V. K. Ahluwalia





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#### **Preface**

Before the invent of instruments, identification of compounds involved chemical methods for the determination of elements, functional group present and degradation of the compounds by chemical means. In addition to these, volumetric and gravimetric procedures were also used for various estimations. However, with the development of instrumental methods, chemical analysis has now been routinely used. These methods need only a few milligram of the compounds, which can also be recovered and used again in most of the instrumental methods.

The book entitled 'Instrumental Methods of Chemical Analysis' is divided into seven Parts.Part I deals with a brief introduction to instrumental methods of chemical analysis (Chapter 1). Part II deals with chromatography which deals with methods of separation of a mixture of compounds into individual components and their structure determination. Various chromatographic methods include paper chromatography, thin layer chromatography, column chromatography, gas chromatography, high performance liquid chromatography, gel chromatography, ion exchange chromatography and electro-chromatography (Chapters 2-10).

Part III deals with thermal methods of chemical analysis. These include thermogravimetric analysis, differential thermal analysis, thermometric titrations and some miscellaneous thermal methods like derivative thermogravimetric analysis, thermobarography, differential scanning calorimetry, thermomechanical analysis and electric thermal analysis. (Chapters 11-14). Electrochemical methods are described in Part IV. These include coulometric method of analysis, polarography, amperometric titration, potentiometric titrations, conductometric titrations, spectrophotometric titrations, high frequency titrations, pH measurements and calorimetry (Chapters 15-23).

Part V of the book deals with instrumental methods for structure determination of organic compounds. These include infrared spectroscopy, ultraviolet spectroscopy, nuclear magnetic resonance spectroscopy, ultraviolet spectroscopy, Electron Spin Resonance Spetroscopy, Mass Spectrometry, Polarimetry and Optical Rotatory Dispersion and Circular Dichroism. (Chapters 24-30).

Instrumental methods of analysis of inorganic compounds form the subject-matter of Part IV. Various methods include Microwave spectroscopy, Nuclear Quadrupole Resonance Spectroscopy, Raman spectroscopy, Mossbauer spectroscopy, Emission Spectrometry (Chapters 31-35). Some of the methods described in this part are also useful for the indentification of organic compounds.

Some miscellaneous instrumental methods are described in Part VII. These include atomic absorption spectroscopy, flame photometry, fluorimetry and phosphorimetry, nephelometric and turbidimetric techniques, refractory and interferometry and X-ray methods (Chapters 36-41).

Theory underlying each instrumental, procedure and application are described for all instrumental methods. Besides a set of problems are also included at the end of each chapter.

It is hoped that this book will be of immense use to all students in chemistry of all colleges and universities. Any suggestions for the improvement of the book will be of great help and will be gratefully acknowledged and incorporated in the next edition of the book.

I am grateful to Dr Suman Dudeja for the help rendered and useful discussions.

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V.K. Ahluwalia
(Author)

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#### **About the Author**

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# Part-I

**Introduction to Instrumental Methods of Chemical Analysis** 



Chapter 1

# Introduction to Instrumental Methods of Chemical Analysis

#### 1.1 CHEMICAL ANALYSIS

Chemical analysis has been defined in different ways by different investigators. However, it is reasonable to state that chemical analysis involves identification of a substance, the components of a mixture or determination of the structure of chemical compounds. Detection of substances present (qualitative analysis) and estimation of their amounts or concentration (quantitative analysis) is also included chemical analysis.

Before the invent of instruments, identification of compounds involved chemical methods for the determination of elements, functional groups present, degradation by chemical means and final conformation by preparation of derivatives. Volumetric and gravimetric procedures were also used for the various estimations. However, with the development of instrumental methods, chemical analysis has now been widely used. These methods need only few milligrams of the compounds.

#### 1.2 INSTRUMENTAL METHODS-THEIR CLASSIFICATION

A large number of instrumental methods are now available for chemical analysis. These methods can be classified in different groups (Table 1.1).

 Table 1.1
 Types of instrumental methods and their examples

Types of Instrumental methods	Examples
Chromatographic methods	<ul> <li>Paper chromatography</li> <li>Thin Layer chromatography</li> <li>Column chromatography</li> <li>Gas chromatography</li> <li>High performance liquid chromatography (HPLC)</li> <li>Gel chromatography</li> <li>Ion exchange chromatography</li> <li>Electro chromatography</li> </ul>
2. Thermal Methods	<ul> <li>Thermo gravimetric analysis</li> <li>Differential Thermal analysis</li> <li>Thermometric Titrations</li> <li>Miscellaneous Thermal methods</li> <li>Derivative Thermo gravimetric analysis</li> <li>Thermobarography</li> <li>Differential scanning calorimetry</li> <li>Thermo mechanical analysis (TMA)</li> <li>Electric Thermal analysis (ETA)</li> </ul>
3. Electrochemical Methods	<ul> <li>Columetric method of analysis</li> <li>Polarography</li> <li>Amperometric Titrations</li> <li>Potentiometric Titrations</li> <li>Conductometric Titrations</li> <li>Spectrophotometric Titrations</li> <li>High Frequency Titrations</li> <li>pH measurement</li> <li>Calorimetry</li> </ul>
Instrumental Methods for structural determinations of organic compounds	<ul> <li>Infrared Spectroscopy</li> <li>Ultraviolet Spectroscopy</li> <li>Nuclear Magnetic Resonance Spectroscopy</li> <li>Electron Spin Resonance (ESR) Spectroscopy</li> <li>Mass Spectrometry</li> <li>Polarimetry</li> <li>Optical Rotatory Dispersion and Circular Dichroism</li> </ul>
Instrumental Methods for Analysis of Inorganic compounds	<ul> <li>Microwave Spectroscopy</li> <li>Nuclear quadruple Resonance Spectroscopy</li> <li>Raman Spectroscopy</li> <li>Mossbauer Spectroscopy</li> <li>Emission Spectrometry</li> <li>Atomic Absorption Spectroscopy</li> </ul>
Miscellaneous Instrumental     Methods	<ul> <li>Flame Photometry</li> <li>Fluorimetry and phosphorimetry</li> <li>Nephelometric and Turbidimetric Techniques</li> <li>Refractory and Interferometry</li> <li>X-ray methods</li> </ul>

#### 1.3 SELECTION OF THE INSTRUMENTAL METHOD

As already stated (section 1.2) a large number of instrumental method are available for chemical analysis. The choice depends on the nature of the compound to be investigated, its amount, purity and the nature of the investigation one is looking for. It should be kept in mind that in some of the instrumental methods, the sample is destroyed and cannot be recovered back (destructive methods) while in other methods, the sample can be recovered back (non-destructive methods) and can be reused. Non-destructive methods are used if the availability of the sample is very small.

Most of the instrumental methods deal with quantitative analysis. The application of all the methods has been discussed in the relevant chapters. A summary of the methods, including their application and some important information, is discussed in section 1.4.

#### 1.4 APPLICATION OF INSTRUMENTAL METHODS/ TECHNIQUES

#### 1.4.1 Chromatographic Methods

These are used for purification of compounds, identification of the components of a mixture and their confirmation. Using this procedure, even few milligrams of a mixture can be separated. Each component of the mixture can be identified by cochromatography. Alternatively, their structures can be established by spectroscopic techniques like infrared spectroscopy, ultraviolet spectroscopy, NMR spectroscopy and mass spectrometry. Using the appropriate type of chromatography, cations, amino acids, carbohydrates etc. can be separated and identified.

#### 1.4.2 Thermal Methods

**Thermo gravimetric analysis** is useful for the determination of thermal stability of salts, analysis of mixtures and determination of Curie temperature. Besides these, the method is also useful for studying some organic compounds. For example, decomposition of malonic acid.

**Differential Thermal Analysis** is useful for determination of heat of reaction, specific heat, identification of substances, identification of the products of a reaction and for quantitative analysis.

Thermometric titrations can be performed successfully in cases where the end point is difficult to infer due to colour of the solutions like neutralization titrations, precipitation titrations, complexation titrations and redox titrations.

#### 1.4.3 Electrochemical Methods

These are useful for different types of titrations including neutralization titrations, precipitation titrations, complexometric titrations and redox titrations. Besides

these, organic compounds can also be determined, for example, trichloroacetic acid and picric acid are quantitatively reduced at mercury cathode where potential is suitably controlled.

**Polarography:** It is useful for the estimation of cations and anions, estimation of organic compounds containing reducible functional groups and determination of stability constants of metal complexes.

**Amperometric Titrations:** These are useful for the following:

- (i) Determination of sulphate (with lead nitrate), which cannot be determined potentiometrically
- (ii) Determination of Co, Cu and Pd using α-nitroso-β-naphthol
- (iii) Determination of iodide with mercuric nitrate
- (iv) Determination of nickel with dimethylglyoxime
- (v) Determination of lead with dichromate ion
- (vi) Simultaneous determination of Cl<sup>-</sup>, Br<sup>-</sup> and I<sup>-</sup>.

A related technique, viz. ampherometic titration with two indicator electrodes (also called dead stop end point procedure) is applicable when oxidation-reduction system is involved before and after the end point. Examples include titration of iodine with thiosulphate, titration of Fe<sup>2+</sup> by Ce<sup>4+</sup> ion, titration of V<sup>4+</sup> by Fe<sup>2+</sup> and titration of hexacyanoferrate (II) with Ce<sup>4+</sup>.

Potentiometric Titrations: These are useful for neutralization titrations, oxidation-reduction titrations, precipitation titrations and complexometric titrations.

Two related techniques are differential titration and automatic titration.

Conductometric Titrations: These are useful for the titration of strong acid against strong base, titration of weak acid against strong base, titration of strong acid against weak base and titration of weak acid against weak base. The procedure is also used in conductometric precipitation reactions.

**Spectrophotometric Titrations:** These are useful for acid-base titrations, redox titrations, complexometric titrations and precipitation titrations.

High Frequency Titrations: These are useful for acid-base titrations and complexometric titrations. These are also useful for the determination of dielectric constants and analysis of binary mixtures.

pH measurements: pH can be determined by potentiometry using hydrogen electrode, glass electrode, quinhydrone electrode, using a pH meter and also by using pH indicators.

Calorimetry: It has been used for the estimation of Cu<sup>2+</sup> in an unknown solution.

#### 1.4.4 Instrumental Methods for the Determination of the **Structure of Organic Compounds**

**Infrared spectroscopy:** It normally gives information about the presence or absence of various functional groups, particularly hydrocarbons, carboxyl groups, hydroxyl groups, carboxylic acids, carboxylate anion, esters, amines, amino acids, amides, ethers, nitriles, isonitriles and nitro compounds. IR spectroscopy is also helpful to study the progress of a reaction, keto-enol tautomerism, conformational studies, geometrical isomerism, rotational isomerism, identification of inorganic ions and study of inorganic complexes.

