Nuclear Magnetic Resonance Spectroscopy

Susanta Das

Susanta Das is presently
doing his PhD at IIT,
Guwahati in laser- matter
interaction. His fields of
interest are thin film,
nanoparticle, laser
produced plasma.

Introduction

Nuclear magnetic resonance in condensed matter was discovered simultaneously by Edward Purcell at Harvard and Felix Bloch at Stanford in 1946 using different instrumentation and techniques. Both groups observed the response of magnetic nuclei, placed in a uniform magnetic field, to a continuous radio frequency magnetic field as the field was tuned through a resonance. This discovery opened up a new form of spectroscopy which has become one of the most important tools for physicists, chemists, geologists, and biologists.

The Interaction of Radiation with Matter

Nuclear magnetic resonance, or NMR as it is abbreviated by scientists is one of a large number of phenomena associated with the interaction of electromagnetic radiation (e.m.) with matter. Other, more familiar examples of this type of interaction include the attenuation of X-rays by lead, the visible emission and absorption spectra of atoms, microwave heating of food and radio frequency (RF) induction heating of metals. What distinguishes these examples is primarily the frequency of e.m. radiation involved. X-rays have a typical frequency of around 10^{18} Hz, while the orange light of a sodium lamp corresponds to about 5×10^{14} Hz. Microwave cookers operate at frequencies of the order of 10^9 Hz and RF induction heaters use a frequency of order of 10^6 Hz. The experimental techniques of producing and detecting the radiation in each of these cases are clearly different. But there are similarities in the theoretical description of the processes involved. Using quantum theory one explains these phenomena in terms of transitions between states of different energy. The frequency ν of the radiation is related to the energy difference ΔE $(= E_2 - E_1)$ of the states by Einstein relation: $\Delta E = h\nu$, where h is Planck's constant. It is shown in *Figure* 1.
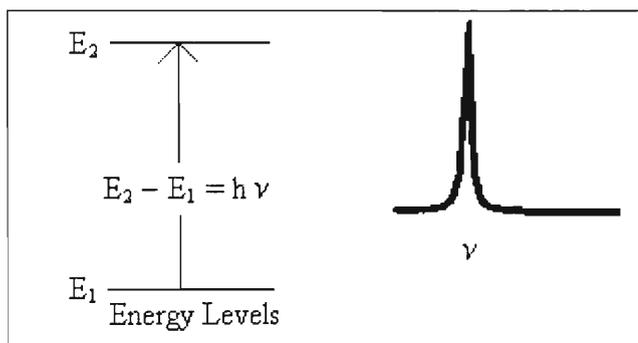
What is NMR?

NMR is a phenomenon which occurs when the nuclei of certain atoms are immersed in a static magnetic field and exposed to a second oscillating magnetic field. Some nuclei experience this phenomenon, and others do not, depending upon whether they possess a property called spin. A list of nuclei, which possess spin, is given in *Table* 1.

It is important to remember that, with NMR, we are performing experiments on the nuclei of atoms, not the electrons.

Theory of NMR Spectroscopy

Two properties of nuclear particles pertinent to an understanding of NMR spectroscopy are the net spin associated with the protons and neutrons (both have a spin quantum number of 1/2) and the distribution of positive charge. NMR spectroscopy is most often concerned with nuclei with spin $I = 1/2$, examples of

Nuclei	Unpaired protons	Unpaired neutrons	Spin
^1H	1	0	1/2
^2H	1	1	1
^{31}P	0	1	1/2
^{23}Na	2	1	3/2
^{14}N	1	1	1
^{13}C	0	1	1/2
^{19}F	0	1	1/2

Table 1.

Figure 2. Energy levels for a nucleus with spin quantum number 1/2 in a magnetic field.
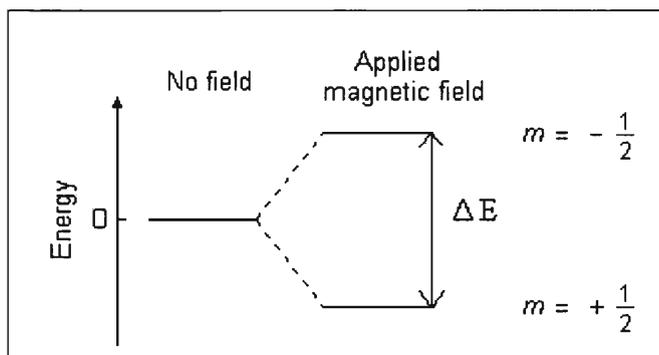
which include H^1, P^{31}, F^{19}. Spectra cannot be obtained on nuclei with $I = 0$ and only in special cases can spectra result from nuclei where $I \geq 1$. For a nucleus with $I = 1/2$, two values for the nuclear spin angular momentum quantum, $m_I = +1/2$ or $-1/2$, indicate the allowed orientations of the nuclear magnetic moment vector in an external magnetic field. The value $+1/2$ corresponds to alignment of the vector with the applied magnetic field and $-1/2$ opposed to it. The quantum number, m_I, has values: I, $(I-1)$,..., $(-I + 1)$, $-I$. For $I = 1$, m has values of $+1$, 0 and -1, corresponding respectively, to alignments with, perpendicular to, and opposed to the field. In the absence of a magnetic field, all orientations of the nuclear moment are degenerate. In the presence of an external field, however, this degeneracy will be destroyed. For a nucleus with $I = 1/2$, the $m_I = +1/2$ state will be in lower energy and the $-1/2$ state higher, as shown in *Figure* 2.

