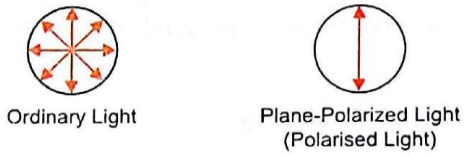
OPTICAL ISOMERISM

What is Optical Activity:
Optical isomerism is type of stereoisomerism. Optical isomers have the ability to rotate the plane polarized light. This property is often referred as Optical Activity.

A beam of ordinary light consists of electromagnetic waves that oscillate in an infinite number of planes at right angles to the direction of light travel. When a beam of ordinary light is passed through a device called a polarizer, or a Nicol prism (made of calcite or CaCO3), light is found to vibrate in only one plane and is said to be plane-polarized. Light waves in all other planes are blocked out.

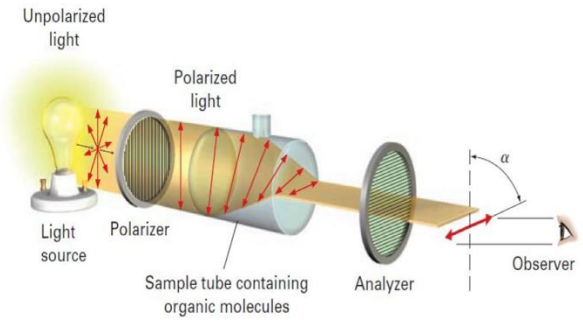


Optically Active Compounds

When a beam of plane polarized light passes through a solution of certain organic molecules, such as sugar or camphor, the plane of polarization is rotated through an angle α. Not all organic substances exhibit this property, but those that do are said to be optically active.

The angle of rotation can be measured with an instrument called a polarimeter. When a solution of known concentration of an optically active material is placed in the polarimeter, the beam of light is rotated either to the right (clockwise) or to the left (anti-clockwise). So the compounds which rotate the plane polarized light (PPL) to the right (clockwise) is said to be Dextrorotatory, and those which rotate the PPL to the left is said to be Levorotatory. Dextrorotatory is indicated by + sign, while Levorotatory by a minus sign (—)

(—)-Morphine, for example, is levorotatory, and (+)-sucrose is dextrorotatory.



Specific Rotation

The extent of rotation depends on the number of optically active molecules encountered by the light beam. This number, in turn, depends on sample concentration and sample path length. If the concentration of sample is doubled, the observed rotation doubles. If the concentration is kept constant but the length of the sample tube is doubled, the observed rotation doubles. In addition, the angle of rotation depends on the wavelength of the light used.

To express optical rotations in a meaningful way so that comparisons can be made, we have to choose standard conditions. The specific rotation, $[\alpha]_D$, of a compound is defined

as "the observed rotation when light of 589.6 nanometer (nm; 1 nm = 10^{-9} m) wavelength is used with a sample path length l of 1 decimeter (dm; 1 dm = 10 cm) and a sample

concentration c of 1 g/cm3".

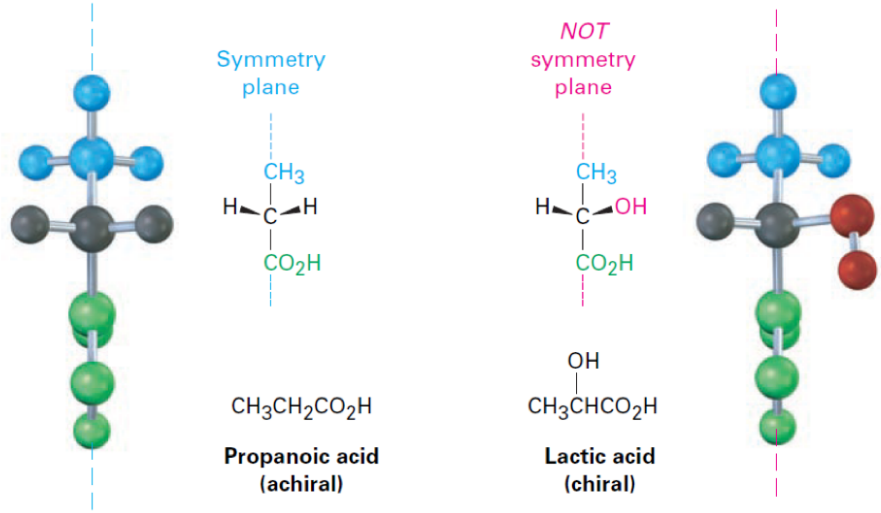
$$[\alpha]_D = \frac{\text{Observed rotation (degrees)}}{\text{Pathlength, } l \text{ (dm)} \times \text{ Concentration, } c \text{ (g/cm}^3\text{)}} = \frac{\alpha}{l \times c}$$

Since the (±) acid is only a mixture of (+) and (-)-forms, in reality lactic acid exist in two forms, the (+)-lactic acid and the (—)-lactic acid. These two acids are exactly identical in physical and chemical properties but differ in their action on the plane polarized light.

So such forms of the same compound which differ only in their optical properties are called Optical isomers and the phenomenon is termed Optical isomerism
An optically active compounds exists in two isomeric forms that rotate the plane polarized light in opposite directions. The optical rotatory power of two isomers are equal in magnitude but opposite in direction.

Plane of Symmetry

A plane which divides an object to two equal halves is said to be plane of symmetry. For example, coffee cup has a plane of symmetry in comparison with a person's hand or shoes which lack a plane of symmetry. An object lacking a plane of symmetry is called Chiral or Dissymmetric, while those having a plane of symmetry is referred to as Achiral.



Chiral Carbon Atom

A carbon atom which is bonded to four different groups. It is also termed as handedness. Chiral carbon lacks the plane of symmetry and therefore called as dissymmetric or Asymmetric.

A chiral object cannot be superimposed on its mirror image. For example, a left hand does not posses a plane of symmetry, and its mirror image is not another left hand but a right hand. The two are not identical and cannot be superimposed. If we lay one hand on the other, the fingers and the thumbs would clash.

Optical Isomerism

An optically active compounds exists in two isomeric forms that rotate the plane polarized light in opposite directions. They are called optical isomers and the phenomena is called optical isomerism.

The optical rotatory power of two isomers are equal in magnitude but opposite in direction. The equimolar mixture of two isomers will therefore, not rotate the PPL at all and is said to be Racemic Mixture.

Optical isomers have the same physical properties: Melting point, boiling point, density etc. They have the same specific rotations but with opposite signs.

Optical Isomerism of Lactic acid

It contains one chiral carbon

Three forms of lactic acid are known, of which two are optically active and the third one is optically inactive.

(+)-Lactic Acid: Rotate the PPL to the right (clockwise) and is dextrorotatory (-)-Lactic Acid: Rotate the PPL to the left (anti-clockwise) and is

levorotatory.

(±)-Lactic Acid: Does not rotate PPL and is optically inactive because it is a racemic mixture of (+) and (-) forms

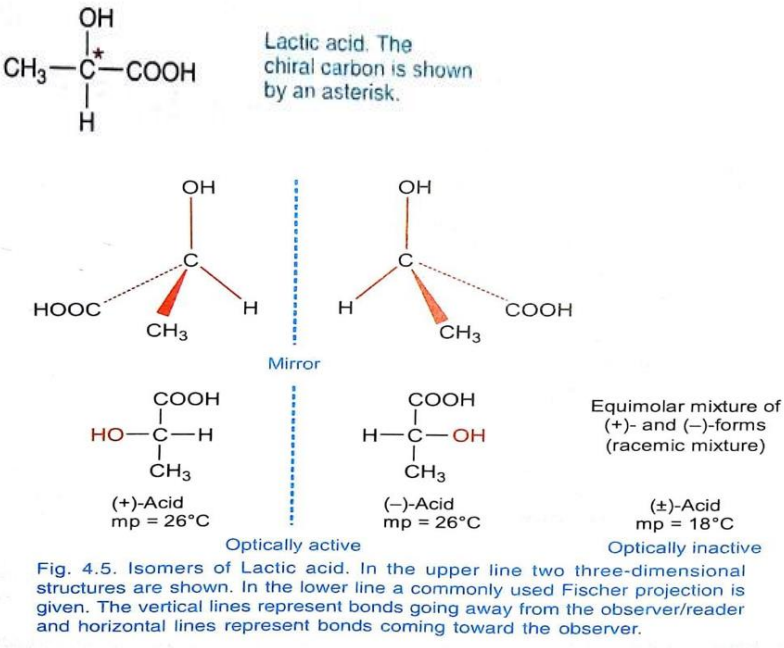


Fig. 4.5. Isomers of Lactic acid. In the upper line two three-dimensional structures are shown. In the lower line a commonly used Fischer projection is given. The vertical lines represent bonds going away from the observer/reader and horizontal lines represent bonds coming toward the observer.

Conditions for Optical Isomerism

Molecule should be dissymmetric. That is, the molecule should not be superimposed on its mirror image. Simply, this dissymmetry results from the presence of a chiral carbon atom. Chiral carbon is the one, which is bonded to four different groups.

The non-superimposable mirror image forms of a chiral carbon are called Enantiomers. They represent two optical isomers (+), and (-). Their opposite rotatory powers are due to the opposite arrangements of groups around a chiral carbon.

Note: It is true that molecules containing chiral carbons are optically active, but it is not always so.

There are some compounds such as meso-tartaric acid which have chiral carbon but is not optically active.

On the other hand, there are some compounds which do not have chiral carbons but are optically active i.e., substituted allenes and biphenyls

OPTICAL ISOMERISM OF TARTARIC ACID

Tartaric acid contains two chiral carbon atoms

Four forms of tartaric acid are known, of which two are optically active and two are optically inactive. The two optically active forms are mirror images of each other but not superimposable, that is, they are Enantiomers.

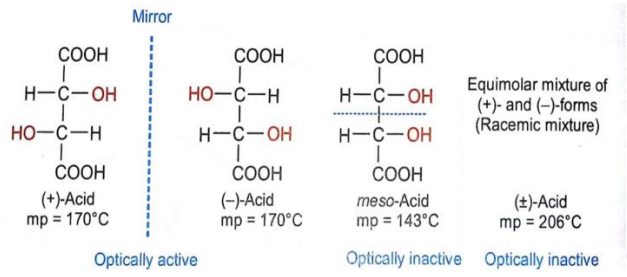
Four forms of Tartaric acid

(+)-Tartaric Acid: Rotate the PPL to the right (clockwise) and is dextrorotatory

(-)-Tartaric Acid: Rotate the PPL to the left (anti-clockwise) and is levorotatory.

meso-Tartaric Acid: It possesses a plane of symmetry and is optically inactive. This optically inactive form is said to be internally compensated; means optical rotation of one chiral carbon is cancelled by that of the other.

(±)-Tartaric Acid: Does not rotate PPL and is optically inactive because it is a racemic mixture of (+) and (-) forms



Terms used to describe Optical Isomerism

Achiral molecule: Which can not be superimposed on its mirror image. It is identical to its mirror image and do not rotate the Plane polarized light.

Chiral center: For a molecule to be chiral, it must have at least one chiral center- a carbon with four nonequivalent groups. Any molecule that contain a chiral center will be chiral unless the molecule has a plane of symmetry, in which case the molecule is achiral (meso)

Chiral molecule: A chiral molecule is one that can not be superimposed on its mirror image. It rotate the PPL

Diastereomers: Diastereomers are stereoisomers that are not mirror images of each other. For a molecule to have a diastereomer, it must contain more than on chiral center.

