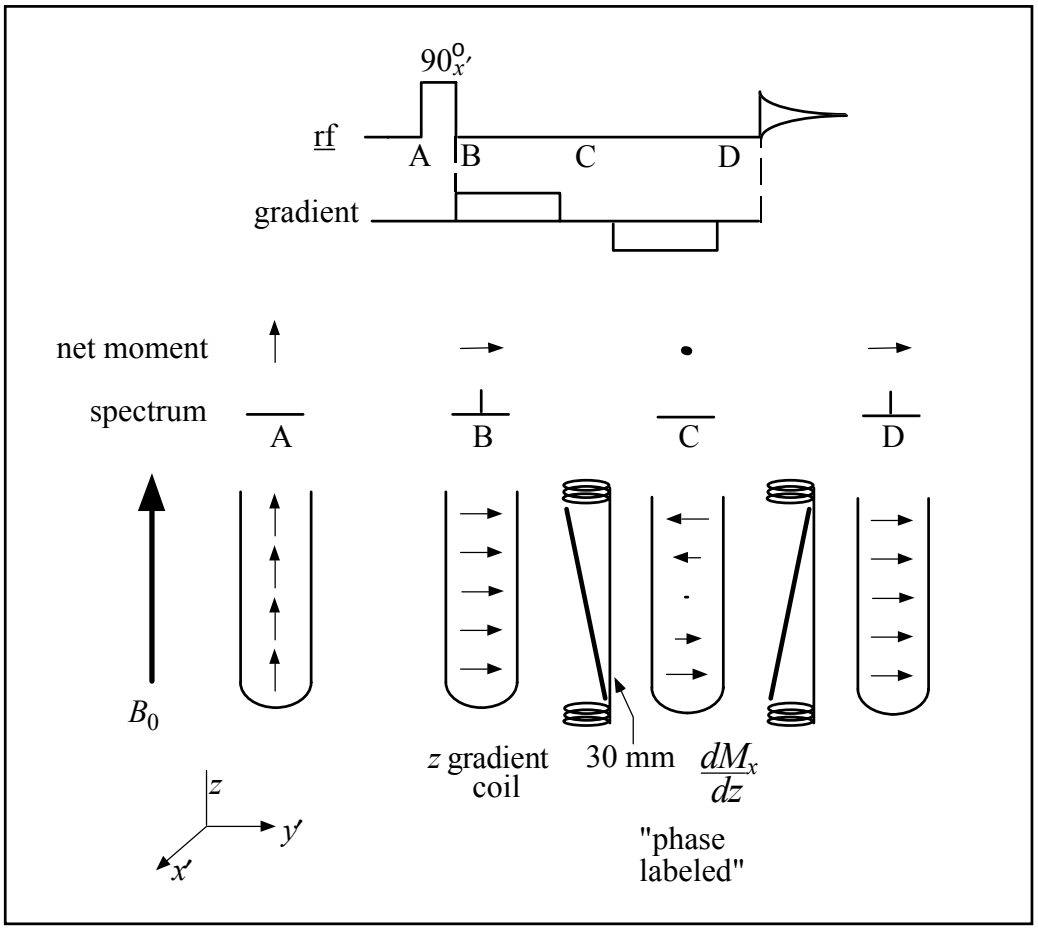


Figure 10-9

As an example, a pair of gradients can be combined with a Hahn spin echo to create a diffusion-selective (therefore molecular weight-selective) echo sequence.

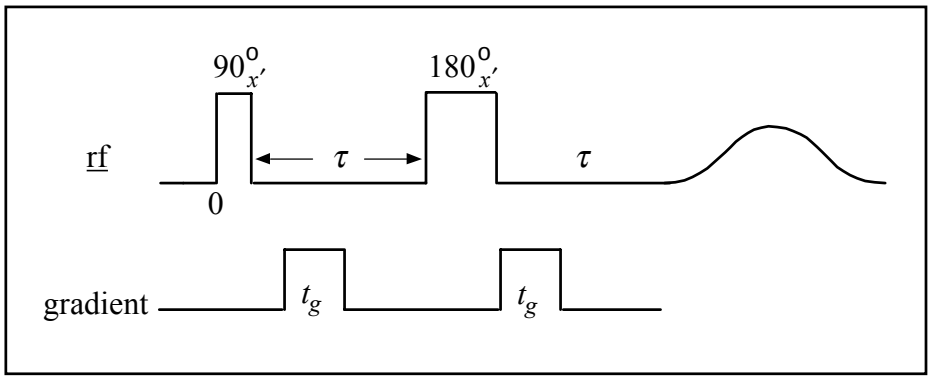


Figure 10-10

The initial 90*x* pulse generates transverse magnetization. The first gradient pulse phase-encodes the magnetization. The 180*x* rf pulse then inverts all spins … so the second gradient pulse should reverse the phase-encoding *unless* the molecules diffuse out of their original volume element during the first echo delay τ. This will diminish the signals of small molecules preferentially to those of big molecules (since the latter diffuse more slowly) … particularly, this provides a way of eliminating the solvent signal in aqueous solutions of biological macromolecules.