25

Nuclear Magnetic Resonance (NMR) Spectroscopy

25.1. INTRODUCTION

Nuclear magnetic resonance (NMR) spectroscopy is the branch of spectroscopy dealing with the absorption of radio frequency radiation by substances held in a magnetic field. Absorption results from interaction of the radiation with the magnetic moment of nuclei in the sample, and it occur at different frequencies for nuclei in chemically different environments within a molcule. NMR has become an extremely important tool for elucidation of molecular structure, including stereochemistry and conformation. The sensitivity of the NMR spectrum to molecular structure makes it an excellent choice for identification testing in pharmaceutical analysis.

Advantages of NMR

NMR has several advantages over other analytical techniques. They are as follows :

1. A reference standard of the substance being analysed is not required.
2. It requires very little sample preparation prior to measurement.
3. NMR is applicable to a very wide range of organic and inorganic compounds.

Disadvantages of NMR

Inspite of several advantages, NMR has a few disadvantages also. They are as follows :

1. The high cost of the NMR instrumentation and its lack of sensitvity.
2. A minimum of 10 mg sample is required for NMR analysis (In some special cases it is very difficult to obtain).

Nuclear magnetic resonance spectroscopy is most often concerned with nuclei with $I = \frac{1}{2}$. Example of such nuclei are ^{1}H, ^{31}P and ^{19}F. Spectra can not be obtained from nuclei with $I = 0$. NMR is a powerful tool for investigating the nuclear structure. The first application of NMR was seen in 1951 to study the structure of ethyl alcohol. In the year 1952, Bloch and Purcell won the noble prize for the discovery of NMR.

25.2. THEORY OF NUCLEAR MAGNETIC RESONANCE SPECTROSCOPY

According to the quantum theory, a spinning nucleus can only have values for the spin angular momentum given by the equation.

Spin angular momentum = $[I(I + 1)]^{1/2}\ h/2\pi$ .....(1)

where I is the spin quantum number of the and h is the Planck's constant.

But $\mu = \gamma \times$ spin angular momentum

$\therefore \quad \mu = \gamma \times [I(I + 1]^{1/2} \cdot h/2\pi$

where

μ = magnetic moment of the nucleus

γ = gyromagnetic ratio

When a nucleus having a magnetic moment is introduced into a magnetic field. H_0, the two energy levels become separate corresponding to $m_1 = -\frac{1}{2}$ (anti parallel to the direction of magnetic field) and to $m_1 = +\frac{1}{2}$ (parallel to the direction of magneitc field). For a nucleus with $I = \frac{1}{2}$. the enegies E_1 and E_2 for the two states with $m_1 = +\frac{1}{2}$ and $m_1 = -\frac{1}{2}$, respectively, are

$$E_I = -\frac{1}{2}\left(\frac{rh}{2\pi}\right)H_0 \qquad \text{.....(2)}$$

and

$$E_I = +\frac{1}{2}\left(\frac{rh}{2\pi}\right)H_0 \qquad \text{.....(3)}$$

The energies of these two states are given in the following Fig. 25.1.

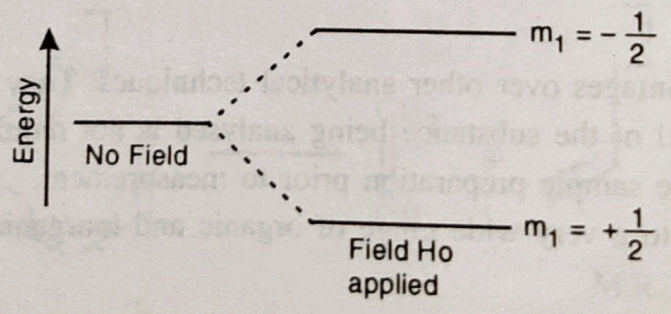


Fig. 25.1. Energy level of a nucleus with I = 1/2 in the absence and in the presence of a magnetic field.

When the nucleus absorbs energy, the nucleus will be promoted form the lower energy (Ground) state E_1 to the higher energy (Excited) state E_2 by absorption of energy, ΔE, equal to $E_2 - E_1$ form this it can be concluded that the absorption of energy (ΔE) changes the magnetic moment from the parallel state $m_1 = +\frac{1}{2}$ to the antiparallel state $\left(m_1 = -\frac{1}{2}\right)$.

From Bohr's relationship, the frequency 'V' at which energy is either absorbed or emitted is given by

$$V = \frac{E_2 - E_1}{h} \quad \text{....(4)}$$

On substituting equation (2) and (3) in (4), we get

$$V = \frac{\frac{1}{2}(rh/2\pi)H_0 + \frac{1}{2}(rh/2\pi)H_0}{h}$$

or

$$V = \frac{r}{2\pi}H_0 \quad \text{....(5)}$$

From equation (5) it is evident that the frequency absorbed or emitted by a nucleus in moving from one energy level to another is proportional to the applied magnetic field (H_0).

When a nucleus is placed in a system where it absorbs energy, it becomes excited. It then loses it energy to return to the unexcited state. It absorbs energy and again enters an excited state. This nucleus which alternately becomes excited and unexcited is said to be in a state of "resonance".