The IR spectrum, broadly speaking has two parts: the functional group region  $(4000-1300~{\rm cm^{-1}})$  and the finger print region  $(1300-500~{\rm cm^{-1}})$ . The finger print region has a large number of peaks and is useful to identify a compound by comparing with an authentic sample, a super impossible IR spectra is obtained.

**Ultraviolet Spectroscopy:** The ultraviolet spectroscopy is comparatively less informative than other types of spectroscopy. However, it is still a valuable technique for the identification of organic compounds. It gives information about the presence or absence of multiple bonded systems, conjugated systems, aromatic and heteroaromatic system. The UV spectroscopy is helpful to determine the absence or presence of certain groups in organic compounds, determination of the concentration of an organic compound in solution, determination of the per cent content of vitamin  $A_1$  and vitamin  $A_2$  in natural fats and oils, differentiation between cis and trans isomers, study of keto-enol tautomerism, conformational analysis, charge complex complexes and for the determination of configuration.

**Nuclear Magnetic Resonance Spectroscopy:** It is the most important technique used for the characterization of organic compounds. It gives information about the different kinds of protons in a molecule (environment of hydrogen atoms in a molecule) and the number of different types of protons. Depending on the value of chemical shifts the protons in alcohol, phenols, carboxylic acids, protons on nitrogen, sulphur, halogen and hydrocarbons can be identified. Besides, NMR spectroscopy is helpful for structure elucidation of organic compounds, distinction between isomeric structures, determination of relative amounts of compounds in a mixture, distinction between *cis* and *trans* isomers, studying progress of a chemical reaction, study of rotational isomerism and study the influence of chirality on the chemical shifts of the enantiomers.

A related technique is <sup>13</sup>C NMR spectroscopy. The <sup>13</sup>C NMR spectra is useful to find the environments of carbon atoms in a molecule. The resonances are the same as those that generate <sup>1</sup>H NMR spectra. The basic difference between <sup>13</sup>C and <sup>1</sup>H NMR spectra is that carbon spectra are usually composed of single lines, one for each carbon atom.

In <sup>13</sup>C NMR spectra, coupling occurs between carbon and hydrogen nuclei, but the primary technique for obtaining <sup>13</sup>C NMR spectra, called broad-band decoupling, eliminates the effect of the proton spins towards carbon nuclei. Interpretation of <sup>13</sup>C NMR spectrum makes use of both chemical shift values and coupled spectra (coupling between a carbon nucleus and its attached protons) obtained by off-resonance or by polarization transfer (DEPT), which reveals the presence of CH, CH<sub>2</sub> and CH<sub>3</sub> groups.

<sup>13</sup>C NMR spectroscopy is helpful to identify functional groups containing carbon atoms and number of protons attached to carbon. The <sup>13</sup>C NMR spectras have also been used to confirm other assignments that have been made from other type of spectras.

**Electron Spin Resonance Spectroscopy:** The ESR spectroscopy has been used to study minute quantities of para magnetic species. It also gives the following information:

- The site of unpaired electrons can be known
- The number and type of nuclei present in the vicinity of the odd electron can be found
- The type of nuclei that is responsible for the splitting can be determined by the relative intensities of the spectrum lines in the ESR spectra
- The value of g can be found.

Besides ESR has been used to study free radicals even in low concentration, for determination of reaction mechanism and the study of inorganic compounds.

Mass spectrometry has been extensively used for determination of structure of organic compounds, molecular weight and molecular formula. It has also been used for the detection of pesticides in the environment and for the analysis of drugs and their metabolites. On the basis of fragmentation the presence of different types of compounds like saturated hydrocarbons, unsaturated hydrocarbons, alcohols, phenols, ethers, carbonyl compounds, carboxylic acids, esters, lactones, amines, amides and nitriles can be identified.

In combination with Gas chromatographs, it is useful to determine the structure of various compounds in a mixture.

Optical Rotatory Dispersion and Circular Dichroism: This technique is useful for determining the stereochemistry of aliphatic amino acids, steroids, location of hydroxyl group in steroidal alcohol-rubijervine, determination of absolute configuration and conformation.

**Polarimetry:** It has been used to determine an optically active substance in presence of other optically inactive substances, study of optical isomerism, differentiation between two enantiomers of a compound and study of configuration.

# 1.4.5 Instrumental Methods of Analysis of Inorganic Compounds

#### **Microwave Spectroscopy**

It has been used to determine the structures of a number of molecules like xenon oxyfluoride, carbon oxysulphide, benzonitrile, ammonia and ozone. Using this technique, barrier heights of certain molecules like  $CH_3OH$  (C—O),  $CH_3OCH_3(C$ —O),  $C_6H_5CH_3(C$ —C) and  $CH_3NO_2(C$ —N) can be measured.

**Nuclear Quadrupole Resonance Spectroscopy:** It has been used for interpretation of eQq data, nature of chemical bond, structures of group III halides, study of charge transfer compounds, chloromethanes, and chloroacetyl chlorides.

Raman Spectroscopy: It has been used to determine the structure of CO<sub>2</sub>, N<sub>2</sub>O, SO<sub>2</sub>, H<sub>2</sub>O, chloro complexes of mercury, HCN and CS<sub>2</sub>. Nature of bonding in tetrahedral complexes have also been determined. Raman spectroscopy also finds applications in physical chemistry in studying ionic equilibria in solution and single crystals. In organic chemistry, Raman spectroscopy is helpful to determine the molecular structure of organic compounds, ring size in cycloalkanes and in quantitative analysis. Polymers have also been studied.

**Mossbauer Spectroscopy:** This type of spectroscopy is useful for the determination iron in tin ores, alloys and industrial wastes. It is useful for determining the nature of the chemical bond, quadrupole effects and determination of structure of iron dodecacarbonyl.

**Emission Spectroscopy** is useful in qualitative analysis, determination of metals and quantitative analysis. It is also useful for the determination of the impurities of Ni, Cr, Si, Mn, Mo, Cu, Al etc. in iron and steel in metallurgical process. Alloys of Al, Mn, Zn, Cu, Pb and Sn can also be analyzed. Also useful for determination of elements like Na, K, Zn, Cu, Ca, Mg, Fe, Ni in animal tissues.

**Atomic Absorption Spectroscopy:** It is an important technique and is useful in the detection and estimation of a number of metals in the environment (water, air and land). Metallic elements in blood serum can also be determined. It is also useful for the determination of lead in petrol, estimation of mercury in water.

#### 1.4.6 Miscellaneous Instrumental Methods

**Flame Photometry:** It finds application in qualitative analysis in detection of alkali metals (Na, K, Li) and rare earth metals (Mg, Ca, Sr, Ba). In quantative analysis for the estimation of Al, Sb, As, Ba, Cu, Fe, Pb, Hg, Ni and Pd. It is also useful for analysis of biological fluids and tissues, analysis of soil for the determination of Na, K, Al, Cu, Co, Fe; analysis of natural and industrial waters and in the analysis of glass, cement and metallurgical products.

Fluorimetry and Phosphorimetry: Fluorimetry is useful for the determination of uranium in salts, analysis of inorganic ions like ruthenium ion in the presence of other platinum metals, determination of Al in alloys, determination of trace elements in steel and determination of cadmium. It is also useful in organic analysis for the determination of vitamin  $B_1$ , vitamin  $B_2$ , adenine, anthranilic acid, aromatic polycyclic hydrocarbons, cysteine, guanidine, indole, skatole, tryptophan, adrenaline, penicillin, phenobarbital etc. Some of the plant products like chlorophyll, ergot, ascorbic acid, folic acid etc. can also be determined.

Phosphorimetry is useful for:

- analysis of aspirin in blood serum.
- procaine, cocaine, phenobarbital and chlorpromazine can also be determined in blood serum in low concentrations.
- analysis of cocaine and atropine in urine.

#### **Nephelometric and Turbidimetric Techniques**

These are used in inorganic analysis for the determination of sulphates and carborides as BaSO<sub>4</sub> and BaCO<sub>3</sub> respectively. Also chlorides, fluorides, cyanides and calcium are determined as AgCl, CaF<sub>2</sub>, AgCN and calcium oxalate respectively. Other applications include determination of sulphur in coke, coal, oils, rubbers, plastics etc.

In organic analysis, turbidimetric procedure is used for the determination of turbidity in sugar products, citrus juices etc. Also benzene in alcohol can be determined.

In biochemical analysis measurement of the growth of bacterium in nutrient medium and finding the amounts of amino acids, vitamins and antibiotics. Determination of protein, yeast, glycogen and beta and gamma globurin in blood and plasma is also possible.

Continuous monitoring of air and water pollution can be done by turbidimetry and nephilometry. In air, dust and smoke can be monitored and in water turbidity can be measured.

Molecular weight of high polymers can also be determined.

#### Refractometry and Interferometry

Refractometry has been used for qualitative and quantitative analysis and also for structural investigations. It is also used for the determination of conjugation and  $\alpha$ ,  $\beta$ -unsaturated compounds.

Inferferometry has been used for the analysis of gases and for the determination of permeability of baloon fabrics to helium and hydrogen. It is also used for analysis of sewage and milk, in biological problems like CO<sub>2</sub> in blood, ethyl alcohol in blood and concentration of heavy water.

**X-ray Methods:** The three types of X-methods are X-ray absorption, X-ray diffraction and X-ray fluorescence. The X-ray absorption is useful for the determination of one element markedly heavier than the others. Other applications include determination of chlorine in hydrogen, plastics, hydrocarbon and for the determination of BaF<sub>2</sub> in C and Ba / Pb in special type of glasses.

X-ray Diffraction is useful for the determination of structure of crystals, characterisation of polymers, metals, nature of complexes, structure of diamonds, graphite, penicillin, Vitamin  $B_{12}$  etc.

X-ray Fluorescence finds use in elemental analysis, in agriculture, medicine, mining, metallurgy etc.



# Part-II

#### Chromatography

- Introduction
- Paper Chromatography
- Thin Layer Chromatography
- Column Chromatography
- Gas Chromatography
- High Performance Liquid Chromatography (HPLC)
- Gel Chromatography
- Ion Exchange Chromatography
- Electro Chromatography



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Chapter 2

## Chromatography

#### 2.1 INTRODUCTION

The term chromatography was coined by a Russian scientist Tswett (1906), who reported the separation of different coloured constituents of leaves by extraction of the leaves by a suitable solvent and passing the extract through a column of calcium carbonate, alumina and sucrose. The name chromatography was derived from the greek word chromon meaning colour. According to the International Union of Pure and Applied Chemistry (I.U.P.A.C.), chromatography is defined as a physical method of separation of the components of a mixture. According to I.U.P.A.C., the components to be separated are distributed between two phases, *viz.*, stationary phase and the mobile phase. Usually, a stationary phase is a column, but may take other forms. The mobile phase is a suitable liquid.