The energy difference between the two states, ΔE, is not very large compared to thermal energies, kT (where k is the Boltzmann constant). Consequently, thermal agitation reduces the excess of nuclei in the lower energy state, and the two states are nearly occupied at normal temperatures.

Box 1.

When the nucleus is in a magnetic field, the initial populations of the energy levels are determined by thermodynamics, as described by the Boltzmann distribution. This is very important, and it means that the lower energy level will contain slightly more nuclei than the higher level. It is possible to excite these nuclei into the higher level with electromagnetic radiation. The frequency of radiation needed is determined by the difference in energy (ΔE) between the energy levels.

Box 2.

A nucleus with a magnetic moment can be treated as though it were a bar magnet which spins on its axis. When placed in a magnetic field, the interaction of the magnetic moment with the external magnetic field produces a *torque*. This torque interacts with the angular momentum (in a classical way) and causes the magnetic moment to precess about the applied field vector, B_o, as indicated in *Figure* 3. The angular frequency of the precession is referred to as the *Larmor frequency*, ω, (in radians $\sec^{-1}$), the magnitude of which depends upon the applied field strength, B_o, the magnetic moment, and the spin angular momentum of the nucleus: $\omega = \gamma B_o$ The quantity, γ, referred to as the *gyromagnetic ratio*, is a constant for a given nucleus and represents the ratio of the nuclear magnetic moment to the nuclear angular momentum.
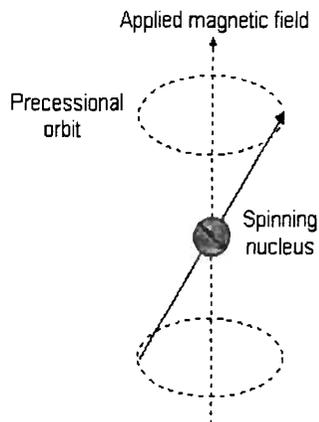
Figure 3. Precession of a nuclear moment in an applied field of strength B_o.

In an NMR experiment one applies a strong homogeneous magnetic field causing the nuclei to precess. Radiation of energy comparable to ΔE is then imposed with a radio frequency transmitter. The experimental procedure for carrying out this operation is shown in *Figure* 4. When the applied frequency from the radio transmitter is equal to the Larmor frequency, the two are said to be in resonance, and energy can be transferred to and
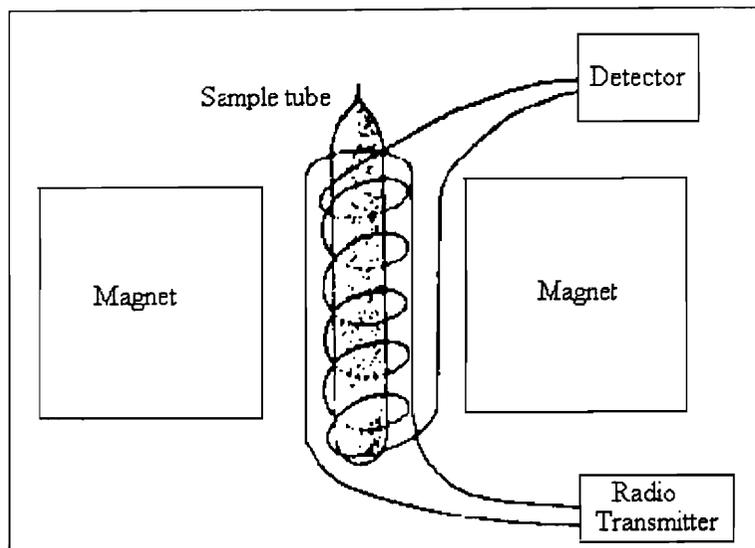
Figure 4. Schematic diagram of a simple NMR spectrometer.

When the applied frequency from the radio transmitter is equal to the Larmor frequency, the two are said to be in resonance, and energy can be transferred to and from the source (i.e., the transmitter) and the sample.

[1] Radio frequency.

from the source (i.e., the transmitter) and the sample. The net result of this resonance is that some nuclei are excited from the low energy (m_I = +1/2) state to the high energy state (m_I = -1/2; *Figure* 2) by absorption of energy from the source at a frequency equal to the Larmor frequency. Since ΔE = h ν and ω = $2\pi \nu$, ΔE is proportional to the Larmor frquency, ω. Energy will be extracted from the rf [1] source only when this resonance condition (ω = $2\pi \nu$) is fulfilled. If a third electronic component, namely a detector, is added to the apparatus (*Figure* 4), one can observe the frequency at which the loss in energy from the transmitter occurs, allowing the resonance frequency to be measured.