Enantiomers: Mirror images of each other are enantiomers

Meso compounds: Compounds that contain a chiral center but is achiral because of plane of symmetry

Optically active: Compounds having the ability to rotate the PPL

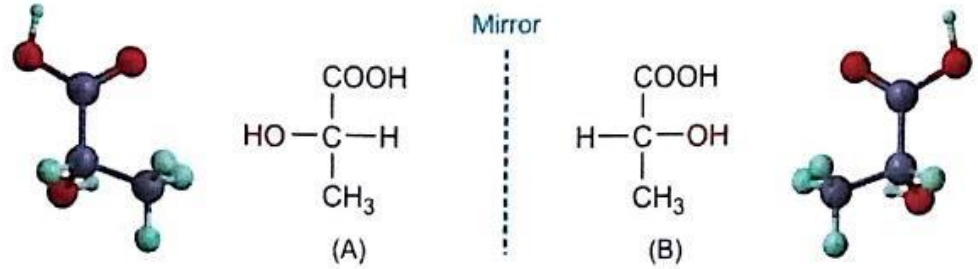
Racemic mixture: A racemic mixture is a 50:50 mixture of two enantiomers. Racemic mixture do not rotate PPL

PROPERTIES OF ENANTIOMERS

Optical isomers that are mirror images are called enantiomers.

These always exist as discrete pairs. For example, there are two optical isomers of lactic acid.

(A) Is the mirror image of (B) and are a pair of enantiomers



Enantiomers are stable, isolable compounds that differ from one another in 3-Dimentional spatial arrangements.

Enantiomers cannot be interconverted under ordinary conditions

Enantiomers have identical properties in all respect except in their interaction with plane polarized light.

They have same melting point, density, solubility, color, and reactivity towards acids and bases.

They differ, however, in the direction in which they rotate the PPL. Both rotate the PPL to exactly the same extent (same angle) but one rotates the plane to the right and other to the left.

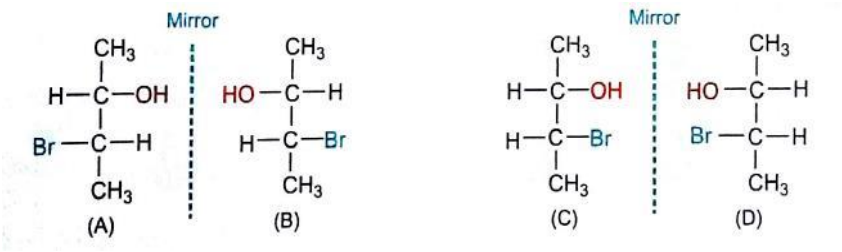
A mixture of equal amounts of two enantiomers is a racemic mixture. Such a mixture is optically inactive because the two components rotate the PPL equally in opposite directions and cancel one other.

PROPERTIES OF DIASTEREOMERS

Diastereomers are stereoisomers that are not mirror images of each other. For a molecule to have a diastereomer, it must typically contain a chiral center.

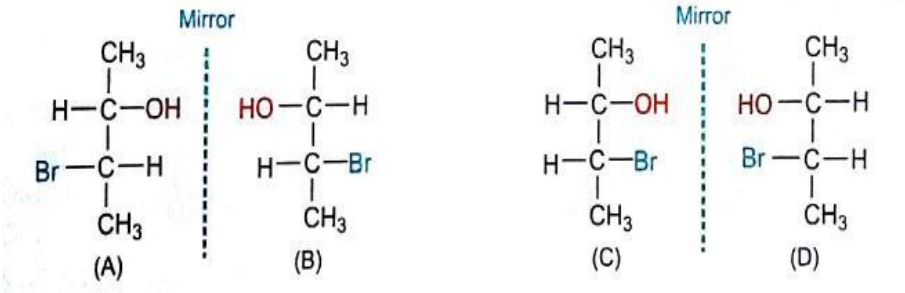
In general, each chiral carbon atom in a molecule doubles the number of theoretically possible isomers. Hence, molecules with n number of chiral carbon atoms have 2^n stereoisomers.

Figure shows the 4 isomers of 3-bromo-2-butanol, which has two chiral carbon atoms



Notice that (A) is the mirror image of (B), (C) is the mirror image of (D). Thus the four isomers are two pairs of enantiomers. Now compare (A) with (C). They are neither superimposable nor are they mirror images. They are called diastereomers. (A) and (D) are also diastereomers, as are (B) and (C), and (B) and (D).

Diastereomers have different properties. Two diastereomers will have different melting points, boiling points, and solubilities. They will also have different reactivities toward most reagents

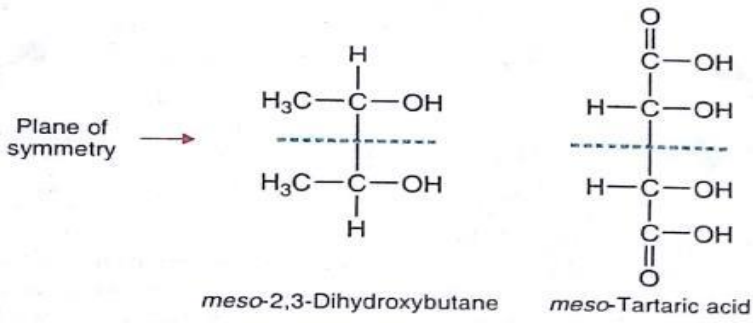


Diastereomers are stereoisomers that are not mirror images. Since we used the right-hand/left-hand analogy to describe the relationship between two enantiomers, we might extend the analogy by saying that the relationship between diastereomers is like that of hands from different people. Your hand and your friend's hand look similar, but they aren't identical and they aren't mirror images. The same is true of diastereomers: they're similar, but they aren't identical and they aren't mirror images.

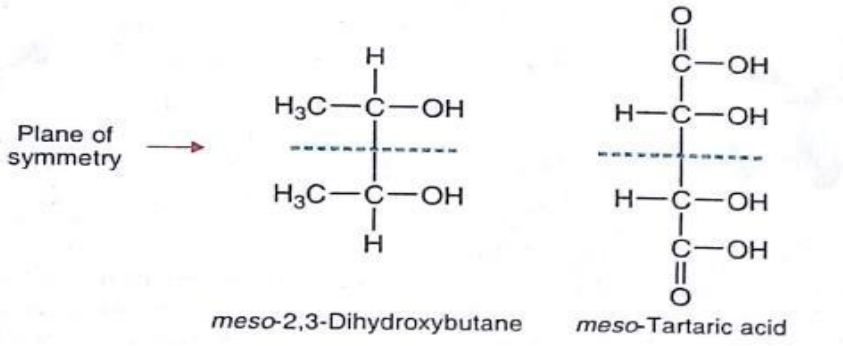
Meso Compounds

A meso compound is a molecule that contains a chiral center, but is achiral as a result of having a plane of symmetry in the molecule.

A compound with two or more chiral carbon atoms but also having a plane of symmetry is called meso compound. The molecules having plane of symmetry dividing them midway between the two chiral carbons in each.



Notice that one half of the molecule is the mirror image of the other. Both molecules are optically inactive, even though each has two chiral centers. Neither will rotate the PPL

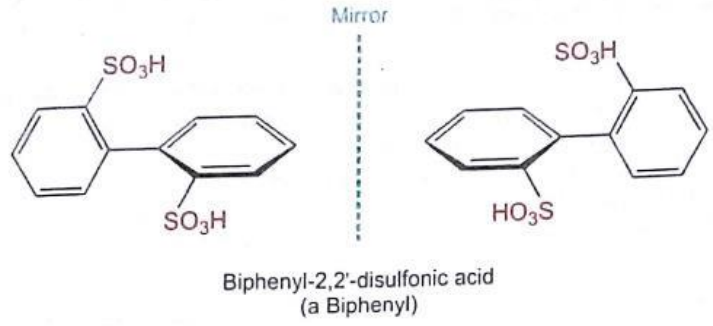


OPTICAL ACTIVITY WITHOUT ASYMMETRIC CARBON

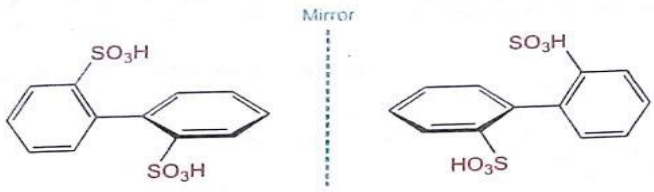
Compounds containing a chiral carbon are optically active. However, there exist some compounds which do not possess a chiral atom but are optically active provided that the molecule is dissymmetric.

BIPHENYL DERIVATIVES

Substituted biphenyls show optical isomerism when substituents in the 2-positions are large enough to prevent rotation about the bond joining the two benzene rings. For example, biphenyl-2,2'-disulphonic acid exist in two forms.



These two forms are non-superimposable mirror images. They do not interconvert at room temperature because the energy required to twist one ring through 180 angle relative to the other is too high. This in turn is because, during the twisting process, the two –SO3H groups must come into very close proximity when the two benzene rings become coplanar and strong repulsive forces are introduced.



Biphenyl-2,2'-disulfonic acid
(a Biphenyl)

RESOLUTION OF RACEMIC MIXTURES

Synthesis of an optically active compound produces a mixture of + and –ve isomers in equal amounts. Recall that these isomers are non-superimposable mirror images and are called enantiomers. Such a mixture is called racemic mixture or a racemate. The separation of a racemic mixture into its two optically active compounds (+ and – isomers) is known as Resolution.

If the enantiomers are separated, the mixture is said to have been resolved.

MECHANICAL SEPARATION

This method is developed by L. Pasteur in 1848. It is applicable to only solid substances which form well defined crystals.
Since crystals of the two forms differ in their shapes, these can be separated mechanically with the help of a magnifying glass and tweezers (forceps). This method

is too tedious for practical purposes and is now of historical interest only because it was the first method which Pasteur employed for the separation of the tartaric acids.

CHEMICAL RESOLUTION

This method is developed by L. Pasteur in 1858. It involves the use of an optically active compound which could react easily and selectively with either of the two enantiomers to produce a mixture of two diasteriomeric compounds. The two compounds so formed have different physical properties and therefore can be separated by conventional method of separation e.g., crystallization. The crystals of diastereoisomers so obtained can be broken down chemically to obtain the two enantiomers separately.

Separation of racemates into their component enantiomers is a process called resolution.

Since enantiomers have identical physical properties, such as solubility and melting point, therefore, resolution is extremely difficult.

Diastereomers, on the other hand, have different physical properties, and this fact is used to achieve resolution of racemates. Reaction of a racemate with an enantiomerically pure chiral reagent gives a mixture of diastereomers, which can be separated.

For example, a solution of racemic lactic acid may be treated with an optically active base such as the alkaloid (Brucine). The resulting product will consist of two salts

(i) (+)- Acid. (—)- Base; and (ii) (—)- Acid(—)-Base

Suppose the two enantiomorphic forms of lactic acid are represented by the symbols

BIOCHEMICAL RESOLUTION

This method is developed by L. Pasteur in 1858. In this method, one of the enantiomers is destroyed biochemically, using a suitable microorganism such as yeast, mold or bacterium. The microorganism utilizes one of the enantiomers for its growth and leaves the other in the solution. The enantiomer left in the solution can be isolated by fractional crystallization.

For example: Penicilium glaucum removes Dextro form from the racemic mixture of ammonium tartrate leaving a levo ammonium tartrate

Disadvantages of biochemical resolution

Half of the material is lost.

This method can not be used if the mixture is toxic to the microorganisms.

Kinetic Method

The fact that enantiomers react with an optically active substance at different rates, is used for the separation of racemic mixtures. The procedure takes advantage of differences in reaction rates of enantiomers with chiral reagents. One enantiomer may react more rapidly, thereby leaving an excess of the other enantiomer behind.

A disadvantage of resolutions of this type is that the more reactive enantiomer usually is not recoverable from the reaction mixture.

