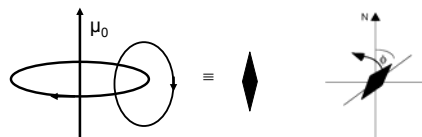


Nuclear Magnetic Resonance Spectroscopy

Some Magnetism

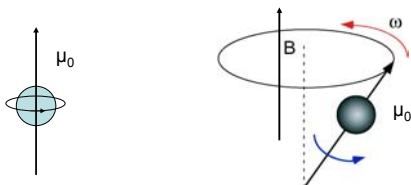
A charge traveling in a loop, is similar to a current that flows through a conducting loop. It generates a magnetic moment μ (dipole) perpendicular to the plane of the conducting loop.

Such dipoles in a magnetic field align with the field direction.



Most spinning nuclei behave like magnets.

However, as opposed to the behavior of a classical magnet the nuclear spin magnetic moment does not *always* align with the direction of the magnetic field B (quantization).



In the absence of a field the magnetic nuclei magnets orient in all possible directions.

Nuclear spin quantum number, I :

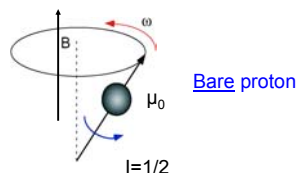
Depending on the nuclear constitution, they would possess a nuclear spin moment.

A Mass #	Z Atomic #	I (nuclear spin q.n.)
Odd	Odd or even	$n/2$ (odd integer)
Even	Even	0
Even	Odd	n (integer)

Any nucleus with an odd Z or an odd A has a net spin.

Nuclear angular momentum of ρ_0 $\rho_0 = \sqrt{I(I+1)}\hbar$

Precessional movement



Bare proton

magnet of magnetic moment (dipole) μ_0

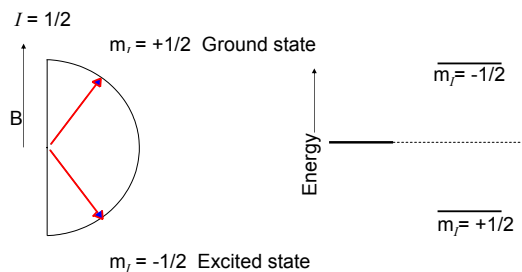
$$\mu_0 = \gamma \rho_0 = \gamma \sqrt{I(I+1)}\hbar$$

magnetogyric ratio of the nucleus

<http://www.chem.queensu.ca/FACILITIES/NMR/nmr/webcourse/index.htm>
<http://www.eclipse.net/~numare/nsinmrpt.htm>

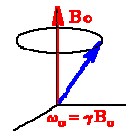
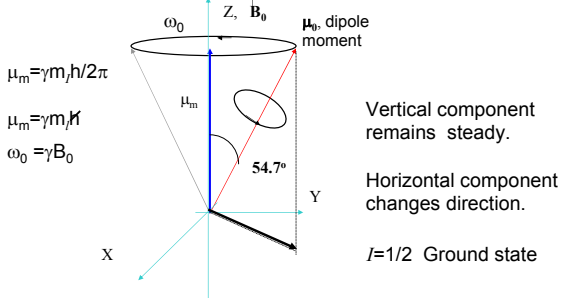
orientations (states) in a $B = 2I+1$

Each state characterized by $m_I = -I, -I+1, \dots, +I$



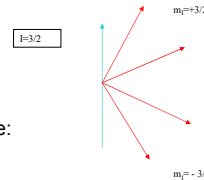
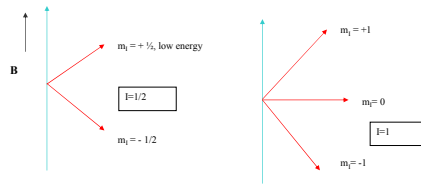
In the presence of a B the two states differentiates.

Precessional cone



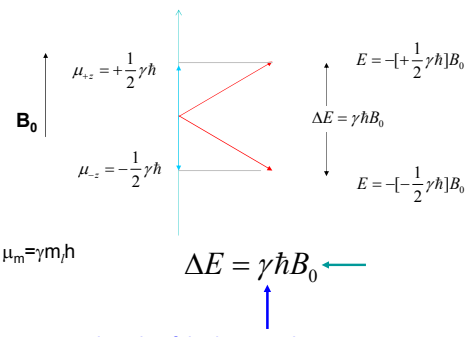
ω_0 = Larmor frequency, rad/s
 The quantity NMR experiment 'measures'.

<http://www.bruker-nmr.de/guide/eNMR/chem/NMRnuclei.html>

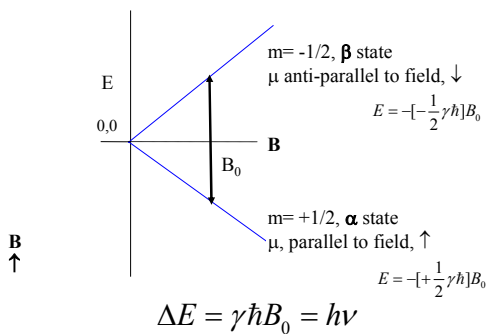


Selection rule:
 $\Delta m_l = \pm 1$

Transitions between states dictated by selection rules

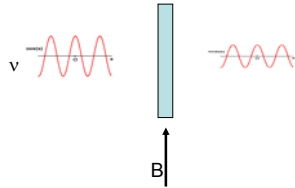


magnetogyric ratio of the bare nucleus

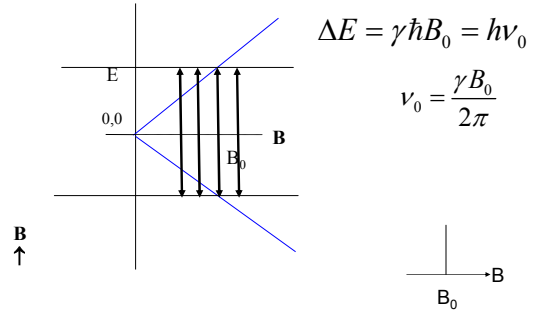


ν falls in the radio frequency region.

NMR Experiment: pseudo classical

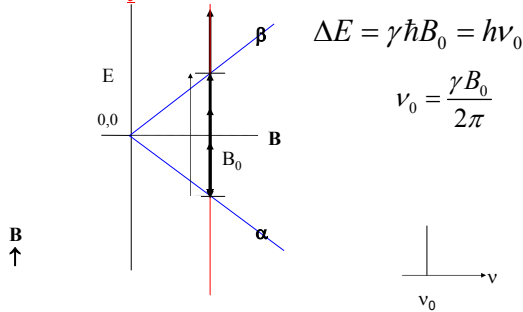


Continuous wave experiment: Field sweep
frequency constant



Continuous wave experiment: Frequency sweep

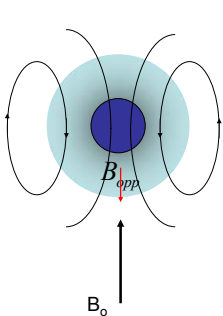
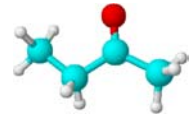
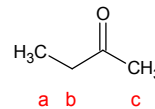
B constant at B₀



Protons in a sample are in a different electronic environment than a bare proton. Protons with different *types* of environments can be recognized by NMR.