Chromatography is used for purification of compounds, identification of the components of a mixture and their separation. In fact, chromatography is the best procedure for the purification of compounds. It is used for the purification of solids, liquids and gases as compared to other methods, like crystallization, sublimation, distillation etc., which have only limited applications. A special advantage of chromatography is that even few milligrams of mixture of compounds can be separated, which is not possible by other methods.

Each component of the mixture after separation can be identified by spectroscopic techniques like infrared spectroscopy, ultraviolet spectroscopy, NMR spectroscopy and mass spectrometry.

#### 2.2 PRINCIPLE OF CHROMATOGRAPHIC SEPARATION

A mixture is separated into its components by using two immiscible substances. These are a stationary phase, (which may be a solid or liquid), which adsorbs the

components of the mixture and the mobile phase (which may be a liquid or gas), which during passing through the stationary phase transports the components of the mixture on to it. The components of the mixture are transported at different rates. The strongly adsorbed component move slowly with the moving phase and the weakly adsorbed component move faster along with the moving phase. Thus, as the mobile phase passes through the stationary phase, the differently adsorbed components are separated. The separation of the components is based on selective adsorption when the stationary phase is a solid and the moving phase is a liquid. However, when the stationary phase is a liquid and the moving phase is a liquid or gas, the separation of the components is based on the partition of the components between the two phases.

#### 2.3 TYPES OF CHROMATOGRAPHY

Chromatography can be of different type depending on the process of either adsorption or partition. Different types of chromatography include:

- Partition chromatography
- Adsorption chromatography
- Exclusion chromatography
- Ion exchange chromatography.

#### 2.3.1 Partition Chromatography

In this technique, a mixture of substances are separated by partition between a stationary liquid phase (which is held on a suitable solid support) and a moving solvent (mobile phase). In case the moving phase is a liquid, the chromatography is called liquid-liquid chromatography. However, when the moving phase is a gas, the chromatography is called gas-liquid chromatography. The former type of chromatography (liquid-liquid chromatography) is carried out on cellulose or moist silica gel. Examples include paper chromatography and thin layer chromatography. In case the adsorbant is packed into columns, it is called column chromatography. In all these types of chromatography, the medium acts as a support of water. Different types of partition chromatography are summarised in Table 2.1.

Type of chromatography	Stationary phase	Mobile phase	Principle
Paper chromatography	Liquid	Liquid	Partition
Thin layer chromatography (TCL)	Solid	Liquid	Partition
Column chromatography	Solid	Liquid	Adsorption

Table 2.1 Different types of partition chromatography

#### 2.3.2 Adsorption Chromatography

In this technique, the separation of a mixture into its components depends on the differences in the degree of adsorption of various components of the mixture on a stationary solid phase and elution with a moving solvent (liquid or gas). When the moving phase is a liquid, it is called liquid-solid chromatography or column chromatography. However, when the moving phase is a gas it is called liquid-gas chromatography (GLC). Different types of adsorption chromatography are summarised in Table 2.2.

Type of chromatography	Stationary phase	Mobile phase	Principle
Column chromatography (see also table 2.1)	Solid	Liquid	Adsorption
Gas liquid chromatography (GLC)	Liquid	Gas	Partition

Table 2.2 Different types of adsorption chromatography

#### 2.3.3 Exclusion Chromatography

In this technique, the separation of a mixture into its components is based on the molecular size of the components. This techniques is two types:

- (i) Gel Permeation Chromatography: In this technique the stationary phase consist of gel (beads of porous synthetic material) and the mobile phase is a liquid which percolates through the beads). The gel swells in the solvent resulting in the increase in the spaces between the polymer chains. Only a definite size of the molecule can just penetrate the interior of the gel.
- (ii) Sieving Separation: In this technique, molecular sieve (e.g., zeolite) is used in the column in order to separate small molecules from larger molecules.

#### 2.3.4 Ion-Exchange Chromatography

This technique has been successfully used for the separation of ions of similar properties using ion exchangers. A discussion on the ion-exchange chromatography forms subject-matter of a subsequent section.

In the foregoing sections following types of chromatographic techniques are discussed:

- Paper Chromatography
- Thin-Layer Chromatography
- Column Chromatography
- Gas Chromatography

- High Performance Liquid Chromatography (HPLC)
- Gel Chromatography
- Ion Exchange Chromatography
- Electro Chromatography



- 1. Explain the term chromatography.
- 2. Discuss the principle of chromatographic separation.
- **3.** Explain partition chromatography, adsorption chromatography, exclusion chromatography and ion exchange chromatography.





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Chapter 3

## **Paper Chromatography**

Paper chromatography is the simplest most widely used technique for the separation and identification of the constituents of a mixture. The stationary phase in paper chromatography is a liquid, *i.e.*, water (adsorbed on cellulose of the Whatman filter paper) and the moving phase (known as irrigant) is also a liquid, which may be a solvent or mixture of solvents. Paper chromatography is a type of partition chromatography (partition between two liquid phases). The components of the mixture to be separated travel at different rates and appear as spots on different points of the Whatman filter paper. Following are given various types of paper chromatography in use.

- Circular (or radial) paper chromatography
- Ascending paper chromatography
- Descending paper chromatography.

#### 3.1 CIRCULAR (OR RADICAL) PAPER CHROMATOGRAPHY

In this technique, a circular Whatman filter paper (about 12 cm diameter) is used. A small strip is cut from the central of the filter paper, the strip is bent vertically [Fig. 3.1(a)]. Alternatively a wick made of fine cotton is inserted in a small hole which is made at the centre of the circular filter paper [Fig. 3.1(b)].

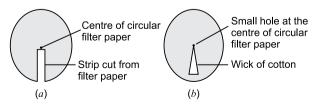


Fig. 3.1 Whatman filter paper used in circular paper chromatography

The filter paper is spotted at the centre with a solution of the compound or the mixture is a suitable solvent using a capillary tube. The filter paper is spotted 2-3 times at the same place so that a reasonable concentration of the spot is achieved. The spotted filter paper is dried in air (so that the solvent evaporates) and is placed on a petri dish containing a suitable irrigant (moving phase). The central paper strip or the cotton wick should dip in the solvent. A glass plate is placed on the filter paper (Fig. 3.2). The solvent moves *via* the strip of filter paper or cotton wick and takes the compound along with it. When the solvent has moved about 4 cm from the centre, the filter paper is taken out, solvent front is marked with a soft pencil and the chromatogram is dried in air.

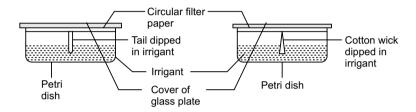


Fig. 3.2 Apparatus used for paper chromatography

The moving phase in circular paper chromatography can be a pure solvent (*e.g.*, benzene, cyclohexane etc.) or a mixture of solvent (like isopropanol-ammonia-water (9:1:2), *n*-butanol-acetic acid-water (4:1:5), or phenol saturated with water etc.) The choice of the solvent is very important. The solvent should be such so that the compound moves along with the solvent in a reasonable time.

In the circular chromatogram, various components appear as circular bands. The coloured bands are visible. Alternatively, the chromatogram is developed with a suitable developer so that the bands become visible. The developed chromatogram appears as in Fig. 3.3.

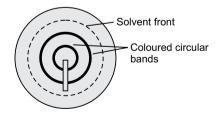


Fig. 3.3 Developed chromatogram

The choice of the reagent for development of the chromatogram depends on the nature of the compound (or mixture) used (Table 3.1).

Compound	Reagent	
Carboxylic acids	Bromo cresol green	
Amino acids	Ninhydrin	
Aldehydes or ketones	2,4-Dinitrophenyl hydrazine (2,4-DNP)	
Basic N or S	Potassium hydrogen phthalate	
Terpenoids	Antimony pentachloride in CCl <sub>4</sub>	

Table 3.1 Developing reagents

The chromatogram can also be developed by putting the circular filter paper in a chamber, (which contains a few crystal of iodine) for about 30 seconds. A large number of compounds give colour with iodine vapour. Alternatively, the bands can be made visible by using an ultraviolet light (from a uv lamp).

Finally, the  $R_f$  value of each band in the chromatogram is calculated by measuring the distance travelled by the component and the solvent from the origin.

$$R_f = \frac{\text{Distance travelled by the compound}}{\text{Distance travelled by the solvent}}.$$

#### Notes:

- The mobile phase or the solvent should be kept in the petri dish for at least half an hour before the experiment is performed so that the air in the petri dish is saturated with the vapours of the solvent, otherwise the solvent will evaporate from the paper faster than it is replaced by capillary action and separation of the components will not be achieved.
- 2. The spotting should be done with a very fine capillary so as to put small spots, two-three times at the same point and the solvent should be evaporated before each spotting (*i.e.*, the spot should be dried before the next spotting is done).
- 3. Paper chromatography is a much better method (as compared to TLC) for calculating  $R_f$  values as these values are far more reproducible in paper chromatography.

## **Applications**

The individual components in a mixture can be identified by comparing their  $R_f$  values with known compounds under identical conditions. Alternatively, as an example, if the mixture contains p-cresol, the mixture is mixed with about 1 mg of p-cresol and the mixture spotted. The chromatogram will show the same circle. No separate circle of p-cresol will be observed.

### 3.2 Ascending Paper Chromatography

In this technique, a strip  $(2 \times 20 \text{ cm})$  of Whatman filter paper is taken. Small spots of the solution of the mixture of compounds are put on the strip at a distance of about 2 cm above the edge of the paper [Fig. 3.4(a)]. The irrigant or the solvent is placed in a glass jar, whose lid has a small hook. The spotted strip of paper is suspended in the glass jar using the hook. Care should be taken so that the lower edge of the strip of paper dips in the solvent, but the spots is the mixture are above the solvent [Fig. 3.4(b)]. The solvent rises up by capillary action on the filter paper and the components of the mixture move up at different rates. When the solvent front has reached near the top of the strip, the strip is taken out, solvent front marked and the strip is air dried.

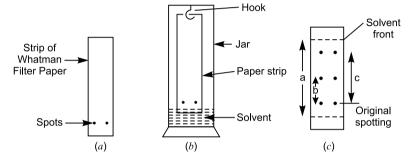


Fig. 3.4 Ascending paper chromatography

Finally, the air-dried strip is developed in a chamber of iodine vapour or by a suitable reagent (see Table 3.1). The spots can also be visualised by UV light. The developed spotted chromagram is shown in Fig. 3.4(c).