In order to determine the resonance frequency, the energy absorbed by nuclei is measured the magnetic field (H_0) is varied. As the field H_0 is increased so that precessional frequency of the nucleus increases and when this frequency becomes equal to the frequency of oscillation field, transitions occur between nuclear energy states. The energy absorbed in this process produces a signal at the detector and this signal is amplified and recorded as a band in the NMR spectrum. An NMR spectrum is plotted between absorption signal and the strength of the magnetic field (H_0). A typical NMR spectrum of ethylbenzene is given in the following Fig. 25.2.

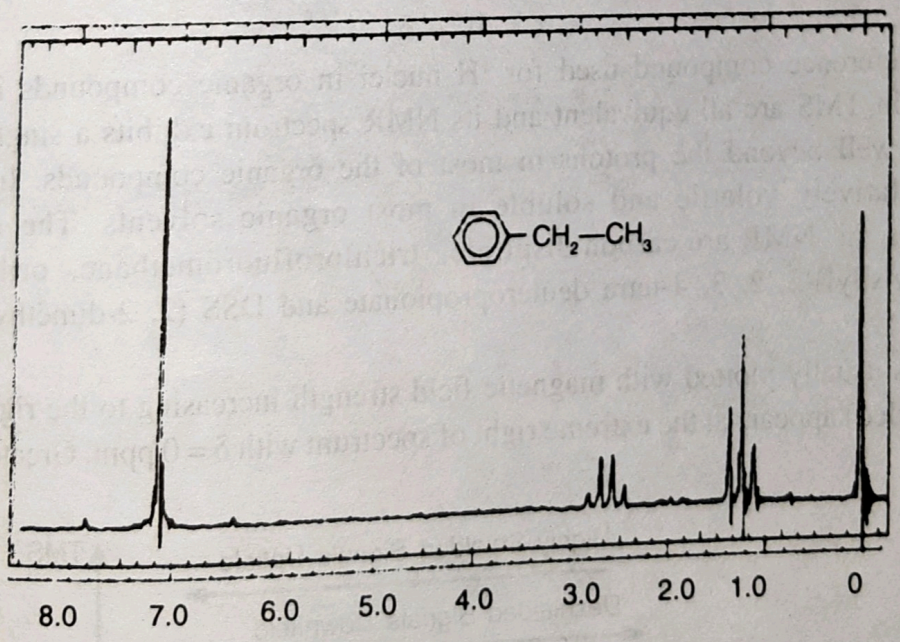


Fig. 25.2. 60 MHz ^1H NMR spectrum of ethylbenzene in $CDCl_3$ with TMS Internal Standard. Phenyl protons. 7.14 ppm; methylene protons, 2.63 ppm; methyl protons, 1.22 ppm.

CHEMICAL SHIFTS

In NMR spectrum, the number of signals gives us information about different kinds of protons in the molecule. In the same manner, the position of the signal tells us what kind of protons they are that is, whether they are aliphatic or aromatic, primary, secondary or tertiary or adjacent to halogen or other atom or group ..... etc. These different kinds of protons have different electronic environments and it is this

electronic environment that determines where a proton absorbs in the NMR spectrum.

According to the basic NMR equation, $V = \gamma H_0/2\pi$ in which gyromagnetic ratio (r) is an intrinsic property of the nucleus. Only a single peak should obtain from the interaction of radiofrequency energy and a strong magnetic field on a proton. The peak area is proportional to the number of protons it represents. But it is not so. The nucleus is shielded to a small extent by its electron cloud whose density varies with the environment. This variation gives rise to different absorption positions in the range of about 60 MHz to 100 MHz.

A shielded proton requires a higher applied field strength (H_0) and a deshielded proton requires a lower applied field strength in comparison to a naked proton, in order to provide the particular effective field strength at which absorption takes place.

In other words, shielding shifts the absorption up field or higher field, while deshielding shifts the absorption to downfield or lower field. Such shift in the position of NMR absorption, arising from shielding or deshielding is called "Chemical Shift".

The position of the peaks in an NMR spectrum relative to the reference peak is expressed in terms of chemical shift, δ which is given as

$$\delta = \frac{H_0(\text{reference}) - H_0(\text{sample})}{H_0(\text{reference})} \times 10^6 \text{ ppm}$$

The chemical shift δ is dimensionless and is expressed in parts per million (ppm) on account of the factor 10^6.

An alternative system which is generally used for defining the position of the resonance relative to the reference is as signed tau (τ) scale. On this scale the reference is assigned the arbitary position of 10 and the values of other resonances are given by

$$\tau = 10 - \delta$$

The normal reference compound used for ^1H nuclei in organic compounds is tetramethylsilane (TMS). The protons in TMS are all equivalent and its NMR spectrum exhibits a single resonance line at a high applied field well beyond the protons in most of the organic compounds. In addition, TMS is chemically inert, relatively volatile and soluble in most organic solvents. The other examples of reference compounds in NMR are carbon disulfide, trichlorofluoromethane, orthophosphoric acid, sodium 3-(Trimethylsilyl)-2, 2, 3, 4-tetra deuteropropionate and DSS (2, 2-dimethyl-2-2silapentane-5-sulfonate).