Protons in a molecule are different from **bare protons**.

Each *type* of nucleus has the 'same environment'.



Screening constant

$$B_{opp} = \sigma B_0$$

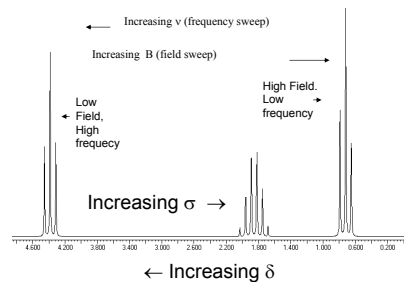
$$B_{eff} = B_0 - \sigma B_0 = (1 - \sigma) B_0$$

$$\nu = \frac{\gamma(1 - \sigma) B_0}{2\pi}$$

σ proportional to the electron density around the atom.

Therefore ν of absorption of each type of H is unique.

In the NMR experiment, the nuclear energy levels are excited and the "absorptions" of the nuclear excitations recorded.



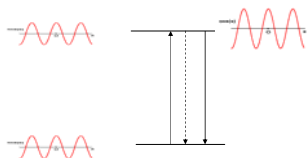
The 'pseudo' NMR experiment is impractical, i.e. absorbance cannot be measured experimentally.

The reason for it is a population distribution issue. And the nature of the interaction of electromagnetic radiation with matter (energy levels of matter).

Boltzmann Law $\frac{N_-}{N_+} \propto e^{-\frac{\Delta E}{kT}}$ ← very small

For protons $\gamma = 26.75 \times 10^7 \text{ rad/T s}$; $\hbar = 1.055 \times 10^{-34} \text{ Js}$,
 $k = 1.380658 \times 10^{-23} \text{ J/K}$. At 298K and $B_0 = 7.05\text{T}$

$$\frac{N_-}{N_+} = 0.99995 \approx 1 \Rightarrow N_- \cong N_+$$



Net Absorption of Radiation $\propto (N_+ - N_-) I_\nu$

When $N_+ \sim N_-$ the net absorption is very minimal, This small absorption is immeasurable.

The 'macroscopic' magnetic moment \underline{M} is extremely small and its field, in comparison to the applied field, \underline{B}_0 , is very small.

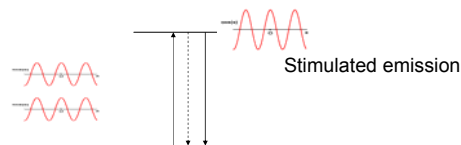
\underline{M} cannot be measured as is.

To measure \underline{M} it must be flipped to a direction orthogonal to \underline{B}_0 . \underline{M} is flipped on to the X-Y plane.

NMR experiment accomplishes the measurement of \underline{M} by flipping the macroscopic magnetic moment \underline{M} to the X-Y plane.

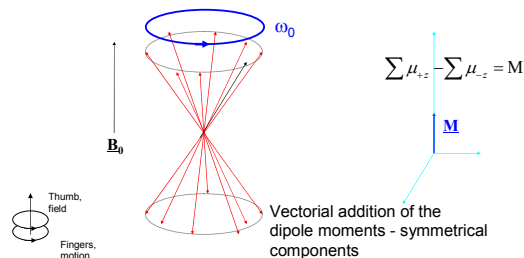
When nuclei are subjected to their resonant frequency at an intensity I_ν ; the transition $\alpha \rightarrow \beta$ (radiation absorption is proportional to $N_+ I_\nu$) occurs and is followed by two other processes.

- $\beta \rightarrow \alpha$ (**Relaxation**, is proportional to N_- , radiation-less part) * desirable as it increases N_+ population.
- Stimulated radiative emission is proportional to $N_- I_\nu$.



Orientation of nuclei in a **sample** (single proton type) in a \underline{B}_0 :

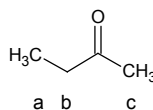
Two orientations possible ($I=1/2$) with a slight excess $\underline{\mu}_+ \parallel \underline{B}_0$ (Z). Horizontal components of dipole moments cancel leaving a small vertical component, net magnetic moment, \underline{M} .



Samples contain millions of molecules. All nuclei of the same *type* would precess at the same Larmor frequency.

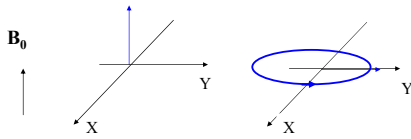
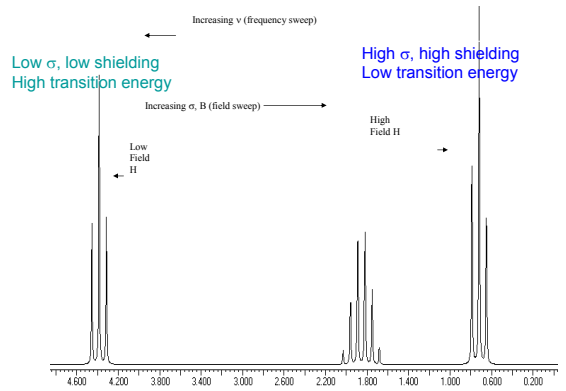
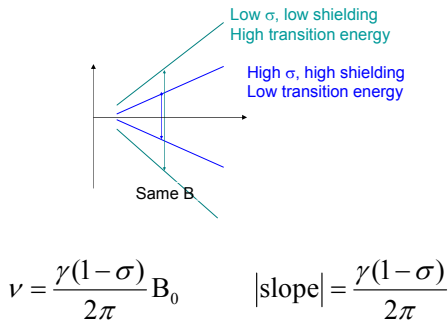
The sum total of the magnetic moment of one *type* of nuclei is a measure of the number of protons of that type in the molecule.

Thus the determination of the magnetic moment for protons in each type of electronic environment forms the essence of the NMR experiment.



$$M_a : M_b : M_c = 3 : 2 : 3$$

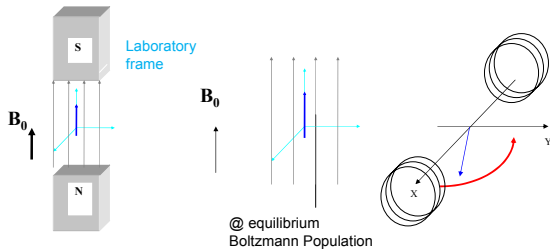
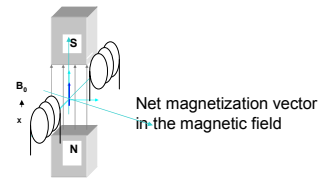
Relationship between energy levels,
magnetic field and shielding of protons in molecules:



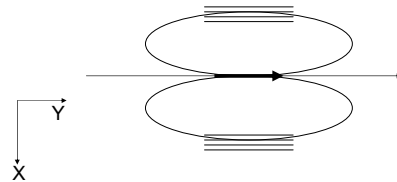
Once flipped on to the X-Y plane, \underline{M} does not remain static. \underline{M} would rotate around z axis at Larmor frequency. In order to measure \underline{M} a detection coil is used.

The rotating field of \underline{M} crossing the coil produce an AC voltage. of which, the amplitude is proportional to \underline{M} and frequency is equal to the Larmor frequency.