Selective Adsorption/Chiral chromatography

Sometimes 'resolution' may be achieved by passing a solution of the racemate over a column of a finely powdered, optically active adsorbent such as starch, sugar or quartz. The surface of the adsorbent, adsorbs selectively one enantiomer and thus the solution emerging at the bottom is richer in the other enantiomer.

Chromatographic methods, whereby the stationary phase is a chiral reagent that adsorbs one enantiomer more strongly than the other, have been used to resolve racemic compounds.

Importance of Resolution

Majority of the drugs have many chiral centers. For example vitamin E has three chiral centers, therefore, 8 forms are possible. Among these 8 forms only one occurs naturally and is 100% potent. Other 7 forms have different potencies less than naturally occurring form. All forms of vitamin E has a positive impact but there are some cases where the other forms have a negative impact on the body are harmful. i.e., one enantiomer is biologically active while the other is either inactive or very harmful.

The Thalidomide Story

Thalidomide was launched on October 1, 1957 as an anti-emetic drug that had an inhibitory effect on morning sickness, that's why given to pregnant womens' to relieve morning sickness. It was later realized that while the (+)-form of the molecule was a safe and effective anti- emetic, the (−)-form was an active teratogen. The drug caused numerous birth abnormalities when taken in the early stages of pregnancy because it contained a mixture of the two forms. Majority of the born kids were having no legs, arms, fingers. This happens because the

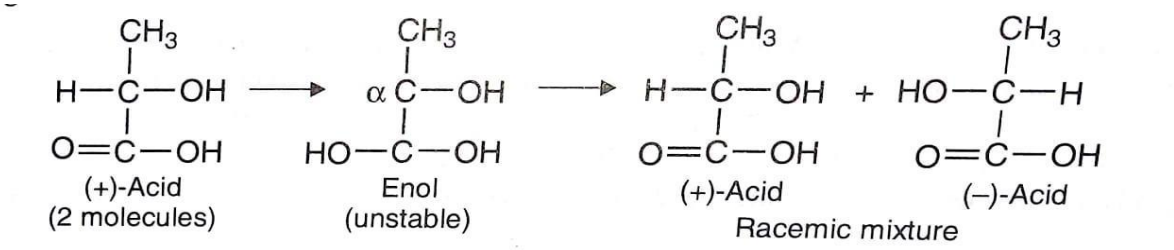
Pharmaceutical company was unable to resolve the + thalidomide

Single Enantiomer vs. Racemic Mixtures

Single enantiomer have less complex and more selective pharmacodyanamic profile as compared to racemic mixture, so have lesser adverse drug reactions, improved therapeutic profile, less chances of drug interactions than racemic mixtures. Single enantiomers seem to be more advantageous over racemic mixtures as - adverse drug reactions occurring due to one enantiomers are avoided, patients are exposed to less amount of drug so body is exposed to the lesser metabolic, renal and hepatic load of drug, there is easier therapeutic drug monitoring of the active pure active enantiomers.

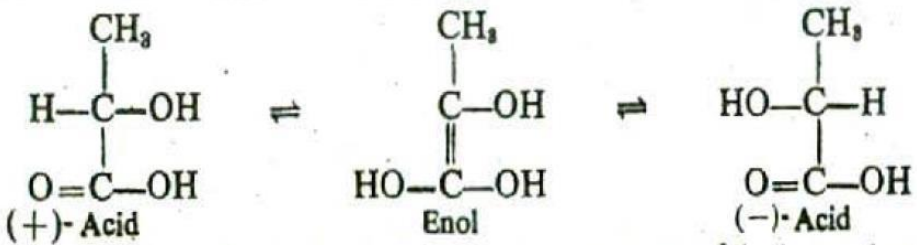
Racemization

The conversion of an optically active compound (+)- or (-)- into racemic mixture (±) is known as Racemization.
Racemization can be accomplished by the means of heat, light or by the conversion of an optically active isomers into an optically inactive intermediate which then reverts to the racemic mixture. The conversion of either of the optically active lactic acids into a racemic mixture by heating its aqueous solution may proceed through an enol intermediate.

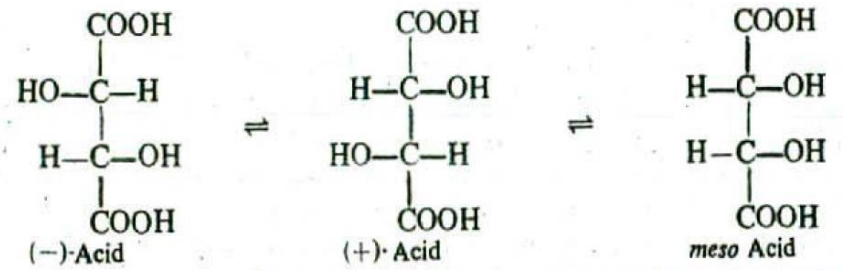


Since the enol is a planar molecule, so the possibility of frontal and rear attachment of H-atom to a carbon atom (in the enol form) is equally probable. Consequently equal number of molecules of (+)- and (-) form would result and racemic mixture would be produced. These changes are reversible and in actual practice we get an equilibrium mixture.

The equilibrium mixture thus obtained contains one molecule of (+)- Acid and one molecule of (-)- Acid i.e., the racemic mixture of the acid.



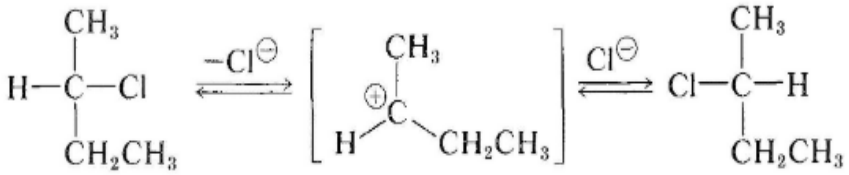
According to Pasteur, if a solution of (+)-tartaric acid is heated for some time at 165°C, it loses its optical activity forming the racemic mixture and meso acid. This is due to the fact that tartaric acid having two asymmetric carbon atoms may undergo inversion of H and OH only about one carbon or both, yielding meso or racemic form, respectively.



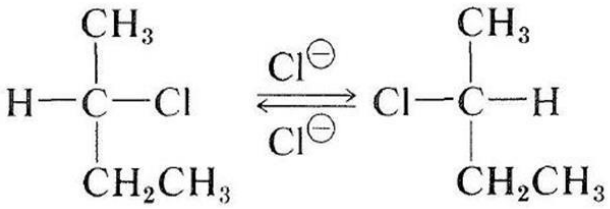
Optically active substances which cannot enolize because they have no hydrogen atom at a carbon to a carbonyl group, do not in general racemize except under conditions which bring about other chemical changes as well.

Racemization of Optically Active Halides

The racemization of optically active halides may take place either by an SN1 or SN2 mechanism depending upon the experimental conditions. In polar solvents, S-butyl chloride racemizes by SN1 mechanism. Dissociation of the compound produces planar carbonium ion which can recombine with the anion i.e., Cl yielding both S and R forms of the compound.



Optically active halides also can be racemized by an SN2 mechanism. A solution of active 2-chlorobutane in acetone containing dissolved lithium chloride becomes racemic. Displacement of chloride ion of the halide by chloride ion (from LiCl) inverts configuration at the carbon atom undergoing substitution. A second such substitution regenerates the original enantiomer. Eventually, this back and forth process produces equal number of R and S forms; the substance is then racemic.

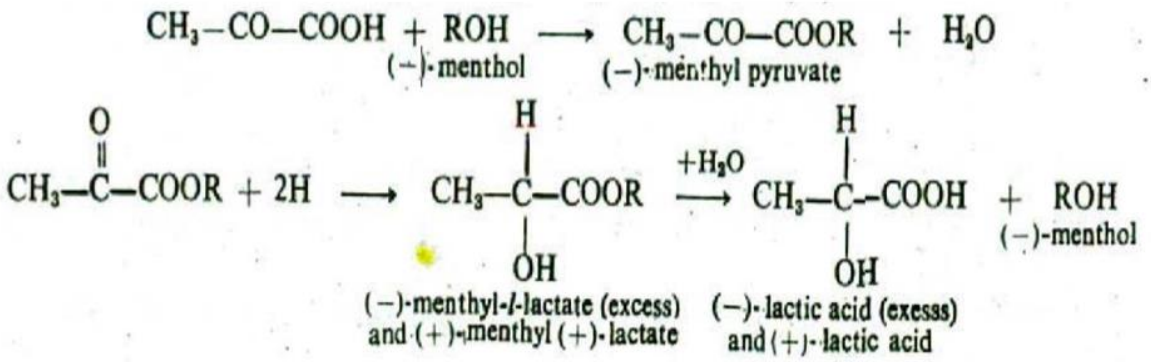


Asymmetric Synthesis

The process in which an asymmetric compound is synthesized from a symmetric compound to yield the (+)- or (-)- isomer directly, is termed Asymmetric Synthesis. OR

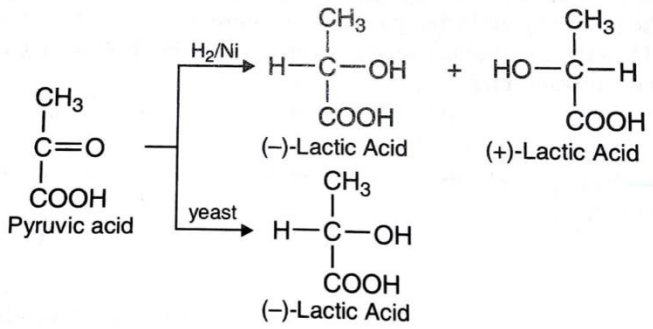
The process in which a chiral compound is synthesized from an achiral compound to yield the (+)- or (-)- isomer directly.

We have already seen that when a compound containing an asymmetric carbon atom is synthesized by ordinary laboratory methods from a symmetric compound, the product is a racemic mixture. If, however, such a synthesis is carried under the 'Asymmetric influence of a suitable optically active reagent, one of the optically active isomers, (+)-or(—)-, is produced in preference and in excess.



When pyruvic acid is reduced as such, it yields (±)- lactic acid. However, when pyruvic acid is first combined with an optically active alcohol, ROH, such as (—)-menthol* to form an ester which is then reduced, the product upon hydrolysis yields (—). lactic acid in excess.

In nature, numerous optically active substances such as terpenes, alkaloids and proteins are produced by asymmetric synthesis, under the influence of optically active enzymes. These enzymes unite with the substance available in plants and when the synthesis is complete, they separate from the product and are thus again free to combine with more of the parent inactive substance and the endless process continues.

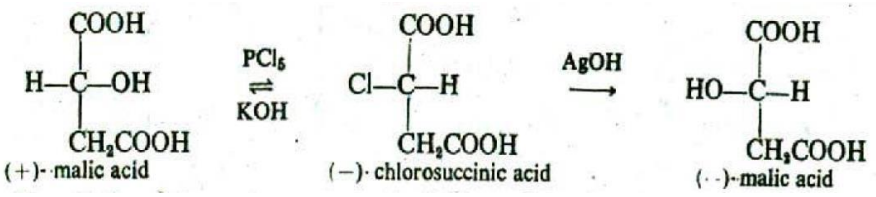


Walden Inversion

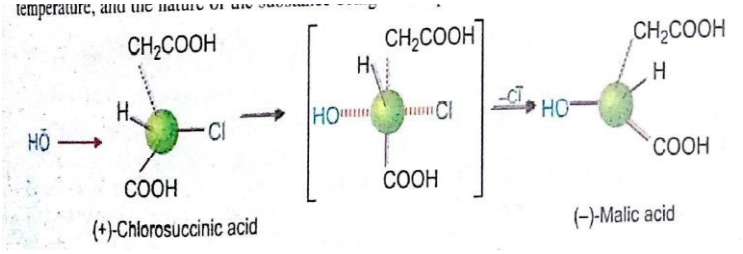
When an atom or group directly linked to chiral carbon atom is replaced, the reaction may proceed with inversion of configuration. This phenomenon was first of all observed by Walden (1895) and hence the name Walden inversion. Walden inversion may also be defined as the conversion of the (+)-form to (—)-form or vice versa. Thus (+)-malic acid may be converted to (—).malic acid as follows.