NMR signal is usually plotted with magnetic field strength increasing to the right. Thus, the signal for TMS (highly shielded) appears at the extreme right of spectrum with $\delta = 0$ ppm. Greater is the deshielding

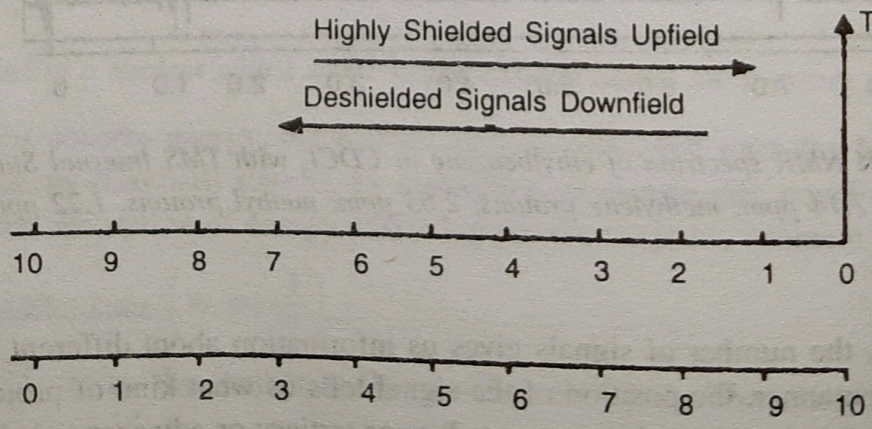
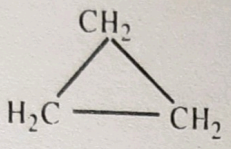


Fig. 24.3. Chemical shift.

of protons, larger will be the value of δ. For most of the organic compounds, signals appears downfield to the left of TMS signal. The value of chemical shifts for protons in different environments are (Table 25.1, 25.2, 25.3 and 25.4).

TABLE 25.1
Chemical Shift for Various Types of Protons.

Types of Protons		*Chemical Shift in ppm*	
		δ	τ
(*i*) Cyclo-propane		0.2*	9.8
(*ii*) Primary	R – CH_3	0.9	9.1
(*iii*) Vinylic	C = C – H	4.6-5.8	5.4-4.2
(*iv*) Acetylenic	C ≡ C – H	2-3.5	8-6.5
(*v*) Aromatic	Ar – H	6-9.0	4-1.0
(*vi*) Fluorides	H – C – F	4-4.5	6-5.5
(*vii*) Chlorides	HC – Cl	3-4	7-6
(*viii*) Alcohols	H – C – OH	3.4-4	6.6-6
(*ix*) Ethers	H – C – OR	3.4-4	6.6-6
(*x*) Esters	H – C – COOR	2-2.2	8-7.8
(*xi*) Acids	H – C — COOH	2-2.5	8-7.5
(*xii*) Aldehydic	RCHO	9-10	1-10
(*xiii*) Hydroxy	R – OH	1-5.5	9 - 4.5
(*xiv*) Phenolic	Ar – OH	4-12	6 to – 2
(*xv*) Enolic	C = C – OH	15-17	– 5 to – 7
(*xvi*) Carboxylic	RCOOH	10.5-12	– 0.5- – 2

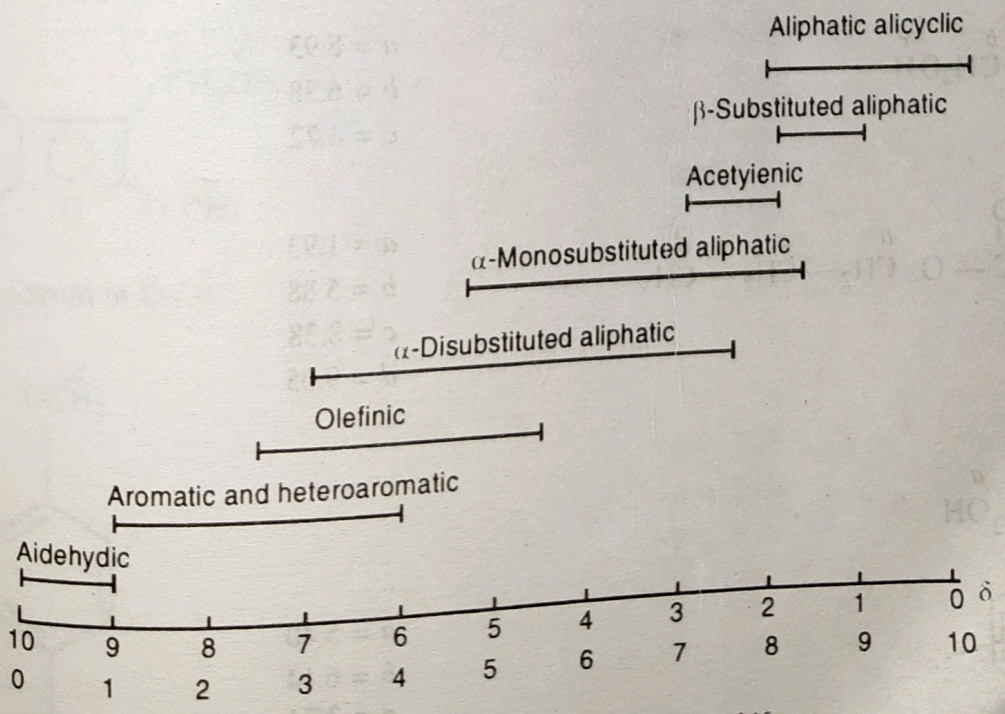
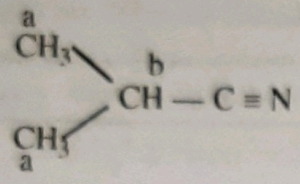
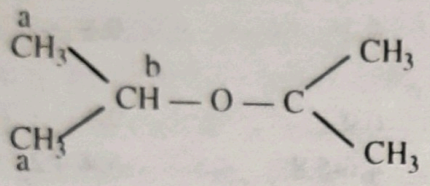
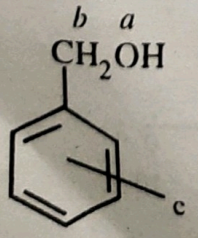