The  $R_f$  of the different components is calculated as in the case of circular paper chromatography

$$R_f = \frac{\text{Distance travelled by the compound}}{\text{Distance travelled by the solvent}}$$
$$= \frac{b}{a} \text{ for compound one and } \frac{c}{a} \text{ for the other compound.}$$

The  $R_f$  value is always less than 1.

The ascending paper chromatography is useful for finding, the number of components in a mixture and finding their identity (as in the case of circular paper chromatography).

#### 3.3 DESCENDING PAPER CHROMATOGRAPHY

In this type of chromatography, the solvent is placed at the top of a tank in a boat shaped container [Fig. 3.5(a)]. The solvent moves down and so the name

descending paper chromatography. The filter paper is spotted at the top as in the case of a ascending paper chromatography and hung in a jar, making sure that only the paper end dips in the solvent and not the spots ([Fig. 3.5(b) and 3.5(c)]. The atmosphere in the tank is saturated with the vapour of the solvent. This is achieved by placing a small amount of the solvent at the bottom of the tank. In this type of chromatography the solvent moves down the paper by capillary action and also by gravitational force. As the rate of solvent movement is fast, the chromatography takes much shorter time.

As the solvent moves, the components of the mixture also move at different speed and get separated. After the chromatography is complete, the filler paper is taken out, solvent front marked and dried in air and the spots developed as in the earlier cases and  $R_f$  calculated.

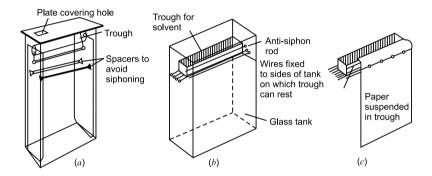


Fig. 3.5 Apparatus used for descending paper chromatography

#### 3.4 PAPER CHROMATOGRAPHIC SEPARATIONS

Following are given some of the typical separations using paper chromatography.

# 3.4.1 Separation and Identification of Group 1 Cations (Pb<sup>+</sup>, Ag<sup>+</sup>, Hg<sup>+</sup>)

- (i) Using a Whatman filter paper strip, one drop of the three cations solution in water is placed near the end of the strip (2 cm above the end) along with a spot of the mixture of the three cations.
- (ii) The paper strip is suspended (after drying) into a cylinder containing 25 mL of the solvent (distilled water or ethyl alcohol).
- (iii) The chromatogram is run for about 2 hours till the solvent front reaches just below the paper strip.
- (iv) The paper strip is taken out, solvent front marked, dried in air and sprayed with potassium chromate solution (0.25 M). Alternatively, the chromatogram is dripped in a solution of 0.25 M K<sub>2</sub>CrO<sub>4</sub>.

- (v) Three spots (one each from individual cation) and three spots of the mixture containing Pb<sup>+</sup>, Ag<sup>+</sup> and Hg<sup>+</sup> appear as yellow, orange-red and orange respectively.
- (vi) By observation of the chromatogram, each of the spots due to Pb<sup>+</sup>, Ag<sup>+</sup> and Hg<sup>+</sup> are located and also confirmed.
- (vii) The  $R_f$  values of each of the spots is calculated and recorded.

# 3.4.2 Separation and Identification of Cations of Group II (Hg<sup>2+</sup>, Cu<sup>2+</sup>, Cd<sup>2+</sup> and Bi<sup>3+</sup>)

Following procedure is followed:

- (i) Spot the Whatman filter paper strip with solutions of mercuric chloride, copper chloride, cadmium chloride and bismuth trichloride. A spot of the mixture of the above four solution is also put along with the earlier spots (in the same line).
- (ii) The paper strip is suspended in a cylinder containing about 25 mL of the solvent (obtained by mixing *n*-butanol and 3N HCl (1:1), shaking the mixture in a separation funnel. The upper layer is used as the solvent for chromatography).
- (iii) The chromatogram is run for 2.5-3 hrs till the solvent front reaches almost at the top of the filter paper strip.
- (*iv*) The chromatogram is taken out, solvent front marked, dried and sprayed with a solution of dithiazone in chloroform. Following spots appear:

 $\begin{array}{ccccc} Brown & - & Cu^{2+} \\ Pink & - & Hg^{2+} \\ Purple & - & Cd^{2+} \\ Brown & - & Bi^{3+} \end{array}$ 

The original spot of the mixture is also resolved into the four spots (as stated above).

(v) The  $R_f$  values of each of the cation is calculated and recorded.

# 3.4.3 Separation and Identification of Cu<sup>2+</sup> and Cd<sup>2+</sup> using Paper Chromatography

- (i) Spot the Whatman filter paper strip with a solution of CuSO<sub>4</sub> in water and cadmium chloride is water on a line equidistance from each other.
   A third spot of a 1:1 mixture of CuSO<sub>4</sub> and CaCl<sub>2</sub> solution is also applied.
- (ii) The chromatogram is run using the solvent (Ethanol-5N HCl; 9:1) taken in a cylinder. It takes about 1 hr for the solvent front to reach near the top of the paper.

- (iii) The filter paper strip is taken out, dried in air and solvent front marked with lead pencil.
- (iv) Develop the chromatogram by spraying with colourless ammonium sulphide solution (obtained by bubbling H<sub>2</sub>S in dilute NH<sub>4</sub>OH solution). Both Cu<sup>2+</sup> and Cd<sup>2+</sup> appear as black spots. The mixture gives two black spots.
- (v) Calculate the  $R_f$  of both  $Cu^{2+}$  and  $Cd^{2+}$  ( $Cd^{2+} > Cu^{2+}$ ).

In place of Ethanol-5N HCl (9:1) as mobile phase, a saturated solution of n-butanol with 3 N HCl can also be used.

A summary of the analysis of cations in given in Table 3.2.

Cations	Irrigant solvent	Spraying reasons	Type of chromatography
Group II Hg <sup>2+</sup> , Cu <sup>2+</sup> , Cd <sup>2+</sup> , Bi <sup>3+</sup>	n-Butanol-3NHCl (1:1) upper layer	Dithiazone solution in chloroform	Paper
Group I Pb <sup>+</sup> , Ag <sup>+</sup> , Hg <sup>+</sup>	Distilled water or ethyl alcohol	$K_2CrO_4$ solution (0.25M)	Paper
Cu <sup>2+</sup> , Cd <sup>2+</sup>	Ethanol-5N HCl (9:1)	Ammonium sulphide solution	Paper
Ni <sup>2+</sup> , Co <sup>2+</sup>	Acetone-water- Conc. HCI (44:3:4)	Sat'd soln. of alizarin containing 0.1% rubeanic acid and 1% Salicylaldoxime	TLC (to be discussed subsequently

Table 3.2 Summary of the analysis of cations

# 3.4.4 Separation and Identification of Amino Acids by Descending Paper Chromatography

Let us suppose that the mixture of amino acid contains DL-alanine, glycine, L-leucinc and L-lysine hydrochloride.

- (i) Prepare a solution of each of above mentioned amino acids (5 mg each) separately in water (0.25 mL). If necessary the solution be warmed to get a clear homogeneous solution. In a similar, way the mixture of abovementioned amino acids is dissolved in water. Thus, we get a total of five solutions.
- (ii) As mentioned in section 3.3, the filter paper strip  $(10 \times 50 \text{ cm})$  is spotted carefully with all the five solution. The spotting be done 2-3 times in each case so that a reasonable concentration of amino acids is spotted.

- (iii) The irrigant (solvent) is prepared by mixing 40 mL *n*-butanol, 10 mL glacial acetic acid and 50 mL distilled water in a separatory funnel. The solution is mixed thoroughly and allowed to settle in upper and lower layers. The upper layer is used as an irrigant and taken in a boat shaped container [Fig. 3.5(a)]. A small amount of the same solvent is also placed at the bottom of the tank in order to saturate with the vapours of the solvent.
- (iv) The spotted filter paper is suspended in the tank [Fig. 3.5(b) and 3.5(c)]. After the chromatogram is complete as indicated by the solvent front reaching near the end of the strip. The filter paper is taken out, solvent front marked with pencil and dried in an oven at 100° for about 5 min.
- (v) The chromatogram is developed by sparing lightly with ninhydrin (0.2 per cent solution in 2-butanol. The spraying is carried out in a fume cupboard. The colour of the spots of amino acids appear by placing the paper in an oven at  $100^{\circ}$ C for 5-10 min. Finally, the  $R_f$  value corresponding to each amino acid is calculated.

# 3.4.5 Separation and Identification of Monosaccharides by Descending Paper Chromatography

Let us suppose that the mixture of monosaccharides contains D-xylose, D-arabinose, D-glucose, D-galactose and D-fructose.

Following procedure is followed:

- (i) Prepare a solution of five monosaccharides as mentioned above and also a solution of the mixture of above monosaccharides.
- (ii) Proceed as in section 3.4.4.
- (iii) The irrigant (solvent) is a homogeneous solution of butan-1ol, acetone and water (4:5:1).
- (*iv*) The development of the chromatogram takes 12-14 hrs. In this case, the solvent is allowed to drip from the paper end.
- (v) The paper is removed, suspended in a fume-cupboard and dried using a hot-air blower.
- (vi) The paper is sprayed with an ethanolic solution of naphthorescinol (0.2 per cent of naphthorescinol in ethanol containing 10 per cent V/V of orthophosphoric acid). The paper is finally dried in an oven at 100°C for 3-5 min or until the spot become clear.
- (vii) The  $R_g$  value of each monosaccharide is calculated

$$R_g = \frac{\text{distance moved by monosaccharide}}{\text{distance moved by D-glucose}}$$

**Notes:** In this case, the  $R_f$  values monosaccharides are low. This necessariates allowing the solvent a drip from the filter paper end. The chromatographic mobility

of each monosacchride is compared with the behaviour of a standard D-glucose. Hence,  $R_{\varrho}$  values are used.



- 1. What is paper chromatography? What is the principle involved in the separation of components of a mixture?
- **2.** Explain  $R_f$  value. How it is determined?
- **3.** What do you understand by ascending and descending paper chromatography.
- **4.** What is two dimensional paper chromatography? What are its applications?
- **5.** Using paper chromatography, how will you show that the two compounds are identical?





Chapter 4

## Thin Layer Chromatography

Thin layer chromatography is an adsorption type of chromatography. In this technique, the adsorbent or the stationary phase is coated as a thin layer on a glass plate and hence the name thin layer chromatography. The moving phase may be a pure solvent or a mixture of solvents. Commonly used adsorbents are silica gel and alumina. The strength of the layer is increased by addition of a small amount of calcium sulphate. The thickness of the adsorbent is generally in the range 0.15 to 2.0 mm. In TLC, besides silica gel and alumina, some other adsorbents like cellulose powder and Kiesleguhr can also be used. Glass plate of different sizes e.g.,  $2.5 \times 7.5$  cm,  $5 \times 20$  cm,  $20 \times 20$  cm (depending on the nature and the amount to mixture to the separated) are used.