Walden inversion is the inversion of a chiral center in a molecule in a chemical reaction. Since a molecule can form two enantiomers around a chiral center, the Walden inversion converts the configuration of the molecule from one enantiomeric form to the other.

Thus (+)-malic acid may be converted to (—)-malic acid as follows.



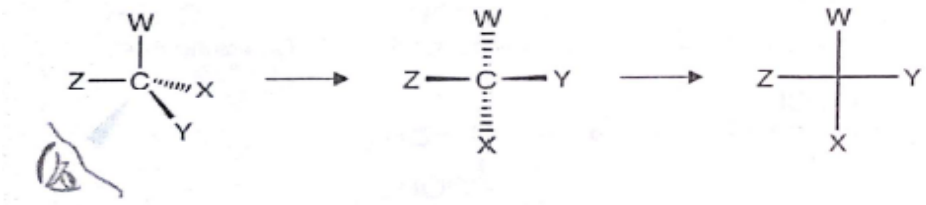
Since SN2 reactions always proceed with inversion of configuration, Walden inversion is a type of SN2 reaction



FISCHER PROJECTION

When we attempt to depict configurations, we face the problem of representing three dimensional structures on a two dimensional surface. To overcome this difficulty we use the so-called Fischer projection. It is the most convenient way of viewing molecules with more than one chiral centers. Fischer projection is a convenient 2-D drawing that represents a 3-D molecule.

To make a Fischer projection, you view a chiral center so that two substituents are coming out of the plane at you (to hug you), and the two substituents are going back into the plane. Then the chiral center becomes a cross on the fischer projection. Every cross in Fischer Projection is a chiral center.

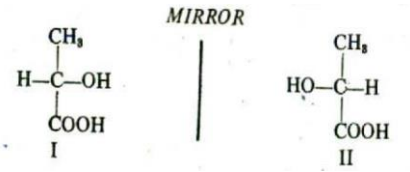


This is the structure of an asymmetric carbon atom drawn in a prescribed orientation and then projected into a planar surface.

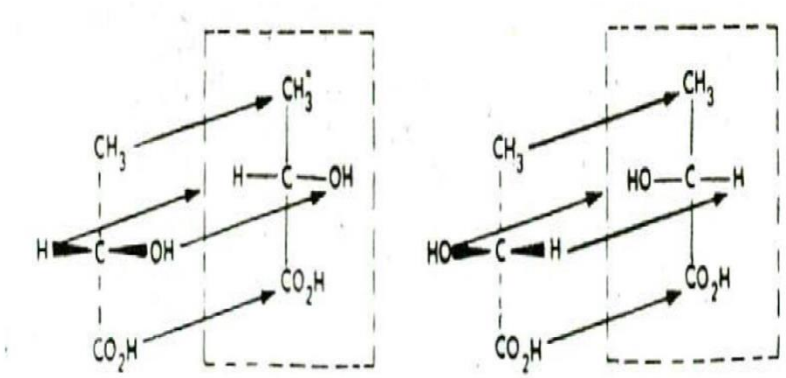
Thus planar formulas of the asymmetric carbon are obtained by placing it so that the two substituents are horizontal and project out towards the viewer (shown by thick wedge-like bonds), while the two other substituents are vertical and project away from the viewer (shown by dotted bonds).

Remember in Fischer projection molecule is oriented so that the vertical bonds at chirality center are directed away from you and horizontal are directed towards you

Planar representation of two forms of lactic acid may be given as



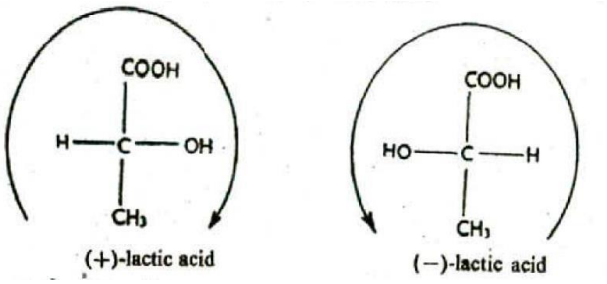
In these formulas the horizontal bonds i.e., C—OH and C—H project towards us out of the plane of the paper whereas the vertical bonds i.e., C—COOH and C— CH3 project away from us. Inspection of the models shows that one interchange of a pair of substituents inverts the configuration (changes one enantiomer into) its minor image), whereas an even number of such interchanges does not. Thus interchanging of —H for —OH in I gives the enantiomer II while the interchange of CH3 for —COOH and —H for —OH leaves the configuration unchanged.



ASSIGNING CONFIGURATIONS TO CHIRAL MOLECULES

Absolute and Relative Configuration

The actual 3-D arrangement of groups in a chiral molecule is called absolute configuration. While discussing optical isomerism, we must distinguish between relative and absolute configuration (arrangement of atoms or groups) about the asymmetric carbon atom. Let us consider a pair of enantiomers, say (+)- and (-)- lactic acids.



We know that they differ from one another in the direction in which they rotate the plane of polarized light. In other words, we know their relative configuration in the sense that one is of opposite configuration to the other, But we have no knowledge of the absolute configuration of the either isomer. That is, we cannot tell as to which of the two possible configurations corresponds to (+)acid and which to the (—)-acid.

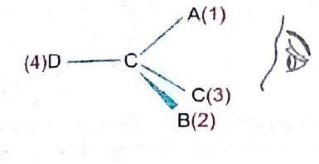
R-S System

The actual 3-dimentional arrangement of groups in a chiral molecule is called absolute configuration. We can specify the configuration by using the R-S system.
This is a newer and more systematic method of specifying absolute configuration to optically active compounds. Since it has been proposed by R.S. Cahn, C.K. Ingold and V. Prelog, therefore known as Cahn-Ingold Prelog system. This system of designating configuration has been used increasingly since the early 1960 and may eventually replace the DL-system.

In this system, four groups attached to the chiral carbon are arranged in decreasing order of priority (1,2,3,4) by applying priority rules. We then view the chiral carbon with the lowest priority group (4) on the side opposite the observer.

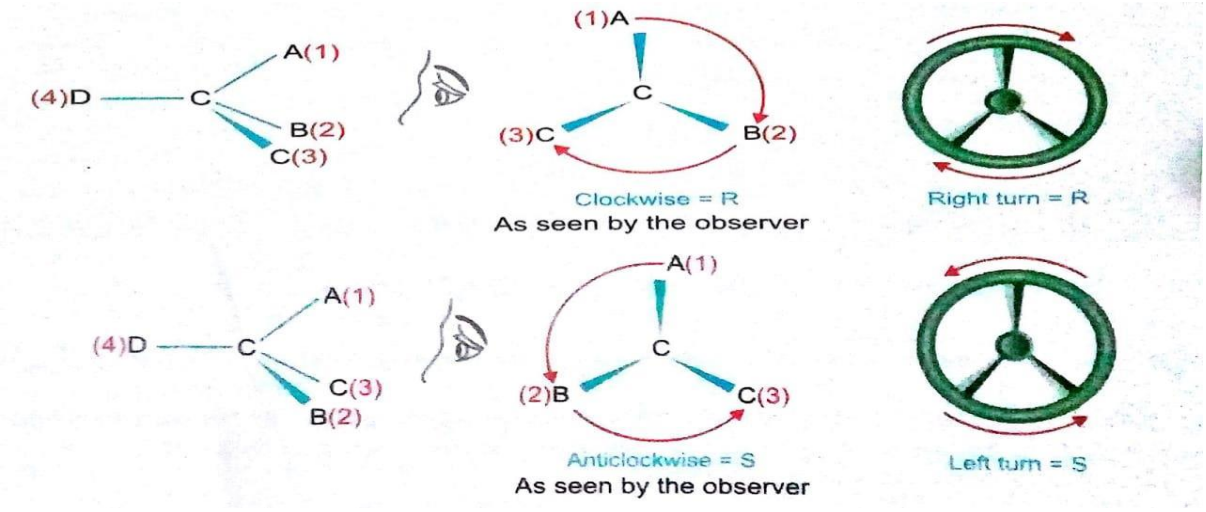
In the above example, A has the highest priority followed by B and C, while D has the lowest priority (4).

(i) If the eye while moving from 1-2-3, travels in a clockwise or right-hand direction, the

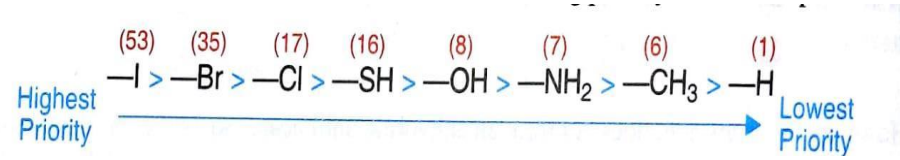


configuration is designated R (Latin, Rectus—right).

(ii) If the eye while moving from 1-2-3 travels in counterclockwise or left- hand direction, the configuration is designated S (Latin, Sinister= left).

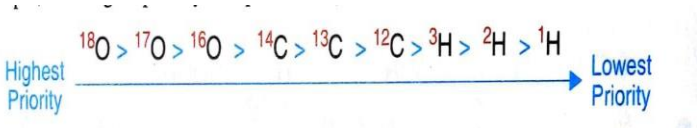


RULE 1: Look at the four atoms directly attached to the chirality center, and rank them according to atomic number. The atom with the highest atomic number has the highest ranking (first), and the atom with the lowest atomic number (usually hydrogen) has the lowest ranking (fourth).



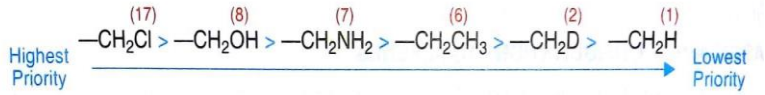
RULE 2:

For isotopes, the higher the atomic mass the higher the priority. For example, deuterium (Hydrogen-2) has higher priority than protium (Hydrogen-1)

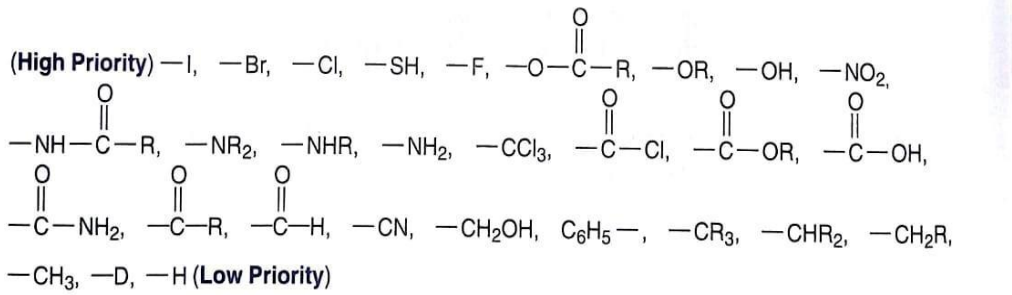


RULE 3: If a decision can't be reached by ranking the first atoms in the substituent, look at the second, third, or fourth atoms away from the chirality center until the first difference is found.

RULE 4: Multiple-bonded atoms are equivalent to the same number of single bonded atoms.