Fig. 25.3a. General regions of chemical shifts.

TABLE 25.2
Tau Values in Some Compounds.

Compound	Tau Values
(*i*) 	*a* = 8.67 *b* = 7.28
(*ii*) 	*a* = 8.95 *b* = 6.44
(*iii*) $\overset{a}{C}H_3-\overset{b}{C}H_2-\overset{c}{C}H_2-I$	*a* = 8.98 *b* = 8.14 *c* = 6.83
(*iv*) $\overset{a}{C}H_3-\overset{\overset{O}{\|\|}}{C}-\overset{b}{N}H-\overset{c}{C}H_3$	*a* = 8.08 *b* = 1.87 *c* = 7.30
(*v*) $\overset{a}{C}H_3-\overset{b}{C}H-\overset{\overset{O}{\|\|}}{C}-\overset{c}{C}H_3$	*a* = 8.93 *b* = 7.52 *c* = 7.88
(*vi*) $\overset{a}{C}H_3-\overset{b}{C}H_2O\overset{c}{H}$	*a* = 8.93 *b* = 6.38 *c* = 4.72
(*vii*) $H-\overset{\overset{O}{\|\|}}{C}-O\ \overset{b}{C}H_2-\overset{c}{C}H_2-\overset{d}{C}H_3$	*a* = 1.93 *b* = 5.88 *c* = 8.38 *d* = 9.05
(*viii*) 	*a* = 5.40 *b* = 6.14 *c* = 2.74

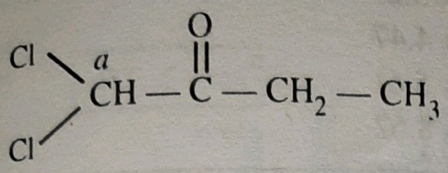
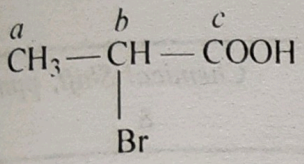
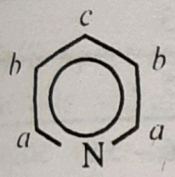
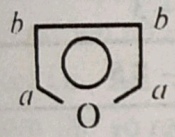
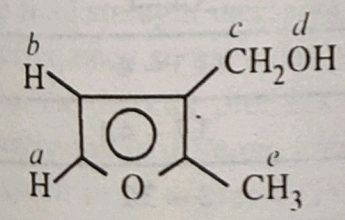
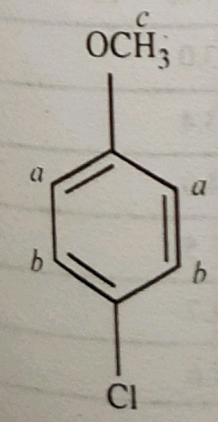
Compound	Tau Values
(*ix*) 	$a = 6.05$ $b = 5.65$ $c = 8.62$
(*x*) 	$a = 8.17$ $b = 5.48$ $c = -1.93$
(*xi*) 	$a = 1.44$ $b = 2.82$ $c = 2.44$
(*xii*) 	$a = 2.66$ $b = 3.71$

TABLE 25.3

Tau Values for Different Sets of Protons in Some More Complicated Structures

Compound	Tau Values
(*i*)  (Spectrum in D_2O)	$a = 2.89$ $b = 3.82$ $c = 5.80$ $d = 5.77$ $e = 7.87$
(*ii*)  (Spectrum in D_2O)	$a = 3.33$ $b = 2.88$ $c = 6.35$

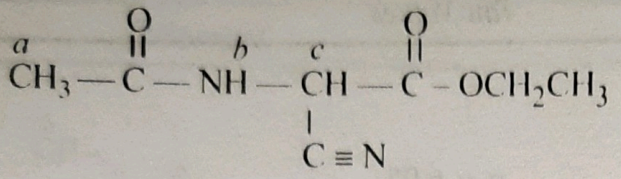
Compound	*Tau Values*
(*iii*) 	*a* = 7.90 *b* = 3.23 *c* = 4.47 *d* = 5.61 *e* = 8.62