TLC technique is superior to other chromatographic techniques. The separation of a mixture into component and their identification can be carried out in a much shorter time (20-30 min). It can be used for both organic and inorganic compounds. The spraying reagents can also be acids without any adverse effects.

#### 4.1 PRINCIPLE OF TLC SEPARATION

As already stated (section 2.3.2), the mixture gets adsorbed on to the adsorbent. The mobile phase, as it rises above the TLC plate transports the components of the mixture on the plate. The components of the mixture are transported at different rates. The weakly absorbed component move faster with the moving phase and the strongly absorbed component moves slowly. Thus, the separation of the components is based on selective absorption on to the stationary phase (adsorbent).

#### 4.2 PREPARATION OF TLC PLATES

This is most important part of the TLC separation technique. In fact a good separation of the components of the mixture depends on the quality of the TLC plate. Following procedure is followed.

The glass plates of appropriate size are first cleaned with a laboratory detergent, washed with distilled water and dried. The clean and dried plates should be handled by the edges or by the under surface which is not to be coated with the adsorbant.

A slurry of the adsorbent (usually silica gel or alumina) is prepared by shaking 30 g of the adsorbent in 100 mL CCl<sub>4</sub> in a wide-necked bottle. A pair of microscope slides are held together and dipped in the slurry, slowly withdrawn and allowed to drain for few sounds while being held over the bottle. The slides are carefully parted and placed in a rack horizontally and allowed to dry for 10–15 min. The rack has the capacity to hold 20–15 plates at a time. The dried plates, are activated by heating at 110°C for 15–20 min. (before use) and kept in a desiccator. Such microscope slides are best for use for following the progress of a chemical reaction.

Large single–glass plate (*i.e.*,  $20 \times 5$  cm) is coated convently by using the easily assembled apparatus (Fig. 4.1). The apparatus consist of a sheet of glass plates ( $20 \times 30$  cm) at the upper and lower ends of which two glass plates ( $20 \times 5$  cm) are secured. The plate to be coated is placed in the central depression and is held in position by the uncoated plates on either size. The thickness of the layer can be adjusted (to 0.25 - 0.3 mm) by wrapping both ends of a glass rod 14 cm long and 7.5 cm diameter with equal lengths (12.5 cm) 2.5 cm of sellotape (this is the spreader).

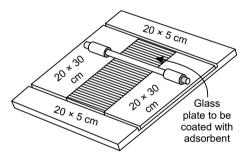


Fig. 4.1 Apparatus for making TLC Plates

The composition of the slurry for spreading depends on the nature of the adsorbent. Follow the instructions given by the supplier of the instrument.

TLC spreaders (Fig. 4.2) are also commercially available (one such supplier is Shadon southern instruments Ltd.). These consist of float frame which can rigidly hold five  $20 \times 20$  glass plates or equal number of  $20 \times 10$  cm or  $20 \times 5$  cm plates. All plates are alligned in the same plane so that a uniform thickness of adsorbent layer is spread on all plates. The slurry of adsorbent in spread on all plates. The slurry of adsorbent in spread with the help of a metal hopper (Fig. 4.3), which draws across the surface. The thickness of the layer can be preselected by adjustment of the nuts.

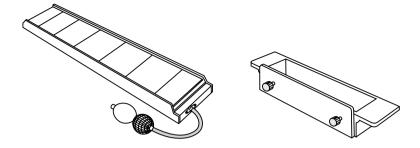


Fig. 4.2 TLC spreader

Fig. 4.3 Metal Hopper

The TLC plates thus prepared are allowed to dry in air (20 - 25 min) and then heated in an air oven at  $100^{\circ}$ C for 10-15 min.

Now a days prepared TLC plates (particularly microscope slides) are available for immediate use.

#### 4.3 PROCEDURE FOR TLC

Following steps are followed:

- (i) Dissolve the mixture to be separated into its components in a suitable solvent (10 mg in 1–2 mL solvent, e.g., 1–2%). Using a fine capillary, it is spotted on the plate about 2 cm above the lower edge of the plate [Fig. 4.4(a)]. The spots must be as small as possible and at the same distance from the lower edge of the plate.
- (ii) The spotted TLC plate is placed in a glass Jar [Fig. 4.4(b)] containing the moving phase (20–25 mL) such that the lower end of the adsorbent layer of the TLC plate is under the solvent but the spots of the applied mixture are above the solvent [Fig. 4.5(a)].

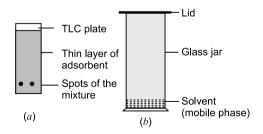


Fig. 4.4 Apparatus for TLC

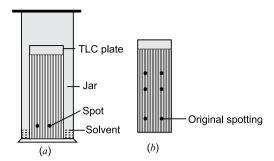


Fig. 4.5 TLC

(iii) The glass jar is covered. The moving phase moves up the adsorbent carrying different components of the mixture at different rates. When the solvent has moved almost to the top end of the adsorbent, the TLC plate is taken out of the Jar and the solvent front is marked with a pencil and the solvent allowed to evaporate.

In case, the components of the mixture are coloured, the coloured spots are visible on the plate. The number to spots is equal to the number of components in the mixture [Fig. 4.5(b)].

(iv) Development of the TLC plate.

If the components are not coloured, the location and the number of components can be found by any of the following procedures:

- (a) The TLC plate may be viewed under uv light. Some components which are fluorescent in nature can be seen in the UV light.
- (b) The plate may be kept in a Jar containing few crystals of iodine. The compounds absorb iodine and the spots become visible. The spots may be yellow or brown in colour.
- (c) The plate may be sprayed with a suitable reagent. For example, if the mixture contains carbonyl compound, the spraying reagent is a solution of 2, 4 dinitrophenyl hydrazine. In case the compound is an amino acid, the reagent used is ninhydrin reagent, which give violent coloured spots.
- (v) Calculation of  $R_f$  values

The  $R_f$  values (known as the retention factor or ratio of fronts) provide a useful index for comparing two compounds but these values are not always reproducible.

$$R_f = \frac{\text{Distance travelled by the compound}}{\text{Distance travelled by the solvent}}$$
 $R_f \text{ of compound } A = x/z, R_f \text{ of } B = y/z, \text{ (Fig. 4.6)}$ 
 $x = \text{Distance moved by compound } A$ 

y =Distance travelled by B

z =Distance travelled by the solvent.

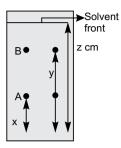


Fig. 4.6 Distance travelled by the various components of the mixture

**Identification of compounds:** The identity of the two compounds is checked by spotting the two compounds on the same plate side by side and following the procedure as described above. Identical compounds have same  $R_f$ 

#### 4.4 PREPARATIVE TLC

This technique is used to separate and isolate the components of a mixture in pure state. The TLC plates used for separation are larger in size  $(20 \times 20 \text{ cm})$  and the adsorbent layer is also thick (about 1mm). For separation, the mixture (about 2g) is dissolved in a suitable solvent and spotted on the plate as a band [Fig. 4.7 (a)]. The spotted plate is dried and placed in a glass jar containing a suitable solvent [Fig. 4.7(b)]. The solvent rises as explained earlier. After the solvent has risen almost to the top of the plate, it is taken out, solvent allowed to dry. If the bands are coloured Fig. 4.8(a), further development is not necessary. However, if the mixture is colourless, the bands are located by covering the preparative plate by a glass plate and spraying about 1 cm on the side of the plate with a suitable reagent [Fig. 4.8(b)]. The developed chromatogram is shown in [Fig. (4.8 (a)].

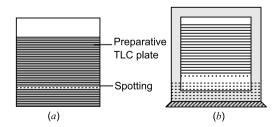


Fig. 4.7 Preparative TLC

For isolation of the components of the mixture, each of the bands in the TLC plates are scraped (with the help of a fine spatula). Each scraped material is

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extracted with a suitable solvent and the component isolated by evaporation of the solvent. In this way each component is isolated individually and weighed.

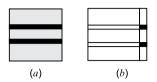


Fig. 4.8 Developed chromatogram

#### Notes:

- 1. The Jar containing the solvent should be saturated with the vapours of the solvent. This is achieved by keeping the solvent in the Jar for about 30 min before the experiment.
- 2. The TLC plates should not touch the sides of the Jar.
- 3. Normally silica gel or aluminate are commonly used as adsorbents. However, the nature of stationary phase may be modified for special needs. For example, silica gel may be modified as follows:
  - Silica gel with a binder like plaster of paris, starch
  - Silica gel with a fluorescent agent
  - Acidic silica gel obtained by the addition of very dilute sulphuric acid, oxalic during the preparation of TLC plate
  - Basic silica gel abtained by the addition of a small amount of KOH during the preparation of TLC places.
- 4. The silica gel for TLC is 100–200 mesh.

After the isolation of each fraction, its structure is determined by spectrophotometric procedures.

#### 4.5 TWO-DIMENSIONAL TLC

It is found that in some TLC procedures, using different types of solvent systems, it is not possible to get a good separation of a mixture. Such problem is overcome in two-dimensional TLC. In this procedure a square glass plate ( $10 \text{ cm} \times 10 \text{ cm}$ ) coated with a suitable adsorbent is used. The TLC plate is spotted with the solution of the mixture in a suitable solvent near the one corner of the plate [Fig. 4.9(a)]. The chromatogram is run in a solvent as usual. After the chromatogram is comple [Fig. 4.9(b)] the plate is removed from the tank. It is dried, rotated through  $90^{\circ}$  so that the separated row of spots become the starting line [Fig. 4.9(c)]. Subsequently, the chromatogram is rerun using another solvent system. The end result [Fig. 4.9(d) is effective separation of the components. Thus, using two-dimensional TLC good separations of even closely related substances can be achieved.

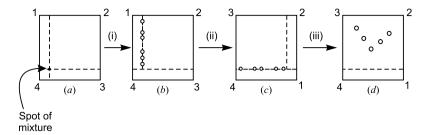


Fig. 4.9 Two-dimensional TLC

- (a) Spotted plate (spot put near one corner)
- (b) Chromatogram run in a solvent [step(i)]
  - (c) Plate rotated through 90° [step (ii)]
- (d) Chromatogram rerun in a different solvent [step (iii)]

The corners of the TLC plates are mark as 1, 2, 3, 4.

Two dimensional chromatography can also be performed on Filter paper (size  $10 \text{ cm} \times 10 \text{ cm}$ ).