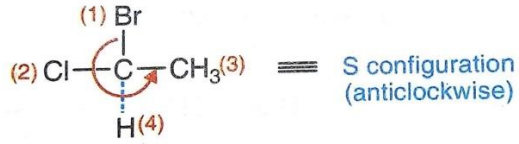


Priority sequence of common groups and atoms



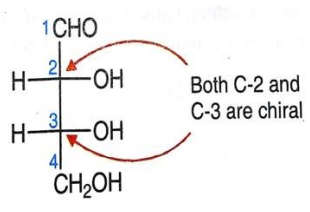
Specify the direction of decreasing priority of the three groups (1- 3). Ignore the lowest priority group that is H. If the groups (1,2,and 3) are arranged in clockwise fashion, the configuration is R. If the groups occurs in anticlockwise fashion, the configuration is S.

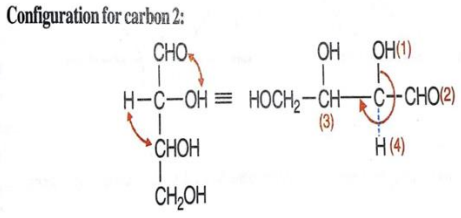
In the above example, configuration is S because groups (1,2 and 3) are arranged in anticlockwise fashion.

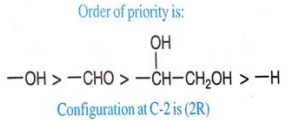


Configuration of compounds with more than 1 chiral center

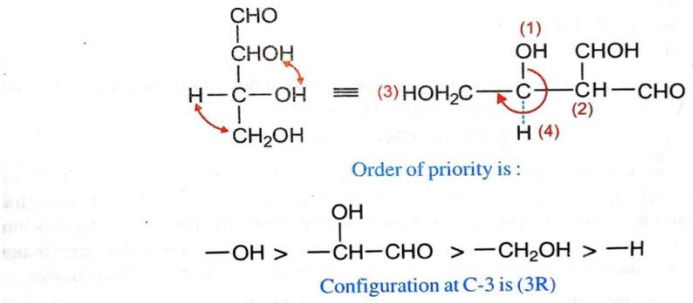
The configuration of compounds with more than one chiral center can also be specified by the R-S system. The configuration of each chiral carbon is determined individually, using the same rules as for compounds with one chiral center.



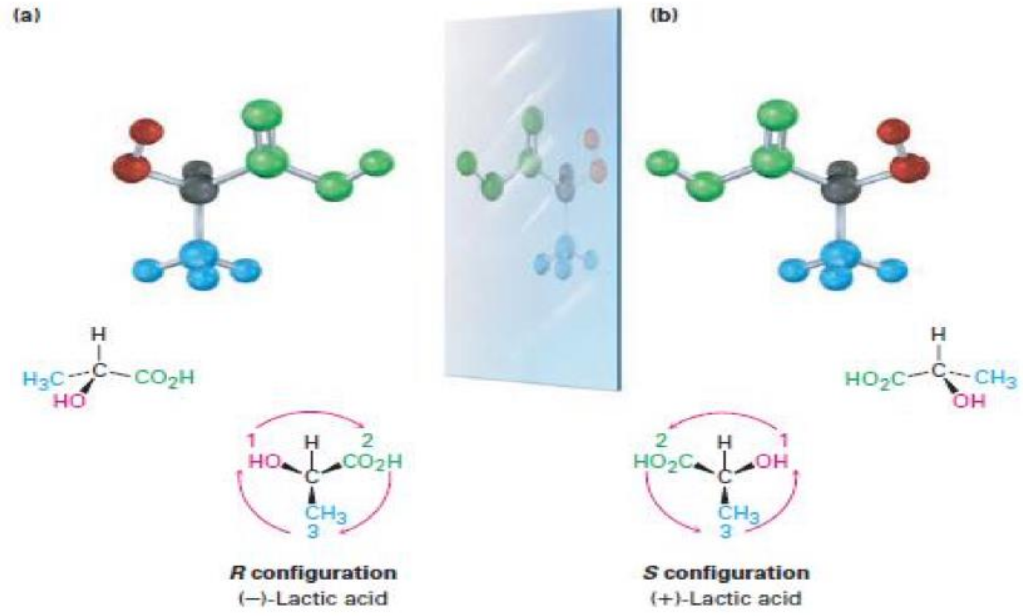




Configuration for carbon 3:

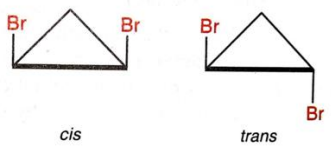


The name of compound is (2R,3R)-2,3,4-trihydroxybutanal

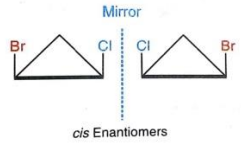


Look at (-)-lactic acid for an example of how to assign configuration. Sequence rule 1 says that OH is ranked 1 and H is ranked 4, but it doesn't allow us to distinguish between CH3 and CO2H because both groups have carbon as their first atom.

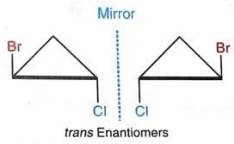
Sequence rule 2, however, says that CO2H ranks higher than CH3 because O (the highest second atom in CO2H) out ranks H (the highest second atom in CH3). Now, turn the molecule so that the fourth-ranked group (H) is oriented toward the rear, away from the observer. Since a curved arrow from 1 (OH) to 2 (CO2H) to 3 (CH3) is clockwise (right turn of the steering wheel), (-)-lactic acid has the R configuration. Applying the same procedure to (+)-lactic acid leads to the opposite assignment.



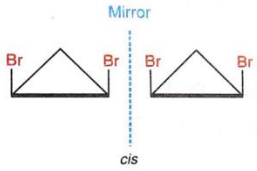
OPTICAL ISOMERISM IN CYCLIC COMPO



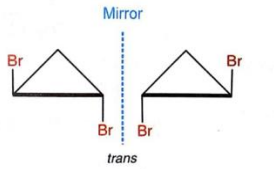
cis Enantiomers



trans Enantiomers



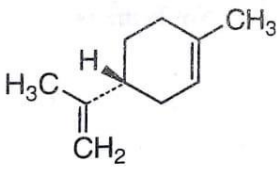
cis
Superimposable mirror images, *meso*



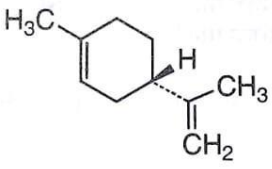
trans
Nonsuperimposable mirror images, *enantiomers*

OPTICAL ISOMERISM IN NATURE

The (+)-limonene has the odor of oranges while (-)-limonene has the odor of lemons

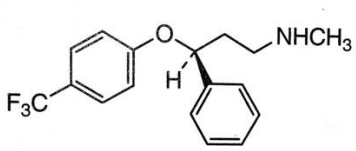


(+)-Limonene



(–)-Limonene

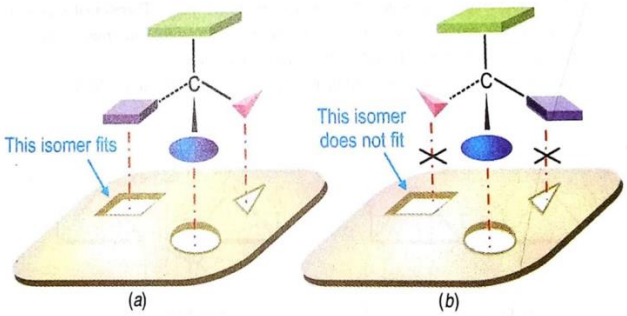
Racemic fluoxetine is an effective antidepressant but it has no activity against migraine. The pure S enantiomer, however, works remarkably well in preventing migraine



(S)-Fluoxetine
(Prevents migraine)

Why do different stereoisomers have different biological activity???

To exert its biological action, a chiral molecule must fit into a chiral receptor at a target site, much as a hand fits into a glove. But just as a right hand can fit only into a right-hand glove, so a particular stereoisomer can fit only into a receptor having the proper complementary shape. Any other stereoisomer will be a misfit like a right hand in a left-handed glove.



Spectroscopy methods

Absorption spectroscopy is the measurement of the amount of light absorbed by a compound as a function of the wavelength of light. In general, a spectrometer irradiates the sample with light, measures the amount of light transmitted as a function of wavelength, and plots the results on a graph. Unlike chemical tests, most spectroscopic techniques are *nondestructive*; that is, the sample is not destroyed. Many different kinds of spectra can be measured with little or no loss of sample.

Infrared (IR) spectroscopy, observes the vibrations of bonds and provides evidence of the functional groups present.

Mass spectrometry (MS), is not a *spectroscopic* technique, because it does not measure absorption or emission of light. A mass spectrometer bombards molecules with electrons and breaks the molecules into fragments. Analysis of the masses of the fragments gives the molecular weight, possibly the molecular formula, and clues to the structure and functional groups. Less than a milligram of sample is destroyed in this analysis.

Nuclear magnetic resonance (NMR) spectroscopy, observes the chemical environments of the hydrogen atoms or the carbon atoms and provides evidence for the structure of the alkyl groups and clues to the functional groups.

Ultraviolet (UV) spectroscopy, observes electronic transitions and provides information on the electronic bonding in the sample.

These spectroscopic techniques are complementary, and they are most powerful when used together. In many cases, an unknown compound cannot be completely identified from one spectrum without additional information, yet the structure can be determined with confidence using two or more different types of spectra.

Electromagnetic waves travel as **photons,** which are massless packets of energy. The energy of a photon is proportional to its frequency and inversely proportional to its wavelength.

Frequency, represented by the Greek letter n (nu), is usually given in hertz (Hz), meaning cycles per second. The **wavelength,** represented by the Greek letter λ (lambda), is the distance between any two peaks (or any two troughs) of the wave. For this reason, we often represent the irradiation of a reaction mixture by the symbol hν.

$E=h\nu$

$E=hc/\lambda$

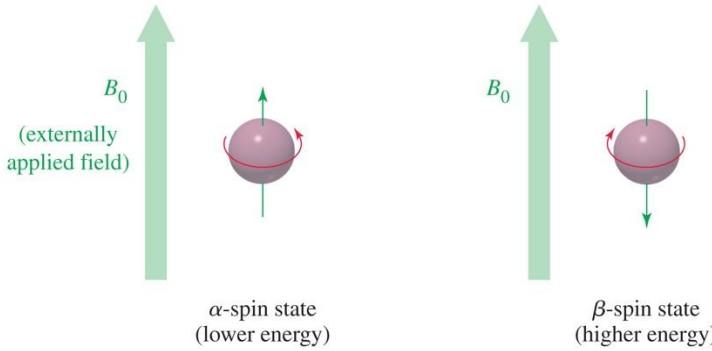
The **electromagnetic spectrum** is the range of all possible frequencies, from zero to infinity. In practice, the spectrum ranges from the very low radio frequencies used to communicate with submarines to the very high frequencies of gamma rays.

The electromagnetic spectrum is continuous, and the exact positions of the divid-ing lines between the different regions are somewhat arbitrary. There are the higher frequencies, shorter wavelengths, and higher energies. Toward the bottom are the lower frequencies, longer wavelengths, and lower energies. X rays (very high energy) are so energetic that they excite electrons past all the energy levels, causing ionization. Energies in the ultraviolet–visible range excite electrons to higher energy levels within molecules. Infrared energies excite molecular vibrations, and microwave energies excite rotations. Radio-wave frequencies (very low energy) excite the nuclear spin transitions observed in NMR spectroscopy.

What is NMR Spectroscopy?