It is possible to match the Larmor frequency and the applied radio frequency either by holding the field strength constant (and hence ω constant) and scanning a variable applied radio frequency until matching occurs or, as is done in the more common NMR apparatus, by varying the field strength until ω becomes equal to a fixed applied frequency. In the latter method, fixed frequency probes (source and detector coils) are employed and the field strength at which resonance occurs is measured.

If the population of nuclei in the ground and excited states were equal, then the probability that the nucleus would emit energy under the resonance condition would equal the probability that the nucleus would absorb energy (i.e., transitions m_I(+1/2) $\rightarrow$ m_I(-1/2) would be as possible as m_I(-1/2) $\rightarrow$ m_I(+1/2)). No net change would then be detected by the radio frequency probe. As we know, in a strong magnetic field there will be a slight excess of nuclei aligned with the field (lower energy state) and consequently a net absorption of energy results. As energy is absorbed from the rf signal, enough nuclei could be excited after a finite period of time so that the population in the lower state would be equal to that in the higher state. Initially, absorption might be detected and this absorption will gradually disappear as the populations of the ground and excited states become equal. When this occurs, the sample is said to be *saturated*. If the NMR instrument is operated properly, saturation usually does not occur, because there are mechanisms for allowing nuclei to return to the

lower energy state without emitting radiation, which are basi-
cally spin-spin relaxation and spin-lattice relaxation. (*Box* 3.)

Box3. Selection Rule for NMR Spectroscopy

We know from spectroscopy that only certain transitions are *allowed* i.e., only certain ones actually take
place. There are usually rules – called *selection rules* – about which transitions can take place; these rules
normally relate to the quantum numbers which are characteristic of each state or energy level. In the case
of NMR, the selection rule refers to the quantum number m; only transitions in which m changes by one
(up or down) are allowed. This is sometimes expressed as

$$\Delta m = m \text{ (initial state)} - m \text{ (final state)} = \pm 1.$$

Another way of saying this is that one spin can flip between 'up' and 'down' or vice versa. In the case of
spin a single spin-half, the change in m between the two states is $(+1/2 - (-1/2)) = 1$, so the transition is
allowed.

Relaxation: How do nuclei in the higher energy state return to the lower state? Emission of radiation is
insignificant because the probability of re-emission of photons varies with the cube of the frequency. At
radio frequencies, re-emission is negligible. We must focus on non-radiative relaxation processes
(thermodynamics!). Ideally, the NMR spectroscopist would like relaxation rates to be fast - but not too fast.
If the relaxation rate is fast, then saturation is reduced. If the relaxation rate is too fast, line-broadening
in the resultant NMR spectrum is observed. There are two major relaxation processes described below:

1. Spin-spin relaxation: Spin-spin relaxation describes the interaction between neighbouring nuclei with
identical precessional frequencies but differing magnetic quantum states. In this situation, the nuclei can
exchange quantum states; a nucleus in the lower energy level will be excited, while the excited nucleus
relaxes to the lower energy state. There is no net change in the populations of the energy states, but the
average lifetime of a nucleus in the excited state will decrease. This can result in line-broadening.

2. Spin-lattice relaxation: Nuclei in an NMR experiment are in a sample. The sample in which the nuclei
are held is called the *lattice*. Nuclei in the lattice are in vibrational and rotational motion, which creates
a complex magnetic field. The magnetic field caused by motion of nuclei within the lattice is called the
lattice field. This lattice field has many components. Some of these components will be equal in frequency
and phase to the Larmor frequency of the nuclei of interest. These components of the lattice field can
interact with nuclei in the higher energy state, and cause them to lose energy (and return to the lower state).
The energy that a nucleus loses increases the amount of vibration and rotation within the lattice (resulting
in a tiny rise in the temperature of the sample). The relaxation time, T_1 (the average lifetime of nuclei in
the higher energy state) is dependent on the magnetogyric ratio of the nucleus and the mobility of the
lattice. As mobility increases, the vibrational and rotational frequencies increase, making it more likely for
a component of the lattice field to be able to interact with excited nuclei. However, at extremely high
mobilities, the probability of a component of the lattice field being able to interact with excited nuclei
decreases.

Detection of NMR Phenomena

There are only two basic methods by which we can arrive at the resonance condition and record a NMR spectrum: a) Continuous Wave NMR (CWNMR) and b) Pulsed NMR or pulsed Fourier Transform NMR (FTNMR).

CWNMR: Transitions between different energy levels occur if the frequency of radiation is equivalent to the energy difference between the two levels: In the old days of NMR, experiments were carried out by varying the frequency of radiation at constant magnetic field ('frequency sweep') and measuring the absorption of radiation by the different nuclei. Equivalently, the magnetic field strength could be varied at constant radiation frequency ('field sweep'). Until the 1970s all NMR spectrometers worked with this continuous wave technique.

FTNMR: A far better resolution and sensitivity in NMR was achieved by the introduction of pulsed Fourier transform techniques. In FTNMR the resonances are not measured one after another but all nuclei are excited at the same time by a radio frequency pulse.

Features of an NMR Spectrum

A simple (^{1}H) NMR spectrum is shown in *Figure* 5.