# 4.6 HIGH-PERFORMANCETHIN-LAYERCHROMATOGRAPHY (HPTLC)

This technique is useful for the separations which are very difficult using other types of TLC. The main features of HPTLC are given below:

- (i) Adsorbent layer: Specially purified silica gel having average particle diameter to 3–5 μm are used. Layers of HPTLC are prepared using the above mentioned silica gel. The layers prepared using improved adsorbents give up to 4000–5000 theoretical plates and provide much better results than usual TLC. Using this, difficult separations can be effected; the separations take much shorter time.
- (ii) Amount of sample: Since the sample capacity of HPTLC layer is lower less amount of sample is required. The sample volume is of the order 100–200 nL in order to get starting spots of 1.0-1.5 mm diameter. Compact separation of spots are obtained after developing the plate for a distance of 3–6 cm.
  - For spotting the TLC plates, the spotting device is a platinum irridium capillary of fixed volume (100 or 200 nL) is used. Also the tip of the capillary is polished to provide a smooth and planar surface of a very small area (about 0.05 mm<sup>2</sup>).
- (iii) Detection of components in HPTLC: Detection of the spots in nanogram range can be affected with uv absorption and in the picogram range with fluorimetric detection. The detection can also be achieved by densitometers. In this procedure, the chromatogram is scanned by a

moving beam of light and the intensity of the light reflected from the surface is measured. The areas of peaks correspond to the amount of materials in the spots.

# 4.7 REVERSED PHASE PARTITION THIN LAYER CHROMATOGRAPHY

In this technique, the stationary phase is non-polar and the mobile phase is polar. This technique finds application for the fractionation of non-polar petroleum products, fats, waxes, carotenoids, steroids and fact soluble vitamins and dyes.

The layers in reversed phase partition TLC are prepared by impregnating thin layers to silicic acid Kielselguhr with non-polar liquids. These layers (which serve as stationary phase) are of paraffin oil (on silicic acid) and silicone.

Usually, the solvent used in reversed phase partition TLC is  $CHCl_3-CH_3OH-H_2O$  (75:20:15), acetone–water (3:2) and acetic acid water (3:1).

#### 4.8 THIN LAYER CHROMATOGRAPHIC SEPARATIONS

### 4.8.1 Separation and Identification of Amino Acids by TLC

Let us suppose for the sake of convenience that the mixture of amino acids contains DL-alanine, L-leucine and L-lysine hydrochloride.

- (i) Prepare solutions of DL alanine, L-leucine and L-lysine hydrochloride by dissolving each of the amino acids (5 mg each) separately in distilled water (0.33 mL). If necessary the solution be warmed to get a clear solutions.
- (ii) Mix one drop of each of the three solution (prepared above) to get a mixture of the three amino acids. The remaining solution of the three amino acids is made up to 1mL with distilled water.
- (iii) Spot the TLC plates (silica gel G plate) with the solutions of the three amino acids and one spot of the mixture of the three amino acids.
- (iv) The spotted air dried plate is inserted in a Jar in the developing solvent [propan-1-ol-conc. aqueous ammonia (d.0.88) (70 mL : 30 mL)]. Adequate precaution is taken to make sure that only the adsorbent of the TLC plates dips in the solvent and not the spots.
- (v) When the solvent front has reached near the top of the plate, the TLC plate is removed, solvent front marked with pencil and the plate is dried at 100°C (10 min). It is then sprayed with ninhydrin reagent (0.2 per cent solution in butan–1–ol and heated the plate at 110°C (5–10 mm) so that the colour of the spots is developed. Mark the centre of each spot with a metallic pin and calculate the  $R_f$  values for each amino acid and record. **Precaution**: Contact of ninhydrin reagent with skin is avoided since it produces long lasting purple discoloration.

### 4.8.2 Separation and Identification of Carbohydrates by TLC

Let us suppose that the mixture carbohydrates contain fructose, lactose, glucose, maltose and sucrose.

Following procedure is followed:

- (i) Prepare solution of individual carbohydrates (fructose, lactose, glucose, maltose and sucrose) in distilled water (0.01 to 0.2% solution). Mix 0.5 mL of each of the above solutions to get a mixture of the carbohydrates.
- (ii) Spot the TLC plate (Kieselgel G plate) with the five carbohydrate solution and another spot of the mixture.
- (iii) The air dried spotted TLC plate is inserted into a Jar having the developing solvent [acetone-butanol- water (50:40:10)]. The spots should not dip in the solvent, only the adsorbent layer of the TLC plate dips in the solvent.
- (iv) After the solvent front has reached near the top of the TLC plate, it is removed, solvent front marked and plate dried in air.
- (v) Spray the TLC plate will spray reagent (20 per cent aqueous trichloroethanoic acid and 0.2 per cent ethanolic solution of 1, 3-dihydroxynaphthalene).
- (v) Calculate the  $R_f$  value of each carbohydrates and record. The  $R_f$  value is in the order fructose, lactose, glocose, maltose and sucrose.

## 4.8.3 Separation and Identification of Ketones

Let us suppose with the mixture of ketones contains acetone, butan-2-one and hexan-3-one.

- (i) The mixtue of ketones is converted into 2,4-dinitraphenyl hydrazones. Also each of the 3 ketones is separately converted into 2, 4-dinitrophenyl hydrazone.
- (ii) Spot the TLC plate (silica gel) with a solution of each of the three 2,4-dinitrophenyl hydrazone in 0.5 mL ethyl acetate. Also put an additional spot of mixture of 2,4-dinitrophenyl hydrazone in ethyl acetate.
- (iii) The air dried spotted TLC plate is inserted into a Jar having the developing solvent (toluene-light petroleum (b.p. 40-60°C) 3:1).
- (iv) After the solvent front has reached near the top of the TLC plate, it is removed, solvent front marked and plate dried in air.
- (v) The 2,4-dinitrophenyl hydrazones appeared as yellow-orange spots. Calculate the  $R_f$  value of each spot and record.

# **EXERCISES**

- **1.** What do you understand by Thin Layer Chromatography? What is its principle?
- 2. Explain the procedure for TLC. How is TLC plate developed?
- **3.** What is preparative TLC? What are its uses?
- **4.** Explain two dimensional TLC? What are its uses?
- 5. What is high performance TLC? How does it differ from usual TLC?
- **6.** Explain reversed phase partition TLC.
- 7. Is TLC superior to paper chromatography? If so explain.





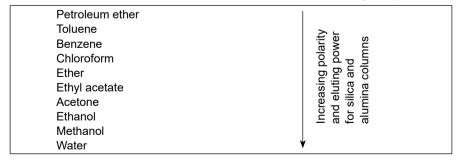
Chapter 5

## **Column Chromatography**

In column chromatography, the stationary phase is a solid and the mobile phase is a liquid. The stationary phase (known as adsorbent) is packed in a glass tube, which is called the column and hence the name column chromatography. Some of the common adsorbents used are alumina (Al<sub>2</sub>O<sub>3</sub>, available as neutral, acidic or basic alumina), silica gel, charcoal, calcium carbonate, starch etc.

The mobile phase (known as eluting solvent) plays an important role is the separation of the components of the mixture. The eluting solvent should be less polar than the components of the mixture, which should, of course be soluble in the moving phase. In case the components are insoluble in moving phase its separation is not possible as the components remain adsorbed at the top of the column and do not move alongwith the mobile phase. A number of solvents are used for chromatographic separations. First, the less polar solvent is used in order to elute the least strongly adsorbed component of the mixture. Subsequently, solvents with increasing polarity and greater eluting power are used for elution of more strongly adsorbed components. Some of the common solvents that are used in column chromatography are given in Table 5.1. A mixture of solvents can also be used for elution.

Table 5.1 Common solvents used in column chromatography



#### 5.1 PRINCIPLE OF COLUMN CHROMATOGRAPHY

The principle of column chromatrograpy is selective adsorption followed by elution. The mixture to be separated is dissolved in a suitable solvent and allowed to pass through a glass column containing the adsorbent. The various components of the mixture are adsorbed in a particular order. Thus, the component which has greatest adsorbing power is adsorbed in the upper part of the column followed by the component with lesser adsorbing power and so on. The net result is that the materials are partially separated in different parts of the column. As the column is eluted with a suitable solvent, the various bands in the column become more clear in different zones. The different zones can be cut (after pushing the adsorbent) or elution continued to get the different components of the mixture. The separation of pure components can be ascertained by TLC.

#### 5.2 PROCEDURE OF COLUMN CHROMATOGRAPHY

The column chromatographic technique consists of the following steps:

(i) Packing of the Column with a Suitable Adsorbent: A column is a long glass tube having a stopper at the end [Fig. 5.1 (a)]. The size of the column depends on the amount of the mixture to be separated. Better separations are achieved by using long thin columns than short wide columns. A small amount of the eluting solvent is added into the dry, clean column after a small ball of cotton or glass wool is pushed near the stoppered end of the tube using a long glass rod. The column is clamped, stopper opened [to allow the solvent flow into a conical flask placed below the column (Fig. 5.1 (b)]. A slurry of the adsorbent and a least polar eluting solvent is added continuously from the top of the column [Fig 5.1 (c)]. The column is frequently tapped gently with a piece of rubber during the addition of slurry. This helps the air bubbles get out of the column.

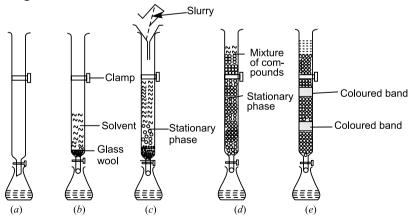


Fig. 5.1 Apparatus used for column chromatography

The column should be evenly packed. After the addition of the required amount of slurry (normally two-third of the column is packed with the adsorbent), the excess solvent is allowed to drain out leaving a small amount of solvent at the top of the solid adsorbent. The stopper is finally closed and the flow of the solvent stopped.

- (ii) Addition of Sample Mixture to the Column: A solution of the mixture (solid or liquid) is dissolved in minimum amount of a solvent and added carefully just on the top of the adsorbent without disturbing the adsorbent. The excess solvent is allowed to drain out, until the liquid lavel is just above the adsorbent [Fig. 5.1 (d)].
- (iii) Elution of the Column: The eluting solvent is added continuously from the top of the column and the eluted solvent is collected in a conical flask.

Continuous addition of the solvent can be done using a separatory funnel, the stem of which dips in the solvent layer on the top of the column.

In case the components of the mixture are coloured, the coloured bands separate out (as the solvent moves) and start moving at different rates [Fig. 5.1(e)]. When the fastest moving coloured band reaches the stoppered end of the column, the eluate is collected in a separate conical flask. After all the colour of the band has been eluted, the component is recovered by distilling the solvent. The residual product on crystallization give one component in pure state. In a similar way subsequent coloured bands are eluted.