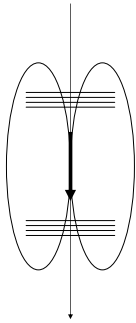
Experimental setup:



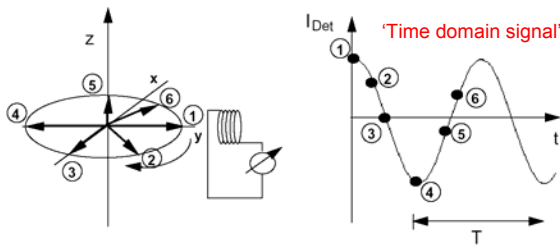
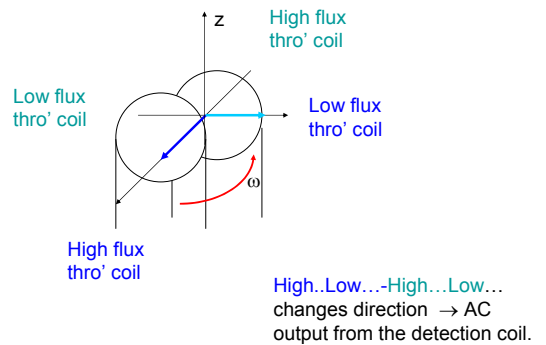
Net magnetization vector in the magnetic field.



Rate of change of magnetic flux through the coil lowest.



Rate of change of magnetic flux through the coil highest.



At the detector coil a time varying voltage signal created. The time variation is a sine/cosine curve.

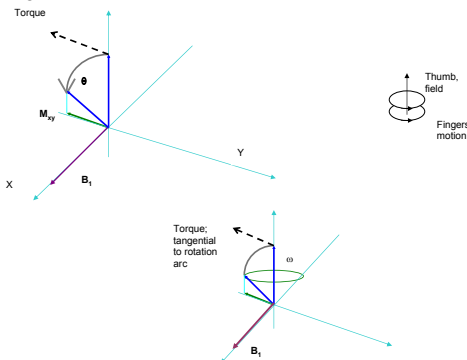
The amplitude of the 'time domain signal' is proportional to the magnetic moment; i.e. number of protons present in the molecule.

The frequency of the 'time domain signal' is the precessional frequency of the type of protons.

In FTNMR spectroscopy, determination of the amplitudes and the respective frequencies from the 'time domain signal' forms of different types of nuclei the crux of the technique.

Note: the quotation marks above.

Flipping the M on to X-Y plane.



To flip the **M** to the XY plane a magnetic a second field **B₁** is utilized.

Application of **B₁** normal to **M** would rotate **M** around **B₁** on a plane normal to **B₁**.

An application of a **B₁** in the 'fixed' X direction will force **M** instantly to take a position that is not normal to **B₁** because of **M**'s rotation about Z. That makes only a part of **M** (component on XY plane on Y direction) to experience **B₁**. Also the magnitude of this component will vary with the position of the rotating **M**.

This leads to a complex movement of the **M** vector in the laboratory frame (XYZ system) and will lead to a less meaningful experiment. Thus application of the second field **B₁** in a fixed direction like X, is fruitless.

To avoid such a complexity, the B_1 must be applied in such a way that it is *constantly normal* to (the rotating) M .

That is, B_1 must **change its direction** and must be **in phase** with M . B_1 must also **maintain a direction normal** to the rotating and 'tipping' M and must be **normal to B_0** .

To satisfy this condition B_1 must rotate on the XY plane at Larmor frequency and should stay in the XY plane. Under such a situation M will interact with B_1 because they are in unison – i.e. at resonance.

To accomplish the 'unison' B_1 must rotate in XYZ frame at Larmor frequency (**Rotating frame**); to generate an effective interaction with the rotating M in XYZ, at Larmor frequency.



Rotating Frame

<http://www.cis.rit.edu/htbooks/nmr/chap-3/c13-1.htm>

Movement of M in laboratory frame

<http://www.cis.rit.edu/htbooks/nmr/chap-3/c13-5.htm>

With such an application of B_1 ; M will rotate at an angular velocity of ω_1 around B_1 .

The angle by which M would move away from Z axis, $\theta = \omega_1 t_p = \gamma B_1 t_p$, where $t_p = \text{time of application of } B_1$.

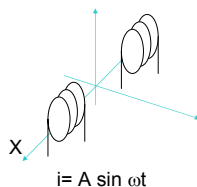
The rotation around B_1 creates a component of M on the XY plane.

It is this 'component' of M on the XY plane that is 'measured' in the NMR experiment.

The movement of M on to x-y plane is accomplished by applying B_1 for a certain time and the time t_p **required** is referred to as a **90° or $\pi/2$ pulse**.

The application of the magnetic field B_1 to M can be accomplished with a *linear oscillating magnetic field* of appropriate frequency ν (therefore angular velocity $\omega = 2\pi\nu$).

The oscillating magnetic field is applied to the sample so that the magnetic vector is oriented in the fixed (say X) direction.



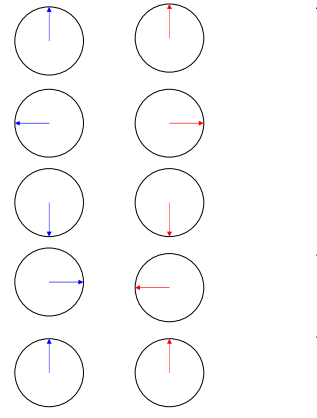
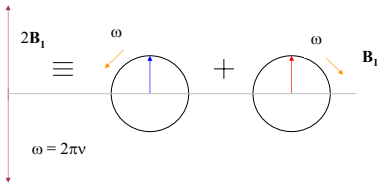
Transmitter coil

It is a **linearly oscillating magnetic field**. To get the amplitude of rotating field equal to B_1 , the amplitude of the linearly oscillating field must be $2B_1$ and the frequency of must be (ν) such that $2\pi\nu = \omega$.



This appears contrary to the explanation given so far, but it is not!

Explanation of rotating B_1 from 'linearly oscillating' field:



A linearly oscillating magnetic vector (or light for that matter) of amplitude $2B_1$ and angular velocity ω can be considered as the sum of right and left circularly polarized vectors; each with an amplitude of B_1 and angular velocity ω .

Only the B_1 with anti-clockwise motion is in unison with that of M , and therefore resonant, as it is *in phase* with the 'anti-clockwise' moving M . The other component is moving at 2ω in the clockwise direction in the rotating frame and therefore it is *off resonance* and will have no interaction with M .

The sample 'absorbs' energy causing the μ 's to flip, therefore M to turn, increasing θ .

In one mode of the CW experiment, the AC frequency can be changed continuously to bring each nuclei resonant with its Larmor frequency sequentially.

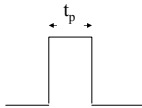
In the pulsed NMR (FTNMR) experiment all Larmor frequencies required for the nucleus, say H nuclei, ([the entire range of frequencies](#)) are provided to the sample at the same time.