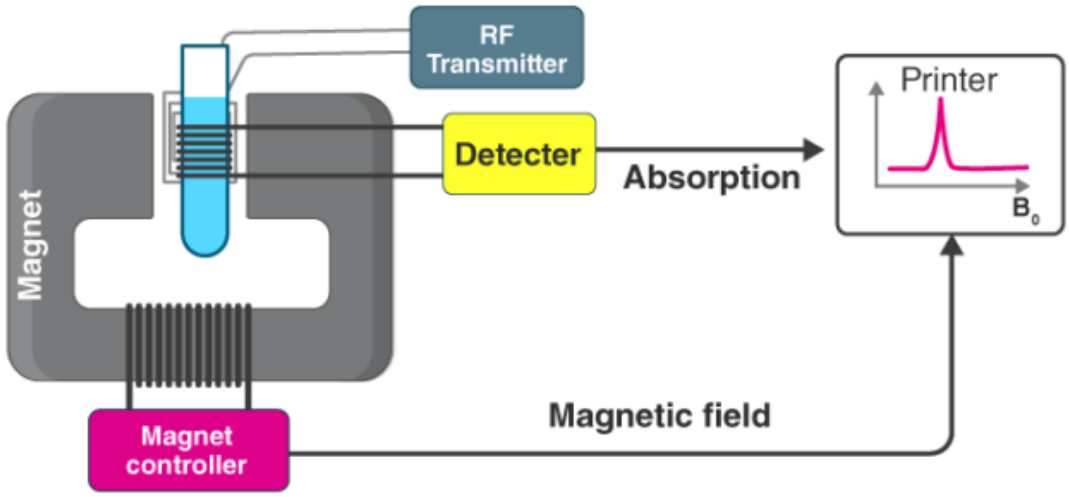
NMR Spectroscopy is abbreviated as ***Nuclear Magnetic Resonance spectroscopy.***

Nuclear magnetic resonance (NMR) spectroscopy is the study of molecules by recording the interaction of radiofrequency (Rf) electromagnetic radiations with the nuclei of molecules placed in a strong magnetic field.



Zeeman first observed the strange behaviour of certain nuclei when subjected to a strong magnetic field at the end of the nineteenth century, but the practical use of the so-called ***"Zeeman effect"*** was only made in the 1950s when NMR spectrometers became commercially available.

It is a research technique that exploits the magnetic properties of certain atomic nuclei. The NMR spectroscopy determines the physical and chemical properties of atoms or molecules.



NMR Spectroscopy Instrumentation

It relies on the phenomenon of nuclear magnetic resonance and provides detailed information about the structure, dynamics, reaction state, and chemical environment of molecules.

Basis of NMR Spectroscopy

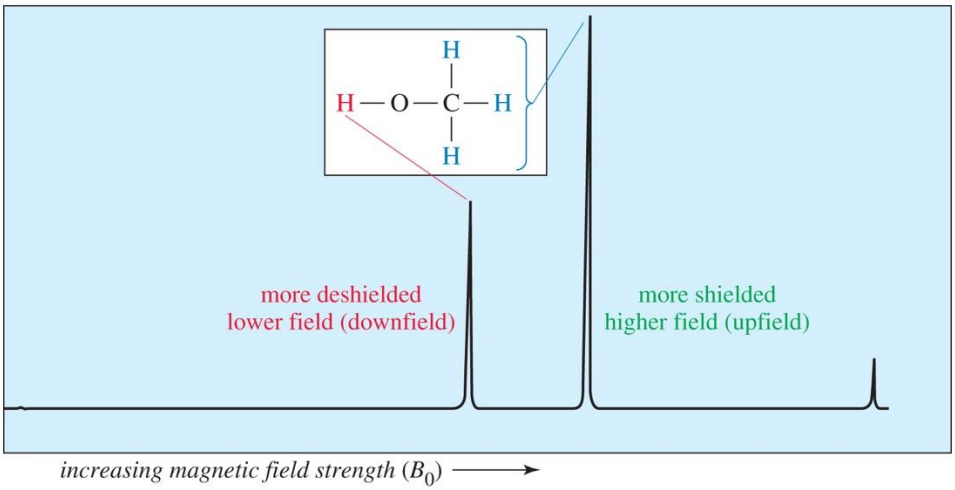
Nuclear Magnetic Resonance (NMR) was first detected experimentally at the end of 1945, nearly concurrently with the work groups Felix Bloch, Stanford University and Edward Purcell, Harvard University. The first NMR spectra was first published in the same issue of the Physical Review in January 1946. Bloch and Purcell were jointly awarded the 1952 Nobel Prize in Physics for their research of Nuclear Magnetic Resonance Spectroscopy. Nuclear magnetic resonance (NMR) spectroscopy is a crucial analytical tool for organic chemists. The research in the organic lab has been significantly improved with the aid of the NMR. Not only can it provide information on the structure of the molecule, it can also determine the content and purity of the sample. Proton (1H)

NMR is one of the most widely used NMR methods by organic chemists. The protons present in the molecule will behave differently depending on the surrounding chemical environment, making it possible to elucidate their structure.

What is shielding and deshielding in NMR?

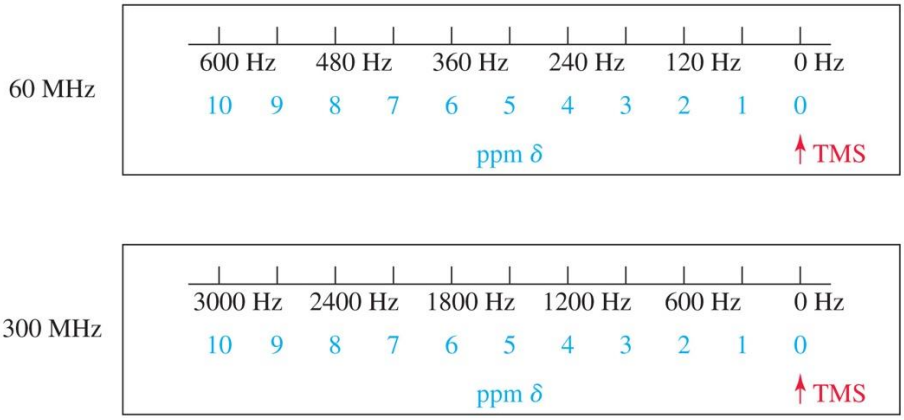
NMR spectroscopy is an ideal technique for identifying the structure of molecules using NMR spectroscopy. Higher electron density around hydrogen atoms creates greater opposition to the applied magnetic field. As a result, the H atom experiences a lower magnetic field and can resonate at a lower frequency. The peak on the NMR spectrum for this H atom would shift upfield. These H atoms are referred to as being shielded.

If the H atom is surrounded by elements that reduce the electron cloud, then, it would experience a higher magnetic field and would resonate at a higher radio frequency. This phenomenon is called de-shielding.

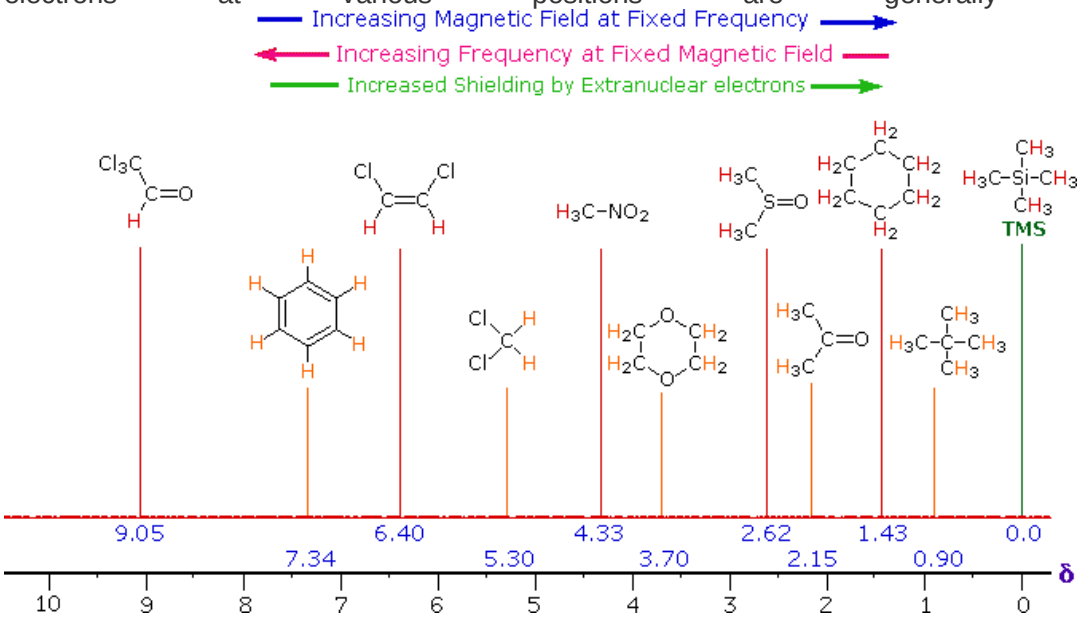


For example, the chemical shift of CH4 protons and CH3Cl protons can be taken here. Chlorine atom is an electronegative atom that will pull the electron density toward it and causes deshielding of the hydrogen nucleus. Therefore, the shift will be to higher ppm. In the case of CH4 the hydrogen nucleus is shielded and therefore, the peak appears on the lower ppm side.

$$\text{chemical shift, ppm } \delta = \frac{\text{shift downfield from TMS (in Hz)}}{\text{spectrometer frequency (in MHz)}}$$



Because of the diverse and complex structures of organic molecules, the shielding effects of electrons at various positions are generally different.

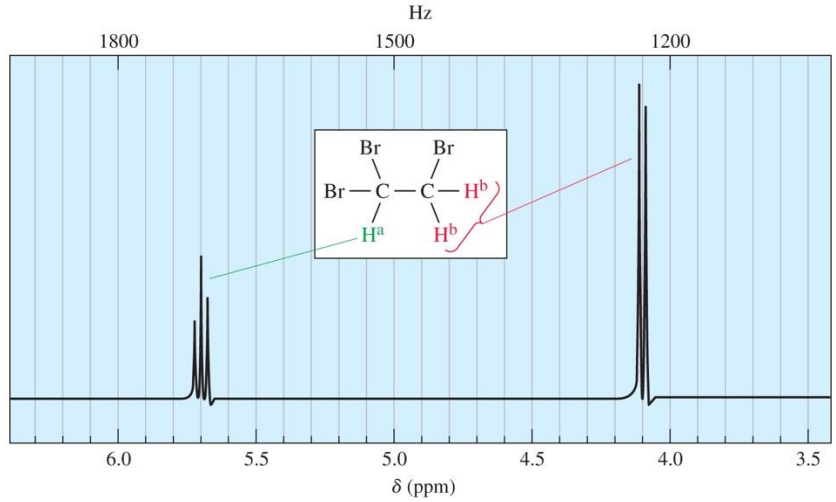


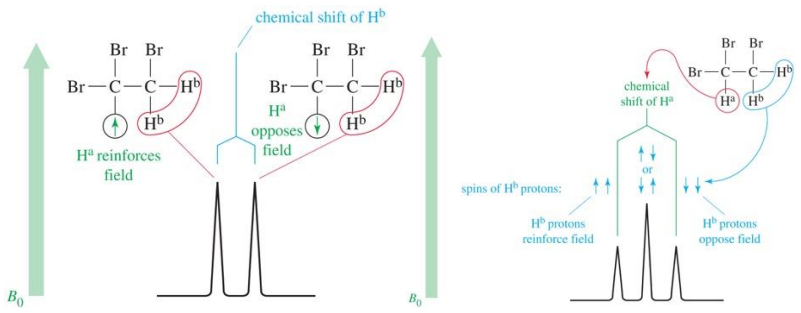
A careful measurement of the field strengths required for resonance of the various protons in a molecule provides us with two important types of information:

1. The number of different absorptions (also called signals or peaks) implies how many different types of protons are present.

2. The amount of shielding shown by these absorptions implies the electronic structure of the molecule close to each type of proton.