If the mixture is colourless, their movement in the column cannot be visualised. In such cases, small fractions of the eluate are collected in separate conical flasks. Fractions containing same compound (as seen by TLC examination of each fraction) are mixed together and distilled to get the product. For collection of different fractions, automatic fraction collector can be used.

#### Notes:

- 1. The stop cock in the column should not be greased.
- 2. The slurry of the adsorbent is stirred continuously white being added to the column.
- 3. The packed column should not contain any air. This can be achieved by gently tapping the column during packing.
- 4. The elution of the column should first be carried out with the least polar solvent and then by systematically increasing the polarity of the solvent. This can be accomplished by not changing the solvent entirely

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but by using mixed solvent by addition of a small percentage of a more polar solvent to the non-polar solvent. The amount of polar solvent can be increased subsequently.

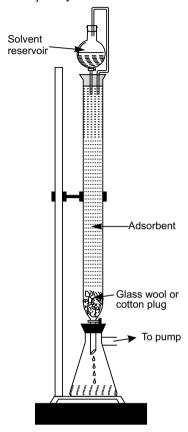


Fig. 5.2 Collection of eluate using small pressure

- 5. In case, the rate of elution becomes very slow (due to so many factors beyond our control), the rate can be increased by application small suction in the collector vessel (Fig. 5.2).
- 6. Under no circumstances the columns be allowed to dry.

#### 5.3 HIGH PERFORMANCE COLUMN CHROMATOGRAPHY

We have seen (section 5.1) that in usual procedure of column chromatography, the liquid phase (eluting solvent) passes through the stationary phase (adsorbent) under the influence of gravity and the overall process is slow. However, high performance or high pressure liquid chromatography (HPLC) is very efficient and gives excellent separation in much shorter time. In this technique, the stationary phase used is of much smaller diameter and high surface area and the liquid phase passing through

the column is by using a pump at a high pressure. By this procedure the speed and versality of column chromatography increases.

In HPLC, the efficiency is much higher than even in TLC.

Though gas chromatography (GC) is also a high performance method, but it can be used for only those substances which are volatile or can be evaporated intact at higher temperatures. However, HPLC can be used for high molecular weight compounds efficiently even at room temperature.

In HPLC, a stainless steel tube (20 cm length, internal diameter 5 mm) is used as a column. The packing material is of uniform, smaller diameter and high surface area. The pressure of the eluting solvent is maintained at 6000 PSL using a suitable pumping system. The sample is injected in the column using a loop type of system, which is connected to the main solvent tube.

HPLC finds applications both in analytical technique and also for preparative purposes.

The detectors are located at the exit from the column. The components in the eluate can be identified with the peaks on the detector recording and can also be separately collected.

#### 5.4 DRY COLUMN CHROMATOGRAPHY

This technique was developed by Loev, Marjorie and Good man (Chem. Ind., 1967, 2026). In this method alumina ( $100-200~\mu m$ ; activity II or III, with incorporated fluorescent indicator) or silica gel ( $100-250~\mu m$ ; activity III, with incorporated fluorescent indicator is packed dry (1g adsorbate : 300~g adsorbent) into a glass column (2-5~cm diameter), which is fitted with an adapter incorporating a sintered disc (Fig 5.3).

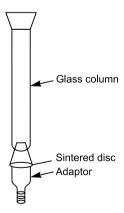


Fig. 5.3 Glass column for dry column chromatography

In place of glass column, Nylon tubing (2-5 cm diameter), which is sealed at the lower end and into which is placed glass wool. In case of glass tube, the packing

of the adsorbent is faciliated by using an ultra-vibrator along the tube. During packing and development of the chromatogram, the tap in the assembly is kept open.

The liquid or solid mixture is dissolved in a suitable volatile solvent like ether, petroleum ether, methylene chloride to which is added adsorbent (about five times the weight of the mixture). The solvent is evaporated and the solid material loaded on to the packed column. Finally, the column is covered with acid-washed sand (0.5 to 1 cm depth).

The column is developed by using a suitable solvent (using a separatory funnel). The development of the column is complete when the solvent front reaches the bottom of the column (20 - 30 min).

Finally, the adsorbent is extruded from the glass assembly. The zones of the components of the mixture are detected by the usual procedures. These zones are cut (using a blade) and extracted with suitable solvent to get individual components.

In case nylon column is used, it is laid horizontally on a sheet of glass, the nylon covering slit lengthwise with a blade. Subsequent steps are same as stated above.

#### 5.5 CHIRAL CHROMATOGRAPHY

We come across a number of biologically important substances that exist as enantiomeric pairs (optical isomers). These enantiomers have identical chemical and physical properties but have different biological properties in medical applications. A typical example of such a compound is thalidomide, which was used as anti-nausea drug for expectant mothers in Great Britain and Europe (1959 – 62). This drug had to be withdrawn since it caused catastropic birth defects to children born to women who took thalidomide during pregnancy. Investigations revealed that this drug was used as a mixture of its two enantiomers and each of these stereoisomers had different biological activity. Although one enantiomer had the desired therapeutic effect. The other isomer was responsible for the birth defects.

In view of the above, it is essential that all chiral drugs be subjected to chiral analysis (separation and identification of the two enantiomers). The technique used for this is called chiral chromatography.

The chiral phase (used as stationary phase) was first developed by W.H. Pickle and T.C. Pochapsy (Chem. Rev., 1989, 89, 347) and is known on Pickle packings.

These were obtained by bonding a asymmetric organic chain to the surface of the conventional silica packing. Though such stationary phase was effective for the separation of enantiomers, these could be used only with THF-hexane or isopropylalchol-hexane as the eluant. Such a stationary phase is not very effective for the separation of hydrophilic materials.

The most commonly used stationary phase, which finds wide applications is human serum albumin (HSA), which is bonded to silica gel. Such stationary phase utilises the affinity of only one enantiomer (J. Hermansson, J. Chromatog,

1984, 316, 537). This is a special form of affinity chromatography and is used for the separation of optically active materials. HSA is temperature sensitive (like all proteins) but is stable at pH 6-8. Separations are unially carried out using phosphate buffer (0.05M) at pH7 using 1-propanol or acetonitrile up to a maximum of 10-15 per cent as organic modifier. Such columns are effective in chiral separations of neutral and acidic materials, particularly drugs.

Thus, using such a column  $(4.6 \text{ mm} \times 150 \text{ mm})$  of chiral HSA and mobile phase (0.05 M) phosphate buffer, pH 6.9 and acetonitrile in 9 : 1 mixture) enantiomeric separation of a benzodiazepine Temazepam could be affected; in this case the UV decrector was set at 254 nm.

#### 5.6 COLUMNS CHROMATOGRAPHIC SEPARATIONS

# 5.6.1 Separation and Identification of a Mixture of o-Nitroaniline and p-Nitroaniline by Column Chromatography

Stationary phase Alumina, packed in a column using benzene

Mobile phase Benzene

Procedure See section 5.2

*o*-Nitroaniline is eluted out first and then *p*-nitroaniline. Since both the components are coloured, the movement of band can be seen. The two compounds cna be confirmed by their IR spectra and NMR spectra. Superimposible IR spectra with authentic samples can also be used to establish their identify.

# 5.6.2 Separation and Identification of a Mixture of *cis*- and *trans*-azobenzene by Column Chromatography

Stationary phase Alumina, packed in a column using petroleum either.

Mobile phase (i) Petroleum either

(ii) Petroleum ether-methanol (95 : 5)

Procedure See section 5.2

*cis*-azobenzene (orange band) is eluted first using petroleum ether-and *trans*-azobenzene (yellow band) is eluted subsequently using petroleum ether-methanol (95:5)

Both the compounds can be identified by their m.ps. and UV spectra.

cis azobenzene, m.p. 71.5°C

 $\lambda_{max}^{\textit{EtOH}}$  281 nm ( $\epsilon$  5260)

trans azobenzene

 $\lambda_{max}^{\textit{EtOH}}$  320 nm ( $\epsilon$  21300)

### 5.6.3 Purification of Anthracene by Column Chromatography

Commercial anthracene contains carbazole and naphthalene as impurity. Anthracene is purified as follows:

- (i) A column of activated alumina (50g) using hexane is prepared. The glass column used is about 60 cm. long and 1.8 cm diameter. The length of activated alumina is about 20 cm.
- (ii) To the column is a added a solution of commercial anthracene (50 mg) in hexane (50 mL). The solution is yellow in colour.
- (iii) Eluate the column with hexane (200 mL)
- (iv) Examine the column with light of an UV lamp.

A deep blue fluorescent zone due to carbazole is seen at the top of the column.

The next zone is a yellow non-fluorescent zone due to naphthalene.

Anthracene appears as a broad, blue-violet fluorescent zone in the lower part of the column.

(v) Continue elution of the column with hexane till anthracene is eluted out completely. The eluate is concentrated to about 2mL and cooled. Anthracent separates out m.p. 215–216°C.

In the final stage of elution the eluate used is a mixture of hexanebenzene (1:1). This hastens the process.



- 1. What is column chromatography? Discuss the principal involved.
- 2. Describe the procedure involved in column chromatography.
- **3.** What is high performance column chromatography? How does it differ from gas chromatography?
- **4.** Write a note on (i) Dry column chromatography, (ii) Chiral chromatography.
- **5.** How will you purify commercial anthracene?
- **6.** What are the applications of column chromatography?





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Chapter 6

## **Gas Chromatography**

#### 6.1 INTRODUCTION

In gas chromatography, the moving phase is a gas, which passes over a stationary phase. In a way, it is similar to column chromatography. The stationary phase can be a solid or a liquid. Thus, the separation can be adsorption or partition. In case the adsorbent is solid, the chromatography is called gas-solid chromatography (GSC). On the other hand, if the adsorbent is liquid, the chromatography is called gas-liquid chromatography (GLC). Both types are now called gas chromatography (GC).

The partition chromatography was invented by Archer John Porter and Richard Laurence Millington Synge. This is a method used for the separation of organic compounds, especially amino acids obtained by hydrolysis of proteins. They also introduced improved methods of gas chromatography, a process for separation of the constituents of various gaseous mixtures. For their work, they were awarded Nobel Prize in Chemistry in 1952.

There has been considerable improvements in gas chromatography. It is now possible to separate very small amounts of complex mixtures containing up to 200 related compounds using partition or adsorption. A limitation in gas chromatography is that it can be used for only volatile materials Thus, non-polar materials are easy to separate than polar materials. However, ionic materials cannot pass through a gas chromatographic column.