TABLE 25.4

Type of Proton		*Chemical Shift, ppm* δ
Primary	RCH_3	0.9
Secondary	R_2CH_2	1.3
Tertiary	R_3CH	1.5
Vinylic	C = C – H	4.6 – 5.9
Acetylenic	C ≡ C – H	2 – 3
Aromatic	Ar – H	6 – 8.5
Benzylic	Ar – C – H	2.2 – 3
Allylic	C = C – CH_3	1.7
Carbonyl Compounds	HC – C = 0	2 – 2.7
Aldehydic	RCHO	9–10
Alcohols	HC – OH	3.4 –4
Ethers	HC – OR	3.3 – 4
Esters	RCOO – CH	3.7 – 4.1
Esters	HC – COOR	2 – 2.2
Acids	HC – COOH	2 – 2.6
Carboxylic	RCOOH	10.5 – 10
	CH_3 – Cl	3.0
	R – CH_2 – Cl	3.4
	R_2CH – Cl	4.0
	CH_3 – C – Cl	1.5
	R – CH_2 – C – Cl	1.7
	R_2CH – C – Cl	1.6

INSTRUMENTATION

The following is the schematic diagram (Fig. 25.4) of NMR spectrometer.

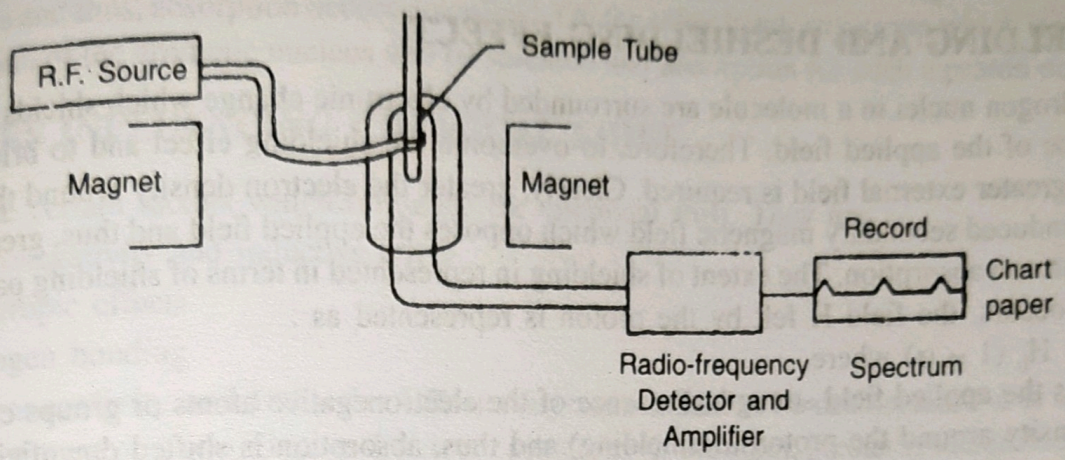


Fig. 25.4. Nuclear magnetic resonance spectrometer.

A NMR spectrometer maues use of a magnet, a radiofreq ency, a detector and an amplifier. The detection system is used to note that energy being transferred from the radiofreq ency beam to the n cle s.

The same nder investigation is tauen in a glass t be which is placed between the pole faces of a magnet. A radio freq ency so rce (V = 60 MHz) is made to fall on the sample. It can be done by feeding energy into a coil wo nd ro nd the sample t be. A signal is detected if the n clei in the sample resonates with the so rce, *i.e.*, ΔE, energy req ired to flip the proton is the same as that of the so rce. Energy is transferred from the so rce via n clei to the detector coil. The o tp t from the detector is fed to a strip chart recorder after req ired amplification.

Protons being in different electronic environments in a molec le cannot resonate at exactly 60 MHz. For practical p rposes, radiofreq ency so rce is held steady at the said freq ency and the field strength (H_0) is varied by placing small electromagnet to the pole faces of the main manget. By increasing the c rrent flowing thro gh these electromagnets, the total field strength is increased.

As the field strength increases, the precessional freq ency of each proton increased ntil resonance with the radiofreq ency so rce taues place. As a proton or set of eq ivalent protons comes to resonance, the signal from the detector prod ces a peau on the chart paper. The NMR spectr m consists of series of peaus that correspond to different applied field stengths. Each peau means a set of protons. The following is the NMR spectr m of m-cresol (Fig. 25.5).