Peaks: There are two peaks because there are two different environments for the hydrogens – in the CH_3 group and attached to the oxygen in the COOH group. They are in different places in the spectrum because they need slightly different external magnetic fields to bring them into resonance at a particular radio frequency. The sizes of the two peaks give important information about the numbers of hydrogen atoms in each environment. It is not the height of the peaks that matters, but the ratio of the areas under the peaks. If you could measure the areas under the peaks in the diagram above, you could find that they were in the ratio of 3 (for the larger peak) to 1 (for the smaller one). That shows a ratio of 3:1 in the number of hydro-
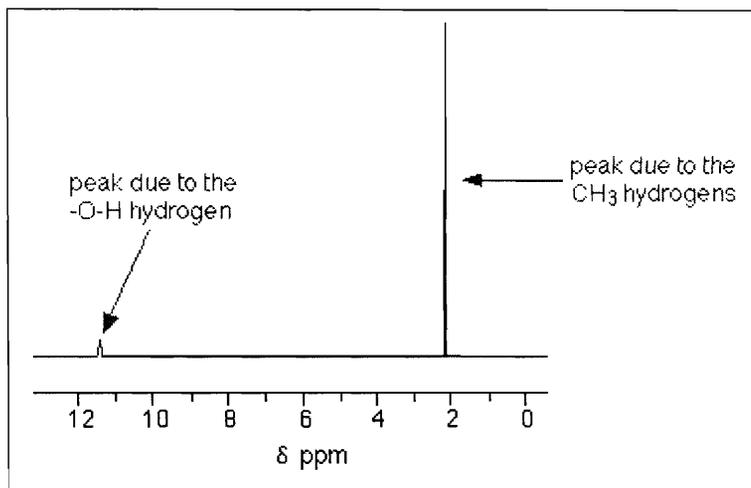
Figure 5. NMR spectrum for CH₃COOH, ethanoic acid.

gen atoms in the two environments which is exactly what you would expect for CH_3COOH.

The need for a standard for comparison – TMS: The diagram has a zero point at the right hand end of the scale. The zero is where you would find a peak due to the hydrogen atoms in tetramethylsilane – usually called TMS, $Si(CH_3)_4$. Everything else is compared with this.

$$CH_3-\underset{\underset{CH_3}{|}}{\overset{\overset{CH_3}{|}}{Si}}-CH_3$$
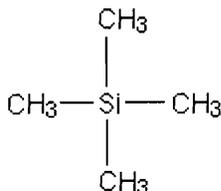
You will find that some NMR spectra show the peak due to TMS (at zero), and others leave it out. Essentially, if we have to analyse a spectrum which has a peak at zero, we can ignore it because that is the TMS peak.

The Chemical Shift: The horizontal scale shown as δ is called the chemical shift and is measured in parts per million (ppm). A peak at a chemical shift of, say, 2.0 means that the hydrogen atoms which caused that peak need a magnetic field two millionths less than the field needed by TMS to produce resonance.

A peak at a chemical shift of 2.0 is said to be downfield of TMS. The further to the left a peak is, the more downfield it is. The chemical shift is explained in detail in the next section.

Solvents for NMR Spectroscopy: NMR spectra are usually measured using solutions of the substance being investigated. It is important that the solvent itself does not contain any simple hydrogen atoms, because they would produce confusing peaks in the spectrum. There are two ways of avoiding this. You can use a solvent such as tetrachloromethene, CCL_4, which does not contain any hydrogen, or you can use a solvent in which any ordinary hydrogen atoms are replaced by its isotope, deuterium – for example, $CDCl_3$ instead of $CHCl_3$. Deuterium atoms have sufficiently different magnetic properties from ordinary hydrogen that they do not produce peaks in the area of the spectrum that we are looking at.

NMR Spectroscopy

Chemical Shift

When an atom is placed in a magnetic field, its electrons circulate about the direction of the applied magnetic field. This circulation causes a small magnetic field at the nucleus which opposes the externally applied field (*Figure* 6).

The magnetic field at the nucleus (the effective field) is therefore generally less than the applied field by a fraction σ, $B = B_0(1-\sigma)$. In some cases, such as the benzene molecule, the circulation
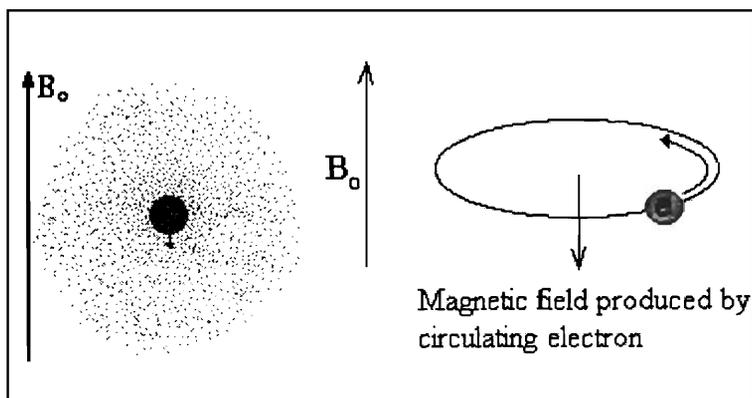
Figure 6. Magnetic field produced by electron which opposes B_0.