Fourier Theorem

$$f(t) = \frac{a_0}{2} + \sum_{n=1}^{\infty} [a_n \cos(\omega_n t) + b_n \sin(\omega_n t)]$$

where $\omega_n = 2\pi\nu_n$

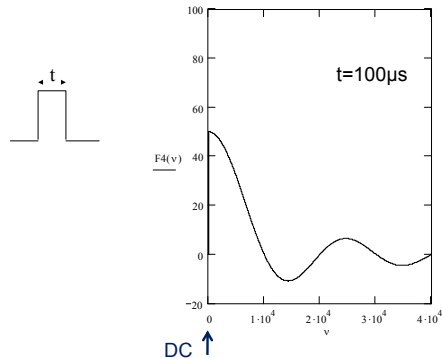
A DC pulse.



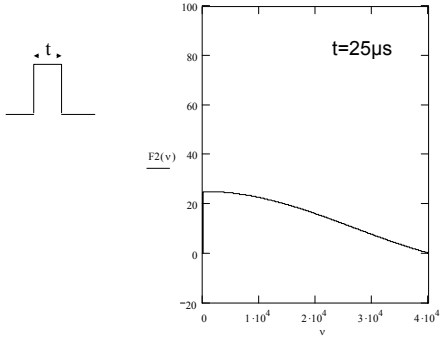
A *pulse* (square wave) is actually a *sum of sine curves* of a range of frequencies and a DC component. The [range of the values of the frequencies](#) that make up a pulse is dependent on the pulse duration.

The power (intensity) of each frequency that *makes up the pulse* is dependent on the pulse duration as well.

Range of frequencies generated from a pulse of duration t



Range of frequencies generated from a pulse of duration t



Linear and Nonlinear Mixing

Two waves with different frequencies can be mixed with an electronic device to produce either of a, b or c below;

- a. Sum wave of the inputs– *linear mixer* (sum/difference)
- b. Modulated wave with new frequencies derived from the input frequencies – *non-linear mixer* (multiplication)
- c. Some combination of 1 and 2. – *non-linear mixer*

A nonlinear mixing operation.

Consider the function $AB\cos(\omega_c t)$. Modulate the amplitude of the function by $[K + \cos(\omega_m t)]$, where K is a constant;

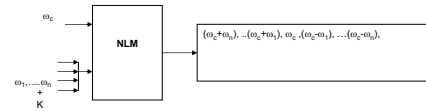
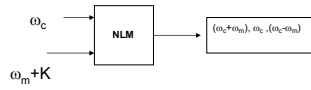
$$V = AB \cos(\omega_c t)[K + \cos(\omega_m t)]$$

$$V = ABK \cos(\omega_c t) + (AB/2)[\cos(\omega_c + \omega_m)t + \cos(\omega_c - \omega_m)t]$$

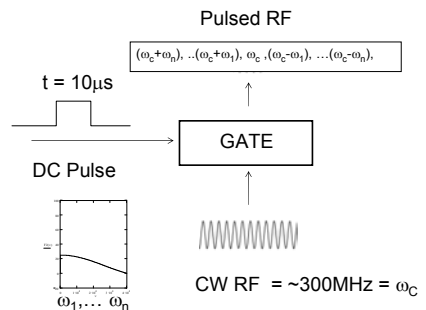
In case voltages are measured, K is a DC voltage.

Resultant is a sum of three cosine functions, of three different frequencies.

- $(\omega_c + \omega_m)$ - upper side band,
- ω_c - center band and
- $(\omega_c - \omega_m)$ - lower side band.



A x MHz frequency AC current allowed thro' a gate opened by a short pulse DC would contain a range of frequencies centered at x MHz.

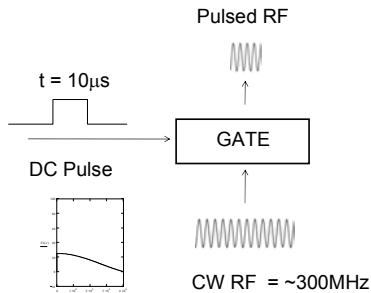


300MHz wave is modulated by the DC pulse.
i.e. 300MHz frequency is nonlinearly mixed with all the frequency components of the pulse.

Excitation of all nuclei is accomplished by nonlinearly mixing the frequencies contained in a DC pulse with an RF frequency that is close to the nuclear precessional frequencies.

The key feature in this pulsed excitation technique is that the sample is subjected to a range of frequencies, thus is capable of exciting nuclei of different *types* (environments) at the same time.

Having excited all the types of nuclei at the same time, that is moved the 'macroscopic' magnetic moments to the XY plane at the same time, the 'time domain signal' from each rotating moment could be observed.



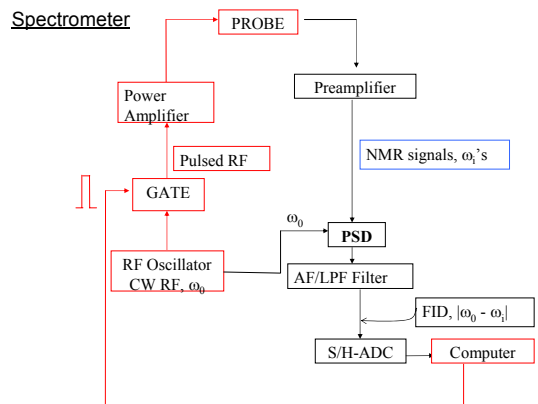
300MHz wave is modulated by the DC pulse.
i.e. 300MHz frequency is nonlinearly mixed with all the frequency components of the pulse.

In FTNMR vocabulary a x MHz instrument would mean an instrument where the applied field B_0 is such that the *bare protons* would rotate at a Larmor frequency of x MHz.

In such an instrument, the AC frequency of the pulse for excitation of bare protons is x MHz.

MSUM - 300MHz instrument; 7.05T magnet.

The pulse generates a range of frequencies centered at 300MHz.

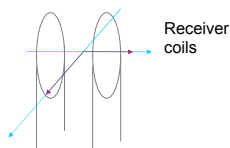


Detection of the Response from Excited Nuclei:

The rotating \underline{M} is associated with its own magnetic field.

To detect \underline{M} (actually the \underline{M}_{xy} component of \underline{M}), a receiver coil could be placed on the XY plane. The output of which will be the 'time domain signal'. Time domain signal is characterized by the amplitude of \underline{M} and the frequency (of precession of the nucleus).

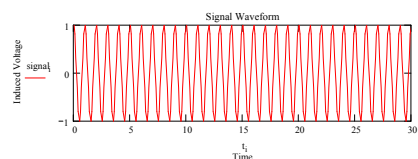
RF Receiver Coil



The lines of forces from such a rotating 'magnet' \underline{M}_{xy} was made to cross a (receiver) coil, it would **induce** an AC voltage (generator) on the receiver.

Receiver coil outputs an AC voltage of frequency ω . ($A_r \cos(\omega t + \phi_r)$, in general) the *amplitude of receiver output* is proportional to the *instantaneous \underline{M}_{xy}* value.

This is the signal that indicates absorption of energy by nuclei.



In FTNMR spectroscopy, determination of the amplitudes and the respective frequencies from the 'time domain signal' forms the basis of the technique.

The mathematical technique of transforming the 'time domain signal', $v(t)$, to frequency domain signal (spectra) is the Fourier Transformation.

According to **Fourier theorem** (any function) time domain signal, here, is a sum of sine and/or cosine waves.

$$f(t) = \frac{a_0}{2} + \sum_{n=1}^{\infty} [a_n \cos(\omega_n t) + b_n \sin(\omega_n t)]$$

Thus determining the frequencies that a time domain signal contains amount to determining the regular spectrum.