Two other aspects of the NMR spectrum we will consider are the intensities of the signals and their splitting patterns:





3. The intensities of the signals imply how many protons of each type are present.

4. The splitting of the signals gives information about other nearby protons.

Protons (in the sample compound) are placed in a magnetic field, where they align either with the field or against it. While still in the magnetic field, the protons are subjected to radiation of a frequency they can absorb by changing the orientation of their magnetic moment relative to the field. If protons were isolated, they would all absorb at the same frequency, proportional to the magnetic field.

But protons in a molecule are partially shielded from the magnetic field, and this shielding depends on each proton's environment. Thus, protons in different environments within a molecule exposed to a constant frequency absorb the radiation at differ- ent magnetic field strengths. The NMR spectrometer was originally developed to vary the magnetic field and plot a graph of energy absorption as a function of the magnetic field strength. Such a graph is called a nuclear magnetic resonance spectrum.

NMR Spectroscopy Principle

- All nuclei are electrically charged and many have spin.
- Transfer of energy is possible from base energy to higher energy levels when an external magnetic field is applied.
- The transfer of energy occurs at a wavelength that coincides with the radio frequency.
- Also, energy is emitted at the same frequency when the spin comes back to its base level.
- Therefore, by measuring the signal which matches this transfer the processing of the NMR spectrum for the concerned nucleus is yield.

NMR Spectroscopy Working

- Place the sample in a magnetic field.
- Excite the nuclei sample into nuclear magnetic resonance with the help of radio waves to produce NMR signals.
- These NMR signals are detected with sensitive radio receivers.
- The resonance frequency of an atom in a molecule is changed by the intramolecular magnetic field surrounding it.
- This gives details of a molecule's individual functional groups and its electronic structure.
- Nuclear magnetic resonance spectroscopy is a conclusive method of identifying monomolecular organic compounds.
- This method provides details of the reaction state, structure, chemical environment and dynamics of a molecule.

Chemical Shift in NMR Spectroscopy

A spinning charge generates a magnetic field that results in a magnetic moment proportional to the spin. In the presence of an external magnetic field, two spin states exist; one spin up and one spin down, where one aligns with the magnetic field and the other opposes it.

Chemical shift is characterized as the difference between the resonant frequency of the spinning protons and the signal of the reference molecule. Nuclear magnetic resonance is one of the most important properties usable for molecular structure determination. There are also different nuclei that can be detected by NMR spectroscopy, 1H (proton), 13C (carbon 13), 15N (nitrogen 15), 19F (fluorine 19), among many more. 1H and 13C are the most widely used. The definition

of 1H as it is very descriptive of the spectroscopy of the NMR. Both the nuts have a good charge and are constantly revolving like a cloud. Through mechanics, we learn that a charge in motion produces a magnetic field. In NMR, when we reach the radio frequency (Rf) radiation nucleus, it causes the nucleus and its magnetic field to turn (or it causes the nuclear magnet to pulse, thus the term NMR).

NMR Spectroscopy Instrumentation

This instrument consists of nine major parts. They are discussed below:

- **Sample holder –** It is a glass tube which is 8.5 cm long and 0.3 cm in diameter.
- **Magnetic coils –** Magnetic coil generates magnetic field whenever current flows through it
- **Permanent magnet –** It helps in providing a homogenous magnetic field at 60 – 100 MHZ
- **Sweep generator –** Modifies the strength of the magnetic field which is already applied.
- **Radiofrequency transmitter –** It produces a powerful but short pulse of the radio waves.
- **Radiofrequency –** It helps in detecting receiver radio frequencies.
- **RF detector –** It helps in determining unabsorbed radio frequencies**.**
- **Recorder –** It records the NMR signals which are received by the RF detector.
- **Readout system –** A computer that records the data.

NMR Spectroscopy Techniques

1. Resonant Frequency

It refers to the energy of the absorption, and the intensity of the signal that is proportional to the strength of the magnetic field. NMR active nuclei absorb electromagnetic radiation at a frequency characteristic of the isotope when placed in a magnetic field.

2. Acquisition of Spectra

Upon excitation of the sample with a radiofrequency pulse, a nuclear magnetic resonance response is obtained. It is a very weak signal and requires sensitive radio receivers to pick up.

NMR Spectroscopy Applications

1. NMR spectroscopy is a spectroscopy technique used by chemists and biochemists to investigate the properties of organic molecules, although it is applicable to any kind of sample that contains nuclei possessing spin.
2. For example, the NMR can quantitatively analyze mixtures containing known compounds. NMR can either be used to match against spectral libraries or to infer the basic structure directly for unknown compounds.
3. Once the basic structure is known, NMR can be used to determine molecular conformation in solutions as well as in studying physical properties at the molecular level such as conformational exchange, phase changes, solubility, and diffusion.

IR Spectroscopy

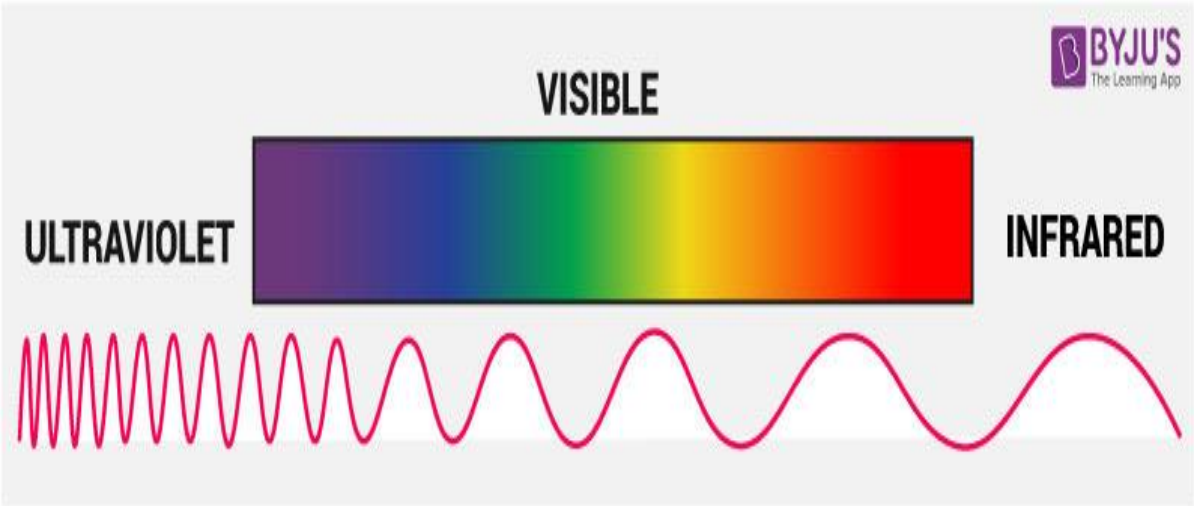
IR spectroscopy (which is short for infrared spectroscopy) deals with the infrared region of the electromagnetic spectrum, i.e. light having a longer wavelength and a lower frequency than visible light. Infrared Spectroscopy generally refers to the analysis of the interaction of a molecule with infrared light.

The IR spectroscopy concept can generally be analyzed in three ways: by measuring reflection, emission, and absorption. The major use of infrared spectroscopy is to determine the functional groups of molecules, relevant to both organic and inorganic chemistry

What is IR Spectroscopy?

An IR spectrum is essentially a graph plotted with the infrared light absorbed on the Y-axis against. frequency or wavelength on the X-axis. An illustration highlighting the different regions that light can be classified into is given below.

IR Spectroscopy detects frequencies of infrared light that are absorbed by a molecule. Molecules tend to absorb these specific frequencies of light since they correspond to the frequency of the vibration of bonds in the molecule.



The energy required to excite the bonds belonging to a molecule, and to make them vibrate with more amplitude, occurs in the Infrared region. A bond will only interact with the electromagnetic infrared radiation, however, if it is polar.

The presence of separate areas of partial positive and negative charge in a molecule allows the electric field component of the electromagnetic wave to excite the vibrational energy of the molecule.

The change in the vibrational energy leads to another corresponding change in the dipole moment of the given molecule. The intensity of the absorption depends on the polarity of the bond. Symmetrical non-polar bonds in N≡N and O=O do not absorb radiation, as they cannot interact with an electric field.

Regions of the Infrared spectrum

Most of the bands that indicate what functional group is present are found in the region from 4000 cm^{-1} to 1300 cm^{-1}. Their bands can be identified and used to determine the functional group of an unknown compound.

Bands that are unique to each molecule, similar to a fingerprint, are found in the fingerprint region, from 1300 cm^{-1} to 400 cm^{-1}. These bands are only used to compare the spectra of one compound to another.

Samples in Infrared Spectroscopy

The samples used in IR spectroscopy can be either in the solid, liquid, or gaseous state.

- Solid samples can be prepared by crushing the sample with a mulling agent which has an oily texture. A thin layer of this mull can now be applied on a salt plate to be measured.
- Liquid samples are generally kept between two salt plates and measured since the plates are transparent to IR light. Salt plates can be made up of NaCl, calcium fluoride, or even potassium bromide.
- Since the concentration of gaseous samples can be in parts per million, the sample cell must have a relatively long pathlength, i.e. light must travel for a relatively long distance in the sample cell.

Thus, samples of multiple physical states can be used in Infrared Spectroscopy.

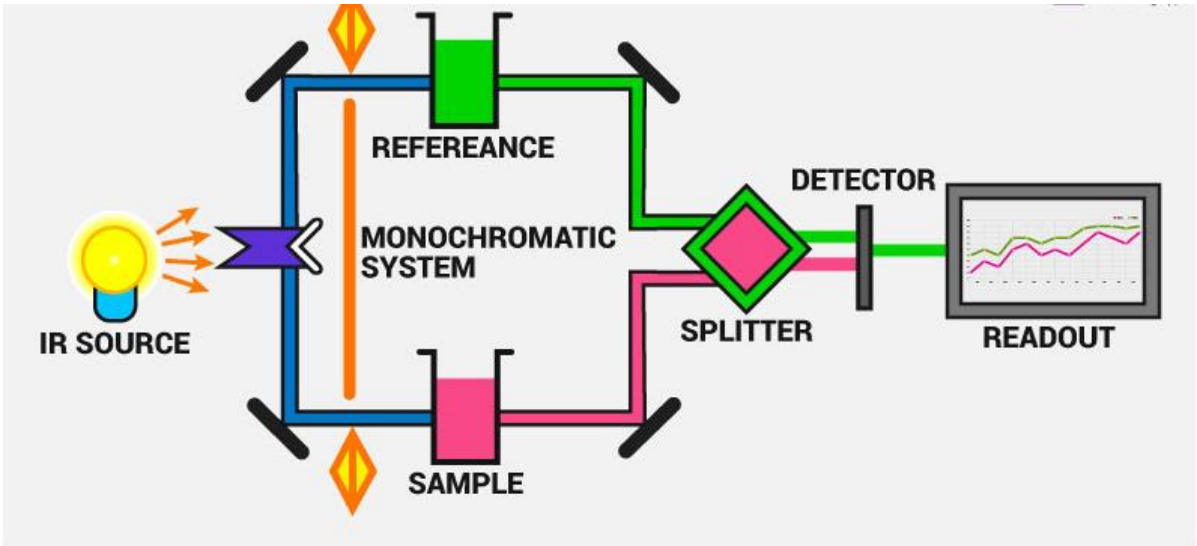
Principle Of Infrared Spectroscopy

The IR spectroscopy theory utilizes the concept that molecules tend to absorb specific frequencies of light that are characteristic of the corresponding structure of the molecules. The energies are reliant on the shape of the molecular surfaces, the associated vibronic coupling, and the mass corresponding to the atoms.

For instance, the molecule can absorb the energy contained in the incident light and the result is a faster rotation or a more pronounced vibration.