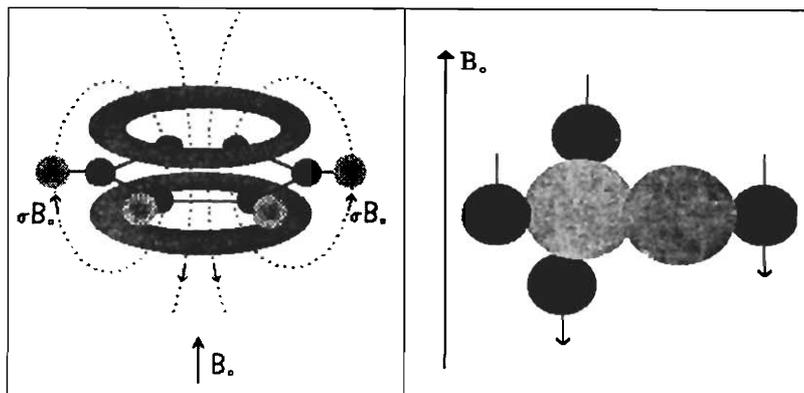Magnetic field produced by circulating electron

Figure 7(left). Deshielding phenomenon.

Figure 8 (right). Methanol molecule.

of the electrons in the aromatic orbitals creates a magnetic field at the hydrogen nuclei which enhances the B_o field. This phenomenon is called deshielding. In this example, the B_o field is applied perpendicular to the plane of the molecule. The ring current is traveling clockwise if you look down at the plane (*Figure* 7). The electron density around each nucleus in a molecule varies according to the types of nuclei and bonds in the molecule. The opposing field and therefore the effective field at each nucleus will vary. This is called the chemical shift phenomenon.

Consider the methanol molecule (*Figure* 8). The resonance frequency of two types of nuclei in this example differ. This difference will depend on the strength of the magnetic field, B_o, used to perform the NMR spectroscopy. The greater the value of B_o, the greater the frequency difference. This relationship could make it difficult to compare NMR spectra taken on spectrometers operating at different field strengths. The term chemical shift was developed to avoid this problem.

The chemical shift of a nucleus is the difference between the resonance frequency of the nucleus and a standard, relative to the standard. The chemical shift is also defined as the nuclear shielding divided by the applied field. The chemical shift is only a function of the nucleus and its environment, i.e., it is a molecular quantity. This quantity is reported in ppm and denoted by the symbol delta, δ. For example, in the case of proton NMR the reference compound is TMS. If the frequency of the

line we are interested in is ν (in Hz) and the frequency of the line from TMS is n_{TMS} (also in Hz), the chemical shift of the line is computed as: $\delta = (\nu - \nu_{REF})/\nu_{REF} = (\nu - \nu_{TMS})/\nu_{TMS}$ which is shown in *Figure* 9 (here the reference is TMS). As all the frequencies scale with the magnetic field, this ratio is independent of the magnetic field strength. Typically, the chemical shift is rather small and it is common to multiply the value for δ by 10^6 and then quote its value in ppm. With this definition the chemical shift of the reference compound is 0 ppm.

$$\delta = (\nu - \nu_{TMS}) \times 10^6 / \nu_{TMS} \qquad (1)$$

Sometimes we want to convert from shifts in ppm to frequencies. Suppose that there are two peaks in the spectrum at shifts δ_1 and δ_2 in ppm. What is the frequency separation between the two peaks? It is easy enough to work out what it is in ppm, it is just $(\delta_2 - \delta_1)$. Writing this difference out in terms of chemical shift given in (1) we have: $(\delta_2 - \delta_1) = (\nu_2 - \nu_{TMS}) \times 10^6 / \nu_{TMS} - (\nu_1 - \nu_{TMS}) \times 10^6 / \nu_{TMS} = (\nu_2 - \nu_1) \times 10^6 / \nu_{TMS}$.
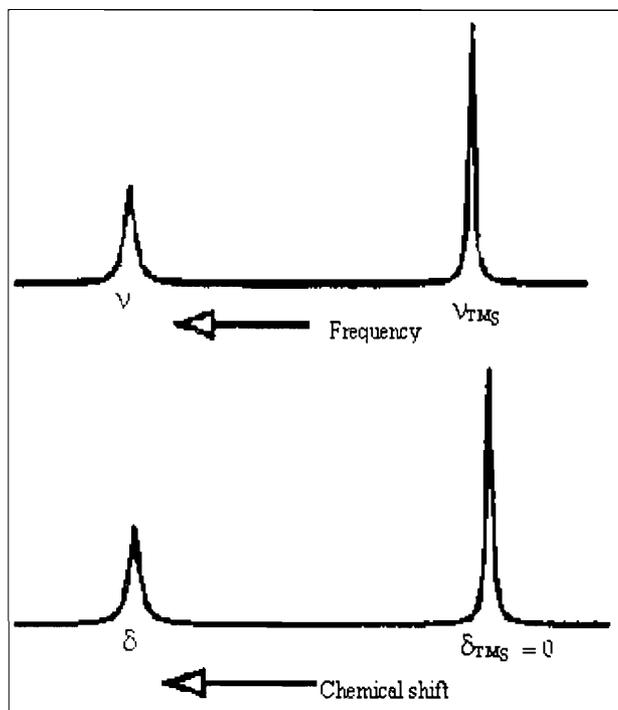
Multiplying both sides by ν_{TMS} now gives us what we want:$(\nu_2 - \nu_1) = \nu_{TMS} \times 10^{-6} \times (\delta_2 - \delta_1)$. The chemical shift is a very precise metric of the chemical environment around a nucleus. For example, the hydrogen chemical shift of a CH_2 hydrogen next to a Cl will be different from that of a CH_3 next to the same Cl. It is therefore difficult to give a detailed list of chemical shifts in a limited space. *Chart* 1 displays a chart of selected hydrogen chemical shifts of pure liquids and some gases.