Using the Euler's identity;

$$e^{ix} = \cos(x) + i \sin(x)$$

The Fourier transform can be written as,

$$F(\omega) = \int_{-\infty}^{\infty} f(t)[\cos(\omega t) - i \sin(\omega t)] dt$$

Fourier transform essentially 'probes' the $f(t)$ for the presence of a waveform of frequency ω . The magnitude of the integral is proportional to the amplitude of the contributing wave of frequency ω .

The frequency of the time domain signal has a frequency in the range of MHz.

Digitization of frequencies in the MHz range is impossible as of now.

Fourier Transform:

The mathematical transform that converts the time domain signal to frequency domain signal (spectrum).

$$F(\omega) = \int_{-\infty}^{\infty} f(t)e^{-i\omega t} dt$$

Note that $F(\omega)$ is a number. It is the amplitude of the co-sinusoidal of frequency ω to the $f(t)$. One can view the multiplication of $f(t)$ by $e^{-i\omega t}$ as an 'interrogation of $f(t)$ for the presence of frequency ω '.

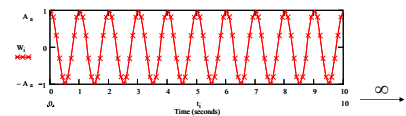
Thus a signal once digitized and stored in the memory can be analyzed for its constituent frequencies using the Fourier transform, viz.

The *digitized data* $f(t)$ when subjected to an *FT* is called a *discrete Fourier Transform*. The high-speed implementation of the Fourier transform is called the FFT.

Plot of amplitude $F(\omega)$ vs ω is the spectrum, in principle.

Some aspects of the time domain signal:

The time domain signal requires digitization, to be subjected to FFT. This by implication demands that the signal be sampled at frequency in the MHz range and higher than the signal frequency.



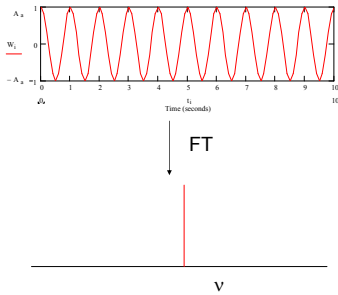
$$f(t) = A \cos(\omega t)$$

e.g. $v=1$ Hz

Sample rate=8 Hz

Dwell Time = DW = 1/8 s

FT of an **infinite** signal of a single frequency would give a sharp peak, FWHM = 0.



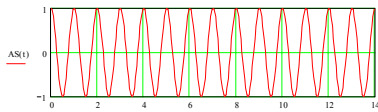
Signal sampling creates discrete points from an analog (signal in time space) output from the probe.

If the sampling rate slower than the rate at which the signal varies the FT would recognize the signal as a signal of frequency lower than the true, actual frequency of the signal.

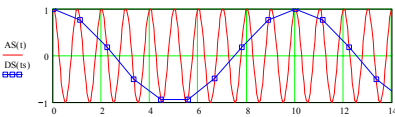
Thus there exists a required minimum-sampling rate, so that the FT would recognize the correct frequency of the signal. The **Nyquist Sampling Theorem** dictates this sampling rate; it states that the sampling frequency (rate of sampling) must be at least twice the frequency of the signal of interest.

There exists no data acquisition system that can acquire data at a frequency in the MHz range.

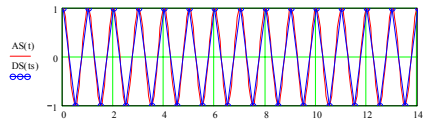
Example coil output.



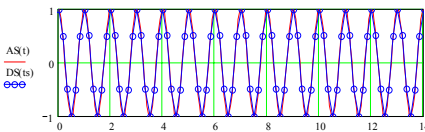
Frequency = 1.0 Hz **Min sampling rate = 2 * (1.0Hz)**



Sampling rate = 0.9 Hz < 2*frequency leads to aliasing.

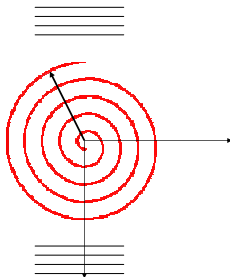


Sampling rate = 2.0 Hz

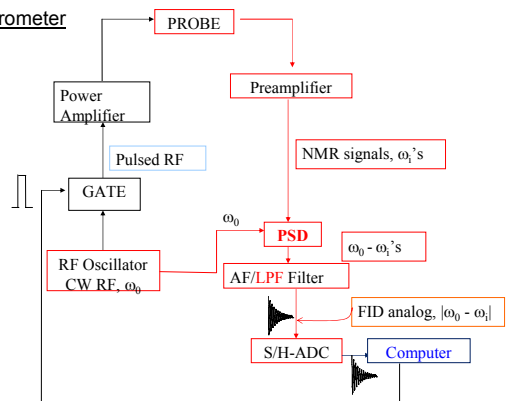


Sampling rate = 6.0 Hz

Decaying M_{xy} with time; vectors traces a decaying spiral

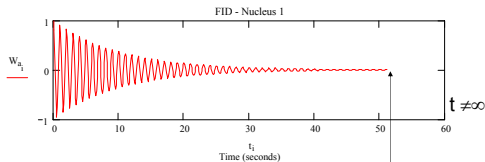


Spectrometer



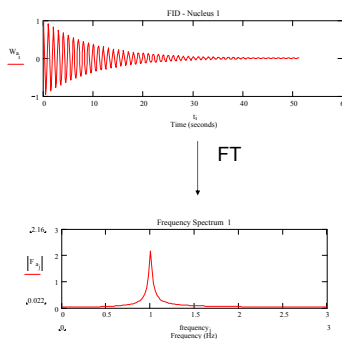
The time domain signal, arising from the excited nuclei, start to decay over a short span of time due to relaxation of the excited state. Free induction decay, FID of time constant, τ .

No signal is monitored up to infinity. Further, collecting data beyond the signal size that is smaller than that of the noise is not advantageous.



$$f(t) = A \cos(\omega t) e^{-t/\tau}$$

FT of a finite time signal would give a line with a non zero FWHM.



FIDs contain the same information as CW spectra.

However FID's can be acquired within seconds as opposed to sequential CW spectra.

FID's can be added as if they were spectra.

Many (2ⁿ) FID's can be added and the added FID is FT'd to produce a spectrum with improved S/N ratio.

S/N per FID = S/N per spectrum in CW experiment.

CW spectra cannot be added, B changes over time significantly.

Relaxation Mechanisms:

Longitudinal/Spin-Lattice Relaxation.

The process by which bulk magnetization **M** is re-established after excitation of nuclei.

The equilibration characterized by a rate constant, ($k=1/T_1$). This process also essential for signal averaging in NMR experiment.

Transverse/Spin-spin Relaxation.

M rotating in xy plane generates an induced current. This signal decays with time mainly due to loss of coherence.

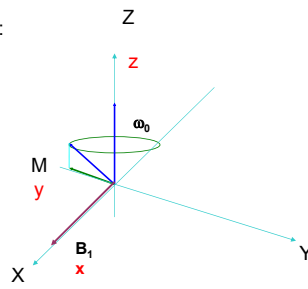
This process does not involve a change in the population of the spin states. This process is characterized by the rate constant ($k=1/T_2$)

Transceiver

In the NMR experiment once the excitation is effected the transmitter coil is turned off, gathering of the FID is initiated with the same coil, located in the probe.