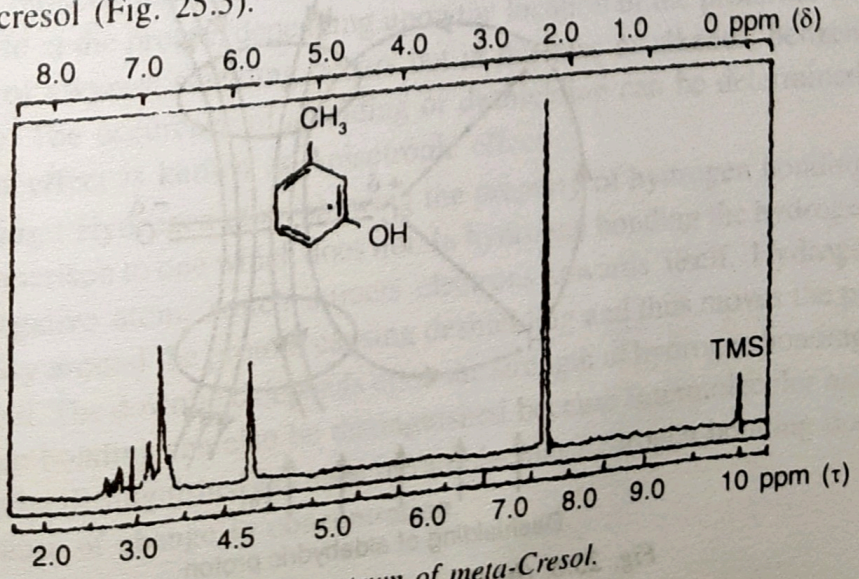


Fig. 25.5. NMR spectrum of meta-Cresol.

Signals and their absorption positions.

(*i*) Three proton singlet (CH_3) = 7.75 τ

(*ii*) One proton singlet (OH) = 4.32 τ

(*iii*) Four protons unsymmetrical pattern = 2.75 – 3.5τ

25.5. SHIELDING AND DESHIELDING EFFECT

Hydrogen nuclei in a molecule are surrounded by electronic change which shields the nucleus from the influence of the applied field. Therefore, to overcome the shielding effect and to bring the protons to resonance, greater external field is required. Clearly, greater the electron density around the proton, greater will be the induced secondary magnetic field which opposes the applied field and thus, greater external field will cause proton absorption. The extent of shielding in represented in terms of shielding parameter α. When absorption occurs, the field H felt by the proton is represented as :

$H = H_0 (1 - \alpha)$ where

H_0 is the applied field strength. Presence of the electronegative atoms or groups cause reduction in electron density around the proton (deshielding) and thus, absorption is shifted downfield. The following is the diagramatic representation of shielding and deshielding [Fig 25.6 (*a*) and 25.6 (*b*)].

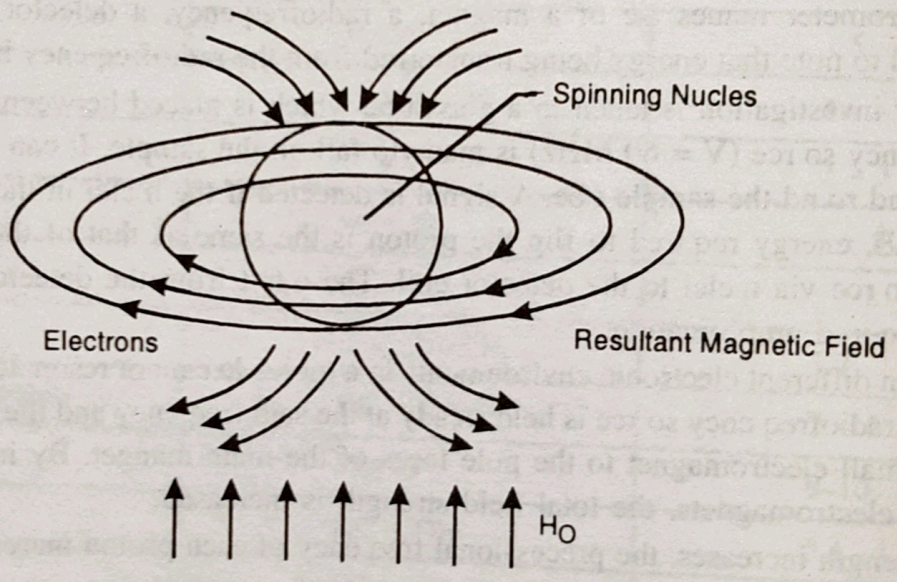


Fig. 25.6. (a) Diamagnetic shielding about the nucleus.

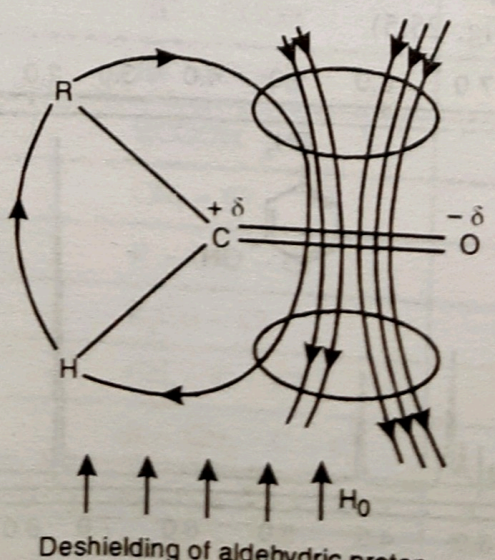


Fig. 25.6. (b) Deshielding of aldehydic proton.