The magnitude of the screening depends on the atom. For example, carbon-13 chemical shifts are much greater than hydrogen-1 chemical shifts.

Figure 9. An NMR spectrum can be plotted as a function of frequency, but it is more convenient to use the chemical shift scale in which frequencies are expressed relative to that of an agreed reference compound, such as TMS compound in the proton spectra.
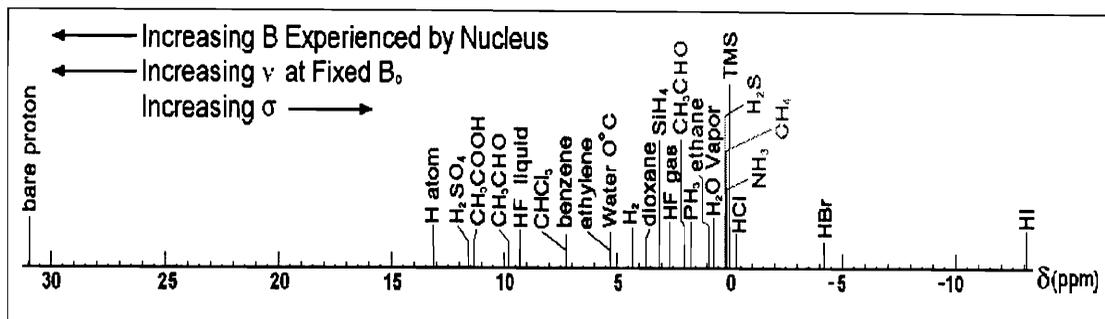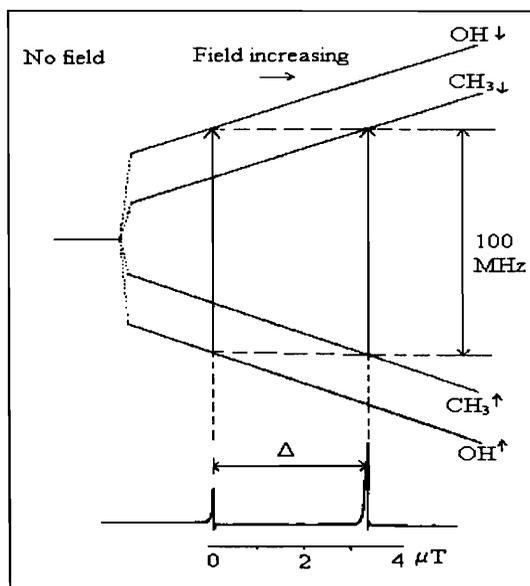
Chart 1.

Example: Chemical shift of CH$_3$OH: As we know that oxygen is a much better electron acceptor than carbon (since oxygen has the greater electronegativity), then the electron density about the hydrogen atom in C-H bonds should be considerably higher than in O-H bonds. We would thus expect $\sigma_{CH} > \sigma_{OH}$ and hence $B_{CH} = B_o(1-\sigma) < B_{OH} = B_o(1-\sigma_{OH})$. Thus the field experienced by the hydrogen nucles in O-H bonds is greater than that at the same nucleus in C-H bonds and, for a given applied field, the CH hydrogen nucleus will precess with a smaller Larmor frequency than that of OH. Put conversely, in order to come to resonance with radiation of a particular frequency (for example 100 MHz), a CH hydrogen requires a greater applied field than OH. The effect of a steadily increasing field on the energy levels of the hydrogen nucleus in the CH$_3$ and OH groups in CH$_3$OH is shown in *Figure* 10. The spectrum of methanol is shown at the foot of the figure where Δ is the chemical shift difference between the OH and CH$_3$.

Figure 10. The effect of magnetic field on the energy levels of the methyl and hydroxyl nuclei of methanol, CH$_3$OH. The applied field is increased rapidly initially (dotted portion) untill near resonance at 2.348T and then the increase is much slower.

Spin-Spin Coupling

Nuclei experiencing the same chemical environment or chemical shift are called equivalent. Those nuclei experiencing different environment or having different chemical shifts are nonequivalent. Nuclei which are close to one another exert an influence on each other's effective magnetic field. This effect shows up in the NMR spectrum when the nuclei are
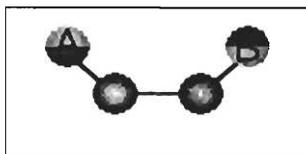
Figure 11. A and B three bonds away.

nonequivalent. If the distance between non-equivalent nuclei is less than or equal to three bond lengths, this effect is usually observable. This effect is called spin-spin coupling or J coupling.