Thus physically the same coil does the transmission and receiving. Hence it is termed the transceiver.

Rotating frame xyz:



Consider a mutually perpendicular axis system xyz defined by vectors $B_1(x)$ on x axis; z colinear with Z and y normal to x and z. **M** will be on y-z plane.

Relative to the frame XYZ, the stationary laboratory frame, xyz frame will be moving at an angular velocity ω_0 .

In the rotating xyz frame, the magnetic moment M is stationary because the frame is rotating at the same angular velocity as M .

Consider a rotating frame rotating at ω_0 and M rotating at ω , Assume $\omega_0 < \omega$. In such a case, M will be moving at $|\omega_0 - \omega|$ in the anti-clockwise direction in the rotating frame.

Consider a rotating frame rotating at ω_0 and M rotating at ω , Assume $\omega_0 > \omega$. In such a case, M will be moving at $|\omega_0 - \omega|$ in the clockwise direction in the rotating frame.

In the rotating frame M will be rotating with an angular velocity equal to the difference $|\omega_0 - \omega|$ of angular velocities.

Assume the RF oscillator frequency is ω_0 , and the Larmor frequency of a 'proton' ω .

Then, in the rotating frame, M from the 'protons' will be rotating at $|\omega_0 - \omega|$.

In NMR the difference between the RF oscillator frequency of the instrument and the Larmor frequencies of the nuclei is relatively low.

Signals in the low frequency region can be digitized as opposed to MHz range!!!

So to digitize one need to 'observe' the FID in the rotating frame, which is a mathematical/conceptual 'construct'.

Linear and Nonlinear Mixing

Two waves with different frequencies can be mixed with an electronic device to produce,

1. Sum wave of (input) frequencies – linear mixer (sum/difference)
2. Modulated wave with new frequencies derived from the input frequencies – non-linear mixer (multiplication)
3. Some combination of 1 and 2. – non-linear mixer

Consider the function $AB\cos(\omega_c t)$. Modulate the amplitude of the function by $[K + \cos(\omega_m t)]$, where K is a constant;

$$V = AB \cos(\omega_c t)[K + \cos(\omega_m t)]$$

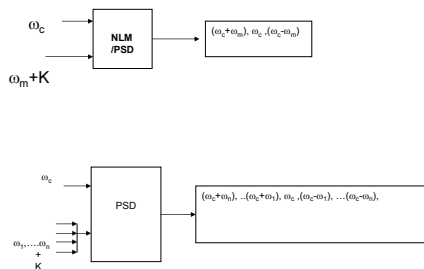
$$V = ABK \cos(\omega_c t) + (AB/2)[\cos(\omega_c + \omega_m)t + \cos(\omega_c - \omega_m)t]$$

In case voltages are measured, K is a DC voltage.

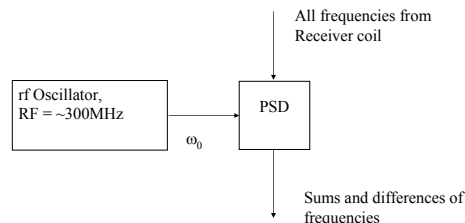
Resultant is a sum of three cosine functions, of three different frequencies.

- $(\omega_c + \omega_m)$ - upper side band,
- ω_c - center band and
- $(\omega_c - \omega_m)$ - lower side band.

This is a nonlinear mixing operation.



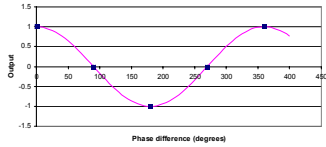
Phase Sensitive Detector



Phase sensitive detector input/output characteristics:

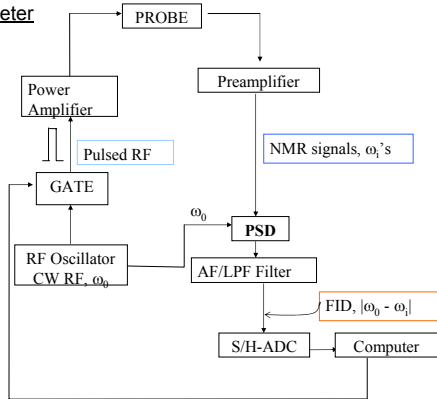
The output of the PSD depends on the phase difference, (among other factors) of the two input signal frequencies.

Phase difference	Output
0	DC
90	0
180	-DC
240	0
360	DC
$\Delta\phi$	$\cos \Delta\phi$



Frequency difference	Phase difference	Output
$\Delta\omega=0$	$\Delta\phi=\text{constant}$	DC
$\Delta\omega\neq 0$	$\Delta\phi$ varying at $(\omega_1 - \omega_2)$ with time	DC varying at $(\omega_1 - \omega_2)$ with time; AC of frequency $(\omega_1 - \omega_2)$

Spectrometer



The output of the PSD can be considered as coming from the Larmor frequencies in the rotating frame, rotating frame rotates in the laboratory frame at an angular velocity of ω_0 .

	¹ H	² H	¹³ C	¹⁴ N	¹⁵ N
Nuclear Spin, I	1/2	1	1/2	1	1/2
Nuclear magnetic moment, μ	+2.79284	+0.85743	+0.70241	0.40376	-0.28319
Gyromagnetic Ratio (rad T ⁻¹ s ⁻¹)	26.7510 x 10 ⁷	4.1064 x 10 ⁷	6.7263 x 10 ⁷	1.9331 x 10 ⁷	-2.7116 x 10 ⁷
Quadrupole moment (m ²)		2.73 x 10 ⁻³¹		1.6 x 10 ⁻³⁰	
Relative Sensitivity	1.00	9.65 x 10 ⁻³	1.59 x 10 ⁻²	1.01 x 10 ⁻³	1.04 x 10 ⁻³
Natural Abundance, %	99.985	0.015	1.10	99.63	0.37
Absolute Sensitivity	5680	8.2 x 10 ⁻⁶	1.00	5.69	0.0219

$$\nu = \frac{\gamma(1-\sigma)}{2\pi} B_0$$

For bare protons $\sigma = 0$. $\gamma = 26.75 \times 10^7 \text{ rad T}^{-1} \text{ s}^{-1}$ and $B_0 = 7.05\text{T}$ the Larmor frequency is;

$$\nu = \frac{\gamma(1-\sigma)B_0}{2\pi}$$

$$\nu := \gamma \cdot (1-\sigma) \cdot \frac{B_0}{2\pi}$$

$$\nu = 3 \times 10^8 \text{ Hz}$$

For bare $^1\text{H}(\sigma=0)$ at 2T field resonate at 90.0000 MHz;

A typical proton would have a frequency of 89.9995 MHz, it is an cumbersome number to report, further if a different instrument (different field) is used the frequency will be different.