IR Spectroscopy Instrumentation

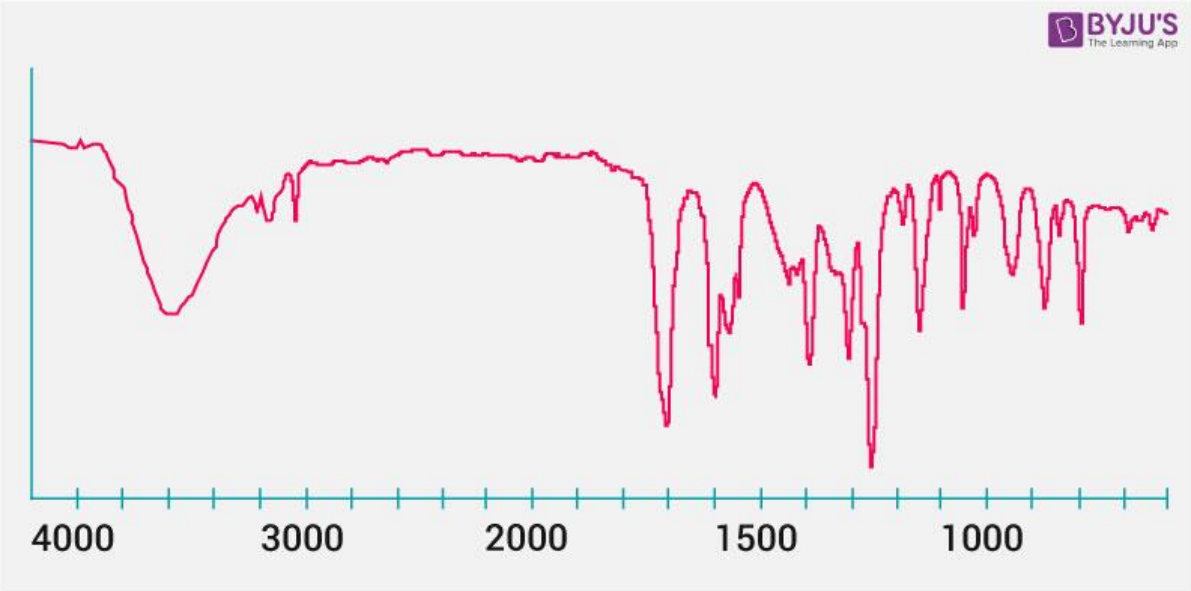
The instrumentation of infrared spectroscopy is illustrated below. First, a beam of IR light from the source is split into two and passed through the reference ant the sample respectively.



Now, both of these beams are reflected to pass through a splitter and then through a detector. Finally, the required reading is printed out after the processor deciphers the data passed through the detector.

Graph of the IR spectrum

Given below is a sample of typical Infrared Absorption Frequencies.



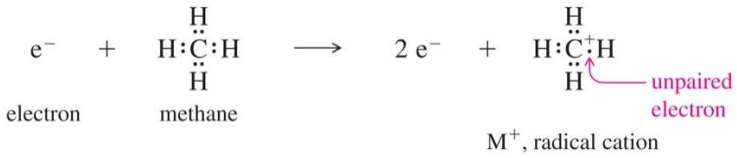
Thus, IR spectroscopy involves the collection of absorption information and its analysis in the form of a spectrum.

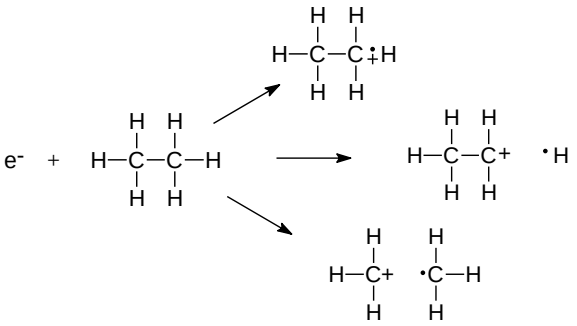
Mass spectrometry

Mass spectrometry (MS) provides the molecular weight and valuable information about the molecular formula, using a very small sample. High-resolution mass spectrometry (HRMS) can provide an accurate molecular formula, even for an impure sample. The mass spectrum also provides structural information that can confirm a structure derived from NMR and IR spectroscopy.

Mass *spectrometry* is fundamentally different from *spectroscopy*. Spectroscopy involves the absorption (or emission) of light over a range of wavelengths. Mass spectrometry does not use light at all. In the mass spectrometer, a sample is struck by high-energy electrons, breaking the molecules apart. The masses of the fragments are measured, and this information is used to reconstruct the molecule. The process is similar to analyzing a vase by shooting it with a rifle, then weighing all the pieces.

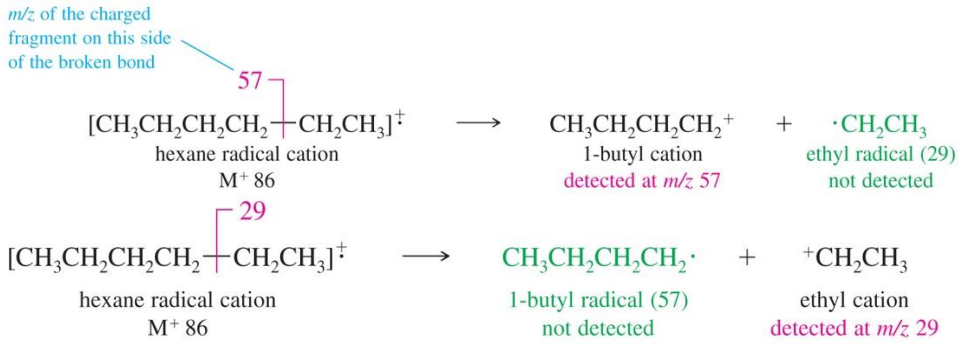
Mass spectrometry refers to destructive methods of analysis. It is based on the ionization of the molecules of the investigated substance and the registration of the mass spectrum of formed ions. There are several methods of ionization, but the so-called electron impact method is the most common at present, when a substance in the gas phase is bombarded by a beam of accelerated electrons. Under these conditions, one electron is initially knocked out of the neutral molecule (M) and a positively charged ion - a molecular ion (the radical cation $M^{+\bullet}$) is formed, which then undergoes a series of successive decays with the formation of smaller positively charged ions (fragment ions) and neutral particles:



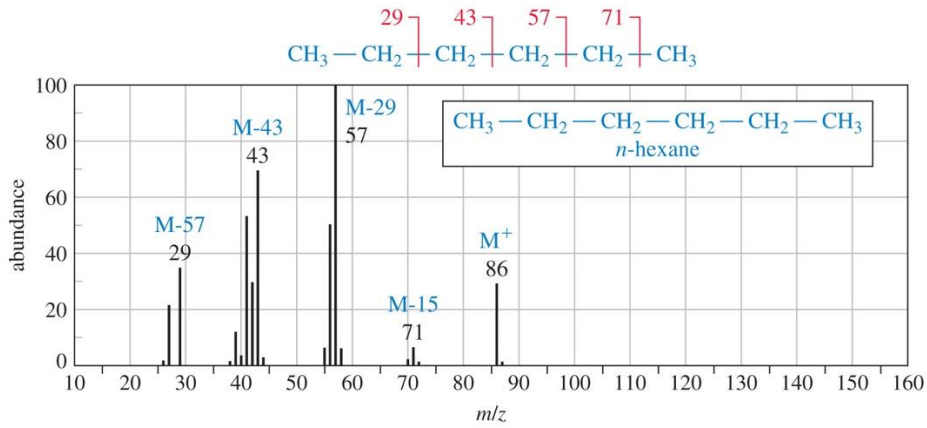


Other fragments can be formed when C—C or C—H bonds are broken during ionization. Only the positive fragments can be detected in MS.

After acceleration in a strong electrostatic field, the flow of positively charged ions is differentially separated in an alternating magnetic field depending on the ratio of their mass to charge (m / z) and is recorded as a spectrum. In the mass spectrum, each positively charged ion appears as a separate signal (peak) whose position is determined by the mass of the ion (more precisely, the mass-to-charge ratio), and the intensity (height) of the signal is proportional to the number of ions with a given mass.

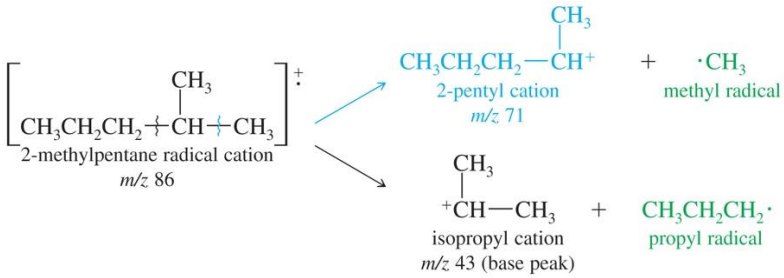


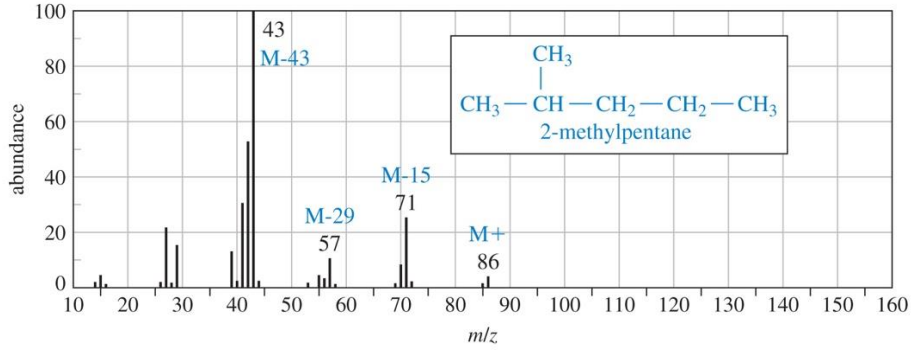
At a low energy of the electron beam (10 eV), the most intense peak in the spectrum, as a rule, corresponds to the molecular ion of the initial molecule. At higher ionization energies (usually 70 eV), the intensity of the peak of the molecular ion falls due to its further decay. The directions of the decay of the molecular ion and the subsequent decays of the fragment ions are determined by the structure of the molecule, so the mass spectrum is characteristic for each compound. In general, the decay of ions obeys the regularities typical for organic reactions and is determined by the location of the charge localization and the stability of the particles formed during the decay. The mass-to-charge ratio for a molecular ion corresponds to the molecular weight of the test substance.



Mass spectrometry is used to establish the structure of organic compounds, their identification and determination of the molecular weight of substances. The high sensitivity of the method, as well as the fact that a small amount of material (up to 10^{-12} g) is sufficient to obtain the result, allows mass spectrometry to be widely used in crimenalistic expertize.

Fragmentation of branched alkanes 2-methylpentane leads to the most stable carbocation fragments form in greater amounts.





A **mass spectrometer** ionizes molecules in a high vacuum, sorts the ions according to their masses, and records the abundance of ions of each mass. A **mass spectrum** is the graph plotted by the mass spectrometer, with the masses plotted as the *x* axis and the relative number of ions of each mass on the *y* axis. Several methods are used to ionize samples and then to separate ions according to their masses. We will empha- size the most common techniques, *electron impact ionization* for forming the ions, and *magnetic deflection* for separating the ions.

Electron Impact Ionization In the **ion source,** the sample is bombarded by a beam of electrons. When an electron strikes a neutral molecule, it may ionize that molecule by knocking out an additional electron.

$e^- + M \rightarrow [M]^{-+} + 2\,e^-$

When a molecule loses one electron, it then has a positive charge and one unpaired electron. The ion is therefore a **radical cation.**