Consider the following example. There are two nuclei, A and B, three bonds away from one another in a molecule (*Figure* 11). The spin of each nucleus can be either aligned with the external field, called spin up ●, or opposed to the external field such that the fields, called spin down ●. The magnetic field at nucleus A will be either greater than B_0 or less than B_0 by a constant amount due to the influence of nucleus B (*Figure* 12). There are a total of four possible configurations for the two nuclei in a magnetic field. Arranging these configurations in order of increasing energy gives the following arrangement (*Figure* 13). The vertical lines in this diagram represent the allowed transitions between energy levels. In NMR, an allowed transition is one where the spin of one nucleus changes from spin up ● to spin down ●, or spin down ● to spin up ●. Absorptions of energy where two or more nuclei change spin at the same time are not allowed. There are two absorption frequencies for the A nucleus and two for the B nucleus represented by the vertical lines between the energy levels in this diagram.

The NMR spectrum for nuclei A and B reflects the splittings observed in the energy level diagram. The A absorption line is split into 2 absorption lines centered on δ_A, and the B absorption
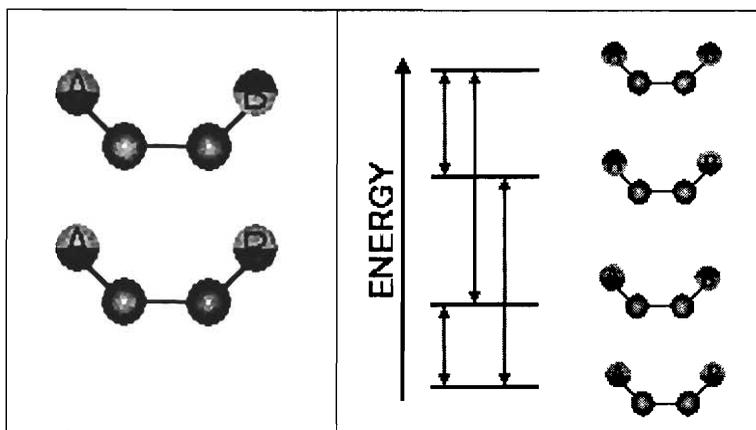

Figure 12 (left). Magnetic field at A.
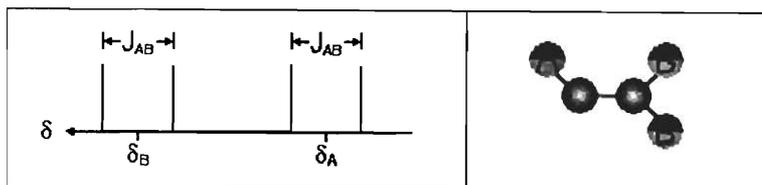Figure 13 (right). Four possible configurations.
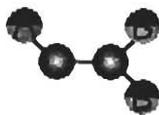
Figure 14 (left). J coupling constant.

Figure 15 (right).

line is split into 2 lines centered on δ_B. The distance between two split absorption lines is called the J coupling constant or the spin-spin splitting constant and is a measure of the magnetic interaction between two nuclei (*Figure* 14).

For the next example, consider a molecule with three spin 1/2 nuclei, one type A and two type B (*Figure* 15). The type B nuclei are both three bonds away from the type A nucleus. The magnetic field at the A nucleus has three possible values due to four possible spin configurations of the two B nuclei (*Figure* 16). The magnetic field at a B nucleus has two possible values (*Figure* 17). The energy level diagram for this molecule has six states or levels because there are two sets of levels with the same energy (*Figure* 18). Energy levels with the same energy are said to be degenerate. The vertical lines represent the allowed transitions or absorptions of energy. Note that there are two lines drawn between some levels because of the degeneracy of those levels. The resultant NMR spectrum is depicted in *Figure* 19. Note that the center absorption line of those centered at δ_A is twice as high as the either of the outer two. This is because there were twice as many transitions in the energy level diagram for this transition. The peaks at δ_B are taller because there are twice as many B type spins than A type spins.

Figure 16 (left). Three possible values of magnetic field at A.