To alleviate such problems absorption positions are reported with respect to a reference.

$$\omega_{\text{ref}} = \gamma_{\text{H}}(1 - \sigma_{\text{ref}})B_0$$

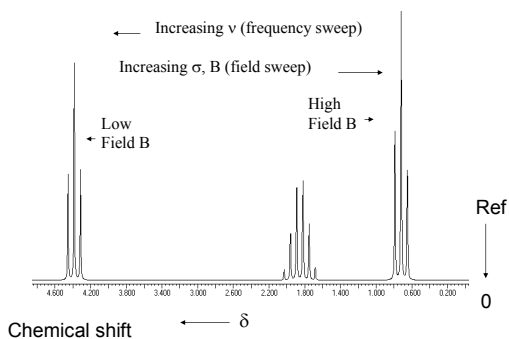
$$\omega_{\text{sample}} = \gamma_{\text{H}}(1 - \sigma_{\text{sample}})B_0$$

$$\omega_{\text{sample}} - \omega_{\text{ref}} = \gamma_{\text{H}}(\sigma_{\text{ref}} - \sigma_{\text{sample}})B_0$$

very small

$$\nu_{\text{sample}} - \nu_{\text{ref}} = \gamma_{\text{H}}(\sigma_{\text{ref}} - \sigma_{\text{sample}})B_0/2\pi$$

General presentation of NMR spectra:



The position of NMR resonance signals is dependent on the external magnetic field strength, B_0 (as opposed to UV, IR, AA etc).

$$\omega_{\text{sample}} = \gamma_{\text{H}}(1 - \sigma_{\text{sample}})B_0$$

Since no two magnets will have exactly the same field, resonance frequencies will vary accordingly. A method for specifying uniquely the position of NMR signals is needed.

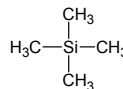
To express the position of NMR signals unequivocally, the signals are expressed relative to signal from a standard compound.

The reference compound is chosen so that the protons of it are well shielded than the typical protons in compounds of Interest, so that reference absorptions does not interfere with the resonances normally observed for organic compounds .

Preferably it should give a single sharp NMR signal.

It should be chemically un-reactive and easily removable from the sample after the measurement.

TMS



The most common reference compound used is tetramethyl silane. TMS is rich in H's(12) and methyl group H's are richer in electrons (larger σ) than in organic compounds and they are structurally equivalent.

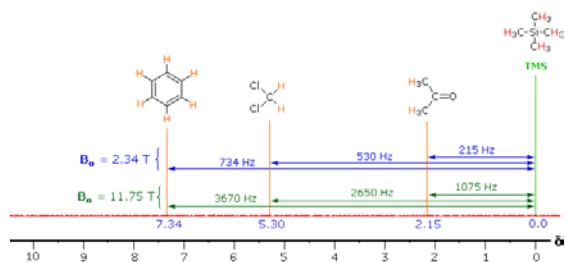
$$\omega_{\text{ref}} = \gamma_{\text{H}}(1 - \sigma_{\text{ref}})B_0$$

$\sigma_{\text{ref}} - \sigma_{\text{sample}}$ is a very small quantity.

Chemical Shift

$$\delta = \frac{V_{\text{sample}} - V_{\text{reference}}}{V_{\text{rf}}} \times 10^6 \text{ ppm}$$

$$\delta = \frac{\text{Shift in Hz}}{\text{Specrometer frequency MHz}} \text{ ppm}$$



Chemical shift is independent of the instrument field strength.

For bare $^1\text{H}(\sigma=0)$ at 2T field resonate at **90.0000** MHz;

A typical proton would have a frequency of 89.9995 MHz, it is an unwieldy number to report, further if a different instrument (different field) is used the frequency will be different.

Thus absorption positions are reported with respect to a reference. If a reference compound resonates at 89.9990MHz.

$$\text{Chemical shift: } \frac{89.9995 \cdot 10^6 - 89.9990 \cdot 10^6}{90 \cdot 10^6} = 5.556$$

$$\text{Chemical Shift (ppm): } \frac{89.9995 \cdot 10^6 - 89.9990 \cdot 10^6}{90 \cdot 10^6} = 5.556$$

$$\text{Frequency shift (Hz)} \quad 89.9995 \cdot 10^6 - 89.9990 \cdot 10^6 = 500$$

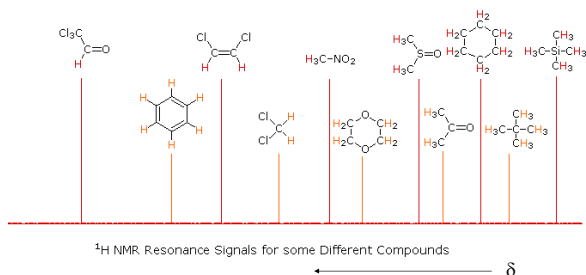
Frequency shift in a **300MHz** instrument (Hz):

$$\frac{(5.556) \cdot 300 \cdot 10^6}{10^6} = 1666.8$$

Entire proton range 300MHz instrument -12ppm (Hz):

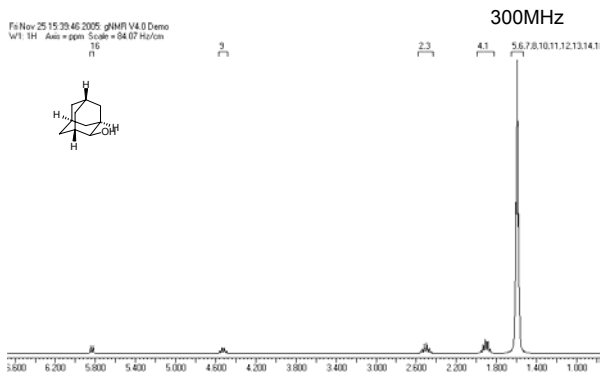
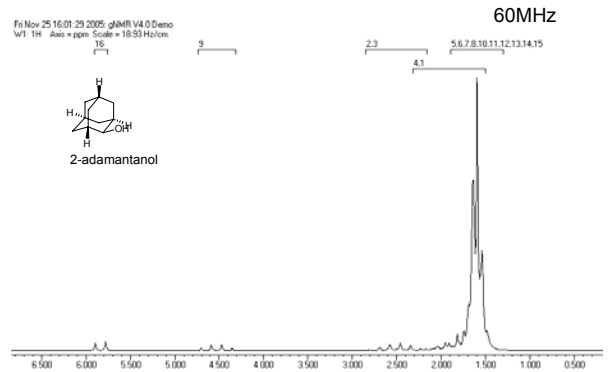
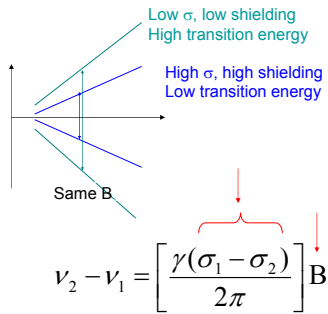
$$\frac{(12) \cdot 300 \cdot 10^6}{10^6} = 3600$$

Chemical shift decreases with increasing shielding



The higher the field the larger the frequency range over which the spectrum spreads. This enhances the resolution of peaks.

The higher the field the larger the frequency range over which the spectrum spreads. This enhances the resolution of peaks.



Precession frequencies of different nuclei.

Different nuclei having different magnetogyric ratios resonate at different frequencies. For bare nuclei of $I=1/2$, for example

$$\nu_0 = \frac{\gamma B_0}{2\pi}$$



Shimming:

Most critical requirement in NMR; *homogeneous B* at the sample
Small variations of B at the sample is corrected by the application of 'small magnetic fields' with coils situated in the magnet – shim coils - so that every part of the sample in the NMR tube experiences the same field strength.