In case of olefines, acetylenes, aldehydes, ketones, acids, esters, nitrites ..... etc., if proton is present in the positive region it will be shielded and absorption occurs upfield. On the other hand, if the proton lies in the negative region, its absorption is downfield. In the case of benzene and other aromatic compounds, strong diamagnetic currents are induced by the applied field. This causes paramagnetic shielding at the aromatic protons and thus, absorption occurs downfield. On the other hand, any group which is present above or below the plane of the aromatic nucleus will be shielded and absorption for such a proton occurs upfield.

25.6. FACTORS INFLUENCING CHEMICAL SHIFT

There are certain factors which influence the chemical shift. They are :

1. Electronegativity and inductive effect
2. Anisotropic effects
3. Hydrogen bonding

1. Electronegativity and Inductive Effect : A proton is said to be deshielded if it is attached with an electronegative atom or group. Greater the electronegativity of the atom, greater is the deshielding caused to the proton. If the deshielding is more for a proton, then its δ value will also be more. Attachment of electronegative atom or group to the carbon bearing the proton causes a downward shift (higher value of δ). This effect is illustrated by the δ values for the protons in methyl halides, CH_3X.

$CH_3 - F$	$CH_3 - Cl$	$CH_3 - Br$	$CH_3 - I$
δ 4.26	δ 3.0	δ 2.82	δ 2.16

The order of electronegativity of the halogens is :

$$I < Br < Cl < F$$

Here iodine; being least reactive is the least effective of the halogens in withdrawing electrons from the protons. Thus the electrons of iodine provide the largest shielding effect. So the signal from the CH_3I protons is found to the farthest upfield (close to TMS signal). In the same manner, one finds the protons in the CH_3F to be the most deshielded than any if the protons in the series and hence their signal is found farthest downfield (Away from TMS signal). The signals of the protons of CH_3Cl and CH_3Br are intermediate. As the distance of the proton from the electronegative atom increases, the deshielding effect of it decreases. This can be shown clearly with the following example.

CH_3–Cl	$CH_3 - C - Cl$
δ 3.0	δ 1.5

2. Anisotropic Effect (Space Effect) : Circulation of electrons, especially the π electrons about the nearby nuclei generates an induced field which can either oppose (causing shielding) or reinforce (causing deshielding) the applied field at the proton, depending upon the location of the proton or the space occupied by the proton. In the case of alkynes, shielding occurs but in the case of alkenes, benzene and aldehydes etc. dishielding takes place. The occurence of shielding or deshielding can be determined by the location of proton in space and this effect is known as Anisotropic effect.

3. Hydrogen bonding : Hydrogen atom showing the property of hydrogen bonding in a compound absorbs at downfield in comparison to one which does not. In hydrogen bonding the hydrogen bonded proton is attached to an electronegative atom, which attracts electrons towards itself. Hydrogen bonding thus decreases the electron density around the proton, causing deshielding and thus moves the proton absorption to the lower field or downfield. The downfield depends upon the strength of hydrogen bonding. Intermolecular and intramolecular hydrogen bonding can also be distinguished because intermolecular and intramolecular hydrogen bonding can also be distinguished because intramolecular hydrogen bonding does not show any shift in chemical shift because of change in concentration.

25.7. SOLVENTS USED IN NMR

A substance free of proton should be used as a solven (*i.e.*, solvent does not give absorption of its own in NMR spectrum). Another important factor is the solvent should be capable of dissolving at least 10% of the substance under investigation. The following is the list of solvents commonly used in NMR spectroscopy (Table 25.1.).

TABLE 25.1
Common NMR Solvents

Carbon Tetrachloride	CCl_4
Benzene – d_6	C_6D_6
Hexachlorobutadiene	C_4Cl_6
Tetrachloro ethylene	C_2Cl_4
Carbon disulfide	CS_2
Chloroform-d_1	$CDCl_3$
Acetone-d_6	$CD_3–CO–CD_3$
Methanol - d_4	CD_3OD
Dimethyl sulfoxide-d_6	$CD_3–SO–CD_3$
Dimethyl formamide-d_7	$(CD_3)_2NCDO$
Pyridine-d_5	C_5D_5N
Acetonitrile-d_3	CD_3CN
Formic acid-d_2	DCOOD
Trifluoroacetic acid	CF_3COOH
Deuterium oxide	D_2O

The important characteristics of solvents used in NMR are as follows :

1. It should be magnetically isotropic and chemically inert.
2. It should be free from protons
3. It should dissolve the sample to a reasonable extent.

25.8. PEAK AREA AND PROTON COUNTING

In an NMR spectrum, various peaks represent equivalent sets of protons. The size or area of each peak tells the number of protons in each set present in the compound under investigation. The area under an NMR signal is directly proportional to the number of protons giving rise to signal. Consider the spectrum of toluene. It shows two types of protons as is clear from the two signals. If the number of squares under each signal are counted, it will be found that the areas under the two peaks have the ratio 5 : 3. From this it is evident that, in toluene, five protons are of one kind and three protons are of another and therefore, we can say that NMR spectrum of toluene represents two kinds of protons which are signal in the ratio 5 : 3.