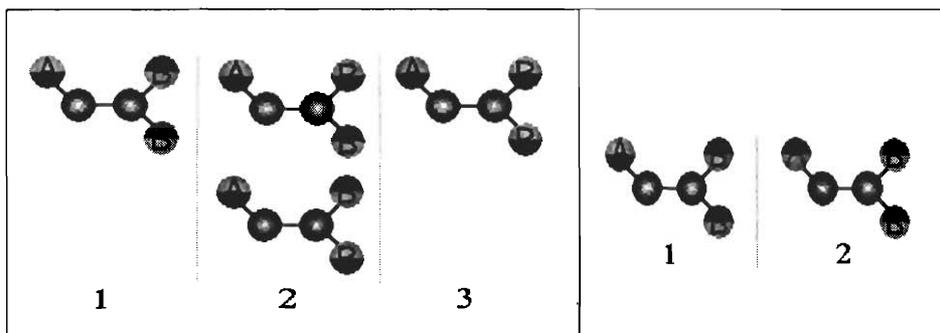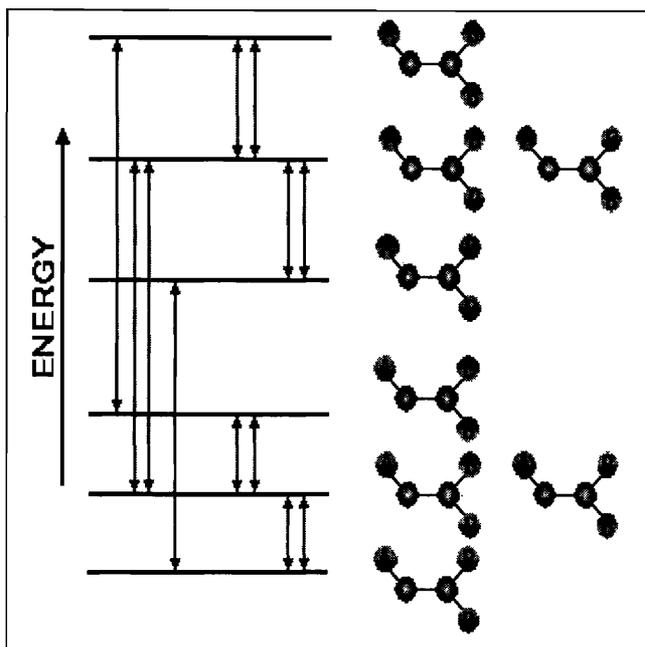Figure 17 (right). Two possible values of magnetic field at B.

Figure 18. Six energy state.

The complexity of the splitting pattern in a spectrum increases as the number of B nuclei increases. The following table contains a few examples.

Configuration Peak Ratios

Configuration	Peak Ratios
A	1
AB	1:1
AB_2	1:2:1
AB_3	1:3:3:1
AB_4	1:4:6:4:1
AB_5	1:5:10:10:5:1
AB_6	1:6:15:20:15:6:1

This series is called Pascal's triangle and can be calculated from the coefficients of the expansion of the equation $(x)^n$, where n is the number of B nuclei in the above table.
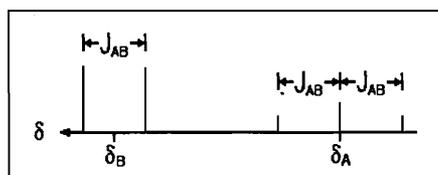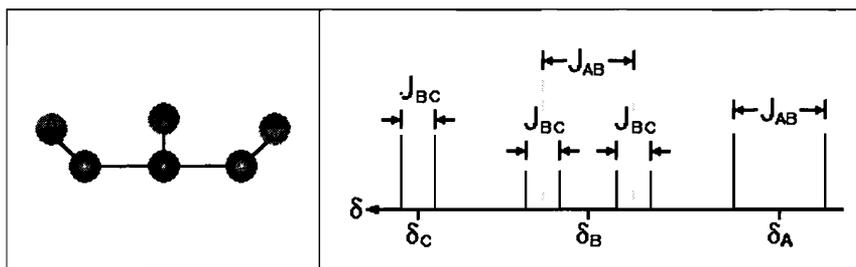
Figure 19. The resultant NMR spectrum.

When there are two different types of nuclei three bonds away (*Figure* 20) there will be two values of J, one for each pair of nuclei. In this case J_{AB} is greater J_{BC} (*Figure* 21).

Applications of NMR

Spectroscopy is the study of the interaction of electromagnetic radiation with matter. NMR spectroscopy is the use of the NMR phenomenon to study physical, chemical and biological properties of matter. As a consequence, NMR spectroscopy finds applications in several areas of science. NMR spectroscopy is routinely used by chemists to study chemical structure using simple one-dimensional techniques. Two-dimensional techniques are used to determine the structure of more complicated molecules. These techniques are replacing x-ray crystallography for the determination of protein structure. Time domain NMR spectroscopy techniques are used to probe molecular dynamics in solution. Solid state NMR spectroscopy is used to determine the molecular structure of solids. Other scientists have developed NMR methods· of measuring diffusion coefficients.

Suggested Reading

[1] B Cowan, *Nuclear Magnetic Resonance and Relaxation*, Cambridge Univeristy Press, 1997.
[2] C Banwell and E M McCash, *Fundamentals of Molecular Spectroscopy*, 4th edition, McGraw Hill, 1994.
[3] C Kittel, *Introduction to Solid State Physics*, Wiley Text Books, 1996.
[4] William Kemp, *Organic Spectroscopy*, Palgrave MacMillan, 1991.
[5] Dallas L Rabenstein, NMR Spectroscopy: Past and Present, *Analytical Chemistry*, Vol.73, 214A-223A, 2001.
[6] RJ Abraham, J Fisher and P Loftus, *Introduction to NMR spectroscopy*, John Wiley and Sons, 1992.

Address for Correspondence
Susanta Das
Department of Physics
Indian Institute of Technology
Guwahati 781039, India.
Email:
das_susanta@yahoo.com