Shimming is optimizing the homogeneity of the magnetic field; which increases the resolution of the peaks.

Careful control of temperature of the environment in the probe.

Spinning the sample; rotation of the sample average out inhomogeneous portions of field in the sample

Shim	Equation for Field Generated	Interaction Type
Z0	1	0
Z1	z	1
Z2	$2z^2 - (x^2 + y^2)$	1
Z3	$z[2z^2 - 3(x^2 + y^2)]$	2
Z4	$8z^2[z^2 - 3(x^2 + y^2)] + 3(x^2 + y^2)^2$	2
Z5	$48z^3[z^2 - 5(x^2 + y^2)] + 90z(x^2 + y^2)^2$	2
X	x	0
Y	y	0
ZX	zx	2
ZY	zy	2
X2-Y2	$x^2 - y^2$	1
XY	xy	1
Z2X	$x[4z^2 - (x^2 + y^2)]$	2
Z2Y	$y[4z^2 - (x^2 + y^2)]$	2
ZXY	zxy	2
Z(X2 - Y2)	$z(x^2 - y^2)$	2
X3	$x(x^2 - 3y^2)$	1
Y3	$y(3x^2 - y^2)$	1 (18 shims)

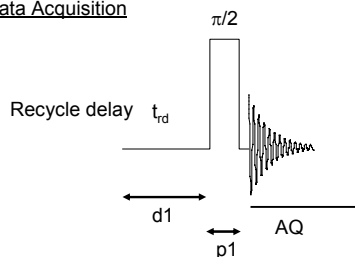
Tuning and Matching:

Tuning: In a probe there is a resonant circuit for each nuclei that is excited H, ¹³C etc. The frequency at which for example the ¹H resonant circuit is very sensitive and must be set to the carrier frequency of the ¹H pulses. Tuning ensures that the carrier frequency is indeed tuned to it.

Matching: Matching is the process of adjusting the impedance of the signal receiving resonant circuit (receiving signals from the Sample) until it corresponds with the impedance of the transmission line connected to it.

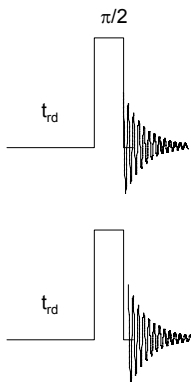
A poorly matched probe reflects a large fraction of the power of the pulses. This would make a 90° pulse much lower than 90°. It deteriorates the strength of the signal.

Data Acquisition



Single pulse experiment
Pulse program.

Phase Correction:



Short delay required to switch modes of the transceiver.

By that time the magnetic vector has moved and data acquisition will be on an FID starting with a phase angle.

This distorts the peak.

Different nuclei would have different starting phases as well.

Spin-Spin Coupling

The signals from non-equivalent 'adjacent' nuclei split into close groupings of resonances. This effect is due to the transmission of nuclear spin configuration information via bonding electrons. The signal splitting of spectra is small ~Hz, and is designated as J coupling (J = coupling constant). J is a molecular property and is independent of B.

In saturated compounds the transmission is not observed beyond three bonds in the usual NMR experiment.

$$\# \text{ split peaks} = 2nI + 1$$

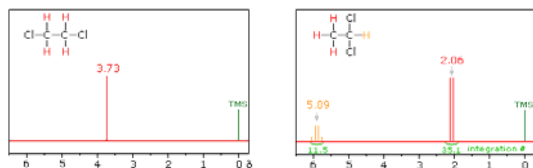
n = # of adjacent equivalent nuclei of nuclear spin q.n. I to nuclei.

$$\text{For 2 sets of adjacent nuclei, } \# \text{ split peaks} = (2n_A I_A + 1)(2n_B I_B + 1)$$

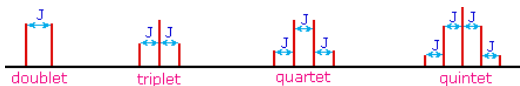
$$\text{If } J_{AC} = J_{BC} \text{ then } \# \text{ split peaks} = 2(n_A + n_B) + 1$$

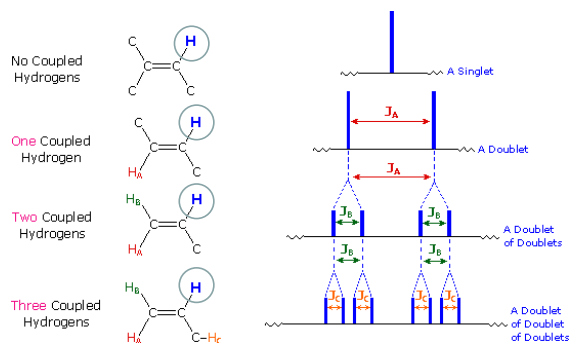
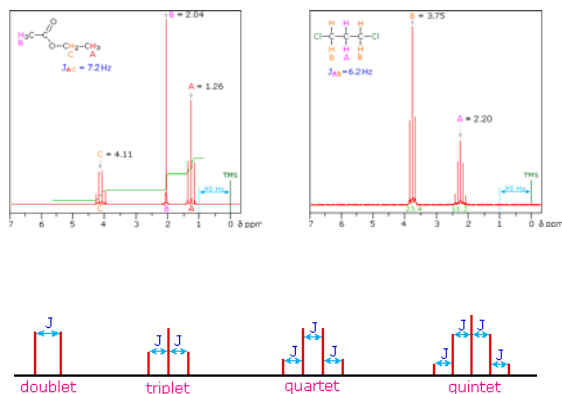
For I=1/2 nuclei the area ratio of split peaks is given by the Pascal's triangle coefficients.

1
1 1
1 2 1
1 3 3 1
1 4 6 4 1
1 5 10 10 5 1



$$J_{AB} = J_{BA}$$





Double Resonance Experiment (Decoupling):

Here two channels are used where one channel is used to saturate one type of nuclei with pulsed RF.

In such a situation the neighboring nuclei would not 'see' the different nuclear 'configurations'.

This technique is referred to as decoupling.

This simplifies the spectrum and give information about 'neighboring' nuclei.

This technique is used to decouple attached protons when generating ^{13}C spectra - broadband heteronuclear decoupling of protons.

Field Lock

The B_0 of the spectrometer might vary over time. The magnetic field in NMR experiments must be held constant during the duration of the experiment. The field lock accomplishes the task of maintaining a constant B_0 . The field lock is a separate NMR spectrometer within the spectrometer, which usually is tuned to the deuterium NMR resonance frequency (deuterium signal comes from the deuterated solvent used to prepare the sample). The 'second' spectrometer monitors the resonance frequency of the deuterium signal constantly. It makes any minor changes necessary to the (slowly) drifting spectrometer magnetic field to keep the deuterium resonance frequency constant. This keeps B_0 constant.

The deuterium lock serves another function. The resonance frequency of the deuterium signals of lock solvents are known values. The difference in resonance frequency of the lock solvent and TMS can be easily established. Therefore TMS need not be added to the sample to set the TMS signal as reference because the spectrometer can use the lock frequency to calculate $\delta = 0$ position of the spectrum.