1. One signal due to five protons (downfield due to deshielding).
2. Another signal due to three protons (upfield).

25.9. SPLITTING OF THE SIGNALS

Each signal in an NMR spectrum represents one kind or one set of protons in a molecule. It is observed that in certain molecules, a single peak is not observed, instead of that, a multiple (group of peaks) is observed. Let us consider a molecule of ethyl bromide, $\overset{1}{C}H_3\overset{2}{C}H_2Br$. This molecule has two kinds of proton

in it and therefore, two signals are expected in it NMR spectrum.

The following is the NMR spectrum of ethylbromide (Fig. 25.7).

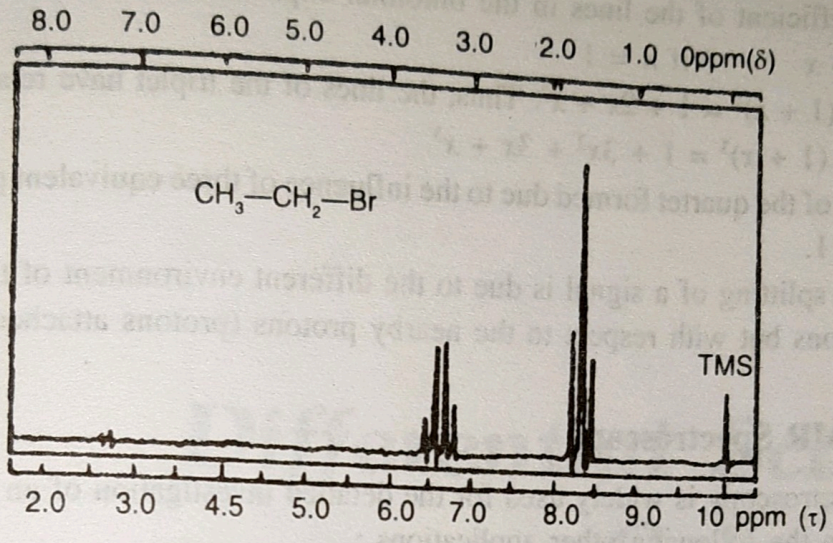


Fig. 25.7. NMR spectrum of Ethyl bromide.

It is evident from the above figure that for each kind of protons, we do not get signlets but a group of peaks are observed. For '1' kind of protons (CH_3), a triplet (*i.e.*, a group of 3 peaks) is observed and a quartet (group of 4 peaks) is noticed for '2' kind of protons ($-CH_2-$). The NMR splitting is due to spin-spin coupling.

25.9. SPIN-SPIN COUPLING

Bands in the NMR spectra of molecules containing more than one magnetic nucleus are frequently split into multiples. The splitting arises from an interaction of the nuclear spins which is mediated by the bonding electrons. This interaction is called spin-spin coupling or scalar coupling. In the simplest cases, the multiplet appears as a series of evenly spaced bands and the distance between the bands in Hertz is the coupling constant. Coupling constants are represented by the symbol ${}^nJ_{ab}$, where the subscripts identify the spins or groups of spins that are coupled, and n is the number of bonds between the nuclei.

To understand it properly, consider a molecule of ethyl bromide, CH_3CH_2Br. The spin of two protons ($-CH_2-$) can couple with the adjacent methyl group ($-CH_3$) in three different ways relative to the external field. The three different ways of alignment are represented as follows :

1. ↑↑ (Reinforcing)
2. ↓↑ ↑↓ (Not effecting)
3. ↓↓ (Opposing)

↑ External field

Thus, a triplet of peaks results with the intensity ratio of 1 : 2 : 1 which corresponds to the distribution ratio of alignment.

Similarly, the spin of three protons (CH_3-) can couple with the adjacent methylene group ($-CH_2-$) in four different ways relative to the external field.

1. ↑↑↑ (Strongly reinforcing)
2. ↑↑↓ ↑↓↑ ↓↑↑ (Weakly reinforcing)
3. ↓↓↑ ↓↑↓ ↑↓↓ (Weakly opposing)
4. ↓↓↓ (Strongly opposing)

↑ External field

Thus, quartet of peaks results with an intensity ratio of 1 : 3 : 3 : 1 which corresponds to the distribution ratio of all the alignments. The relative intensities of the individual lines of a multiple correspond to the numberical coefficient of the lines in the binomial expression.

$(1 + x)^n = 1 + x$ if $n = 1$

If $n = 2$, then $(1 + x)^2 = 1 + 2x + x^2$. Thus, the lines of the triplet have relative intensities 1 : 2 : 1.

If $n = 3$, then $(1 + x)^3 = 1 + 3x^2 + 3x + x^3$.

Thus, the lines of the quartet formed due to the influence of three equivalent protons will have relative intensities 1 : 3 : 3 : 1.

Therefore, the splitting of a signal is due to the different environment of the absorbing proton not with respect to electrons but with respect to the nearby protons (protons attached to the adjacent carbon atom).

Applications of NMR Spectroscopy

The NMR spectroscopy is widely used for the detailed investigation of an unknown compound. In addition to this, it has the following other applications :

1. Identification of structural isomers
2. Detection of hydrogen bonding
3. Detection of aromaticity
4. Distinction between cis-trans isomers.
5. Detection of electronegative atom or group
6. Detection of some double character due to resonance
7. For quantitative analysis.

❑❑

Patience is the best remedy for every trouble.