#### 6.2 PRINCIPLE OF GAS CHROMATOGRAPHY

In gas chromatography, the compounds (mixtures) are separated by partitioning process between the moving gas phase (inert gas like nitrogen or helium) and a stationary liquid phase. The apparatus used for gas-liquid chromatography is

called a gas chromatograph (Fig. 6.1). The mixture of compounds to be separated is injected into the chromatograph and is immediately vapourised. The vapourised sample is swept into the column by the aid of carrier gas. The column is packed with a solid, finely divided substance, the surface of which is coated with a liquid (stationary phase) of relatively low volatility. The column is placed in an oven, whose temperature is controlled by a thermostat and heating elements. In the column, the mixture gets separated into individual bands, which passes through the detector. The detector produces an electronic signal, whose voltage is proportional to the amount of the material present in the gas stream. A plot of the voltage as a function at time is plotted by the recorder. This plot is called a chromatogram (Fig. 6.2).

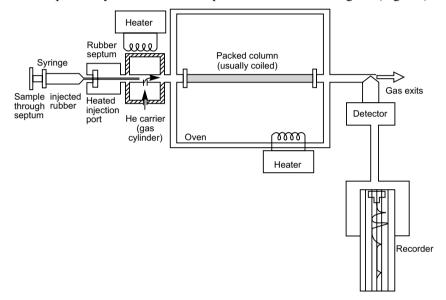


Fig. 6.1 Schematic diagram at a gas chromatograph

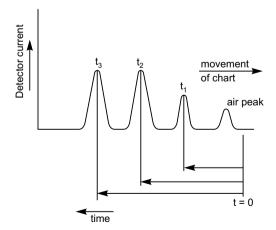


Fig. 6.2 Gas Chromatogram

In gas chromatography, under a given set of conditions, the retention time of a compound is the property of that compound and may be used to identify it. Also, the voltage output of the detector is proportional to the mole fraction of the components being detected in the vapour, the relative areas under two peaks of the chromatogram is equal to their percentage in the mixture [Figs. 6.2 and 6.3).

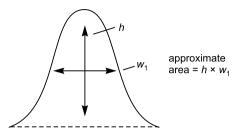


Fig. 6.3 Peak area

#### 6.3 THE CHROMATOGRAPHIC INSTRUMENT

A chromatographic assembly consists of the following components.

#### 6.3.1 Carrier Gas

Commonly used carrier gas include hydrogen, helium and nitrogen. Hydrogen is inflammable and can also react with unsaturated compounds. Helium is used as carrier gas due to its good thermal conductivity and low density. Nitrogen, though cheap gives reduced sensitivity. It should be ascertained that the carrier gas is pure. Normally, a flow rate of 300 – 900 mL min<sup>-1</sup> is maintained.

### 6.3.2 Sample Injection System

A number of devices are available. A common procedure is to use a microsyringe with hypodermic needle, and inserting through a silicone rubber septum into a heated metal block at the beginning of the column. The temperature of the metal block should be such that the liquid is immediately vapourised without getting decomposed. The temperature of the port is normally set near the boiling point of the least volatile compound.

Compounds having high molecular mass being non-volatile cannot be put into the injection system. This difficulty can be overcome by breaking the large molecules into smaller fragments, which are volatile and so can be analysed. This process is commonly known as **pyrolysis gas chromatography** (PGC). Using this technique (carried out in the absence of oxygen), the decomposition products are separated in the gas chromatographic column. The resulting chromatogram (called pyrogram) are used for qualitative and quantitative analysis of the fragments. This procedure of PGC finds application in the analysis of polymers. As an alternative to this process, the high molecular weigh compounds can be pyrolysed first and then injected into the system.

Some compounds, which have high polarity, low volatility or thermal stability are unsuitable for direct injection in the gas chromatograph. This problem has been overcome by converting the compound into their trimehtylsilyl derivatives and then subjecting to gas chromatographic analysis.

It is a common knowledge that inorganic compounds are not as volatite as organic compounds. However, some inorganic compounds which possess the necessary properties can be subjected to gas chromatographic analysis. Gas chromatography has also been used for the separation and quantitative analysis of metals. For this purpose, the metals are converted into metal chelates, which are volatile and so can be subjected to gas chromatographic analysis. As an illustration,  $\beta$ -diketones ligands, *e.g.*, acetyl acetone and its fluorinated derivatives, trifluoroacetyl acetone (TFA) and hexafluoroacetyl acetone (HFA) form volatile and stable chelates with metals like beryllium, aluminium, chromium (III) etc. Thus, it is possible to chromatograph a number of metals as their  $\beta$ -diketone chelates.

The chelates can be extracted with a suitable solvent (like benzene) and then the solution injected into the gas chromatographic column.

#### 6.3.3 The Column

The efficiency of gas chromatographic separations depends of the type of the column, the nature of the solid support, type of liquid phase, packing procedure, length of the column and the temperature employed. Following are given some common types of column which can be used:

- (i) Packed Clumns: The columns used in the earlier developments of gas chromatography were about 5 m in length, 2 4 mm internal diameter and made of glass, metal (stainless steel, aluminium, copper etc.) or high temperature plastic (PTFE). The columns were evenly packed with inert material [like diatomaceous earth, which had been washed and deactivated (by acid treatment) and sieved to get mesh size 60 120], which was uniformly coated with a suitable liquid phase (1 5 percent) (A number of liquids phases are available, which are grouped into non-polar, intermediate polarity and polar). The selection of the liquid phase depends on the nature of the mixture to be separated. A useful guide is 'like separates like'. Some of the examples of different types of liquid phases are given below:
  - (a) Non-polar hydrocarbon-type liquid phase. Examples include paraffin oil (Nujol), Apiezon [Grease silicone-gum rubber (can be used up to 400°C)].

- (b) Liquids phases of intermediate polarity. Examples include esters of high molecular weight alcohols like dinonyl phthalate. Such compounds possess a polar group attached to a large non-polar skeleton.
- (c) Polar compounds: Examples include carbowaxes (polyglycols).
- (d) Compounds possessing a number of H atoms. These are referred to hydrogen-bonding class. Examples include glycol, glycerol and hydroxy acids.
- (ii) Capilliary Columns: Most of the present day instruments have capilliary columns. These columns are made of glass, stainless steel, fused silica or quartz and are coated with a stationary phase as a thin coherent film on the inside surface of a long thin (capilliary) tube. The packed capilliary column can be purchased. The choice of the column is very important. Two types of capillary columns are available.
  - Wall coated open tubular (WCOT) columns. In these columns, the stationary phasor is directly coated on the inner wall of the capilliary tube.
  - Support-coated open tubular (SCOT) columns. These have a finely divided layer of solid support on the inner walls of the cpailliary tube. The stationary phase is then coated on the solid support.

The type of column to be selected depends on the type of the mixture to be separated. In fact, 'Rules of thumb' for column selection is followed. The same, general concept of like separating like is also followed for capillary columns. For example, non-polar materials can be separated on a non-polar phase and polar material on polar phase. The longer the column, better is the resolution. Normally, 25m column is satisfactory. Also, the column efficiency increases considerably with decrease in the internal diameter of the capilliary tube. Best separation are effected by using capilliary tube column of  $100\mu m$  (0.1 mm). For better resolution, the film thickness in the capilliary column should be about  $1\mu m$ . Film thickness more than  $1 \mu m$  leads to bleeding of the column when in use.

### Porous layer open tubular (PLOT) columns

The packed column and capillary columns so far described used only liquid stationary phases. In these columns, as has already been stated, the separation of mixture is by partition mechanism. However, it is now possible to procure porous layer open tubular (PLOT) columns. In these columns a very this layer  $(5-50\mu\text{m})$  of solid adsorbent is evenly deposited on the inside wall of the capaillary tube. The solid adsorbents are similar to that used for packed column adsorption chromatography. The only difference is that in PLOT columns the adsorbents have much smaller size.

PLOT columns containing molecular sieve, alumina, carbosieve (a kind of carbon molecular sieve) are easily available. These columns give good separation of permanent gases or the  $\rm C_1 - \rm C_5$  hydrocarbons. A draw back in such columns is that the coatings are not mechanically stable and so more care is required for using them.

#### 6.3.4 The Detector

A suitable detector is situated at the exit of the column. Its function is to sense and measure small amounts of separated components in the carrier gas stream leaving the column. The information from the detector is fed to a device which produces a trace called a chromatogram. The choice of the detector depends on factors like concentration level of the separated component and also on the nature of the separated component. A number of different type of dectors are available. These include hot-wire dector (HWD), Flame ionisation detector, electron capture detector, photoionisation detector (PID), atomic emission detector (AED) and differential thermal conductivity detector. Integrated detectors are also available; these give signals proportional to the amount of the eluted component. Most of the detectors are commercially available.

### 6.3.5 Temperature Programming

Normally in gas chromatograms, the temperature of the column is kept at a constant temperature. This is referred to isothermal mode of operation. A main disadvantage in such chromatograms, the resolution of the components is poor and the peaks are broad and widely separated. Also, compounds having high boiling points may not be detected. All these problems can be overcome by using programmed temperature gas chromatography (PTGC). In such a chromatogram, the temperature of the whole column is raised during the analysis of the sample. In this the temperature programme involves changes in column temperatures, which can be selected by a microprocessor controller. In this technique, there is an initial isothermal period, a linear temperature rise segment followed by a final isothermal period. The final isothermal period temperature may vary depending on the separation to be effected.

Using programmed temperature gas chromatography, it is possible to separate compounds of wide boiling range more rapidly compared to isothermal operation of the column. Also sharp peaks are obtained so that heights of the peaks can be used to obtain accurate quantitative analysis.

#### 6.4 PREPARATIVE GAS CHROMATOGRAPHY

It is a very useful technique for an organic chemist. In this procedure packed column (1-2 cm in diameter and 12-20 ft length) (packed with suitable adsorbents are used. Samples (up to 100 mL) can be convently used. Mixtures of compound that are not difficult to separate can be separated easily using such procedures. The compounds as they leave the column are collected using suitable cooled traps. The compounds obtained by this procedure are ultrapure.

Commercial gas chromatographs are now available for the separation of mixtures. The procedure finds use in the preparation of pharmaceutical drugs and other fine chemicals.

#### 6.5 APPLICATIONS OF GAS CHROMATOGRAPHY

(i) Separation of a mixture of compounds and identification of the various components.

As already stated (section 6.2, Fig. 6.2), in gas chromatography, the retention time of a particular compound is the property characteristic of that compound that can be used to identify it. Retention time is the time which lapses between the injection of the sample and its appearence at the detector. In place of retention time, the distance on the chromatogram from the starting of the separation to the peak of a particular substance can also be used for identification of a particular constituent in a mixture.

An authentic procedure for the identification of various components is an follows. Suppose a peak in a chromatogram is attributed to 2–hexanone. This can be confirmed by mixing the mixture with a pure speciman of 2–hexanone. The peack, which becomes much larger in height and size is that of 2–hexanone. In a similar way the other peaks can also be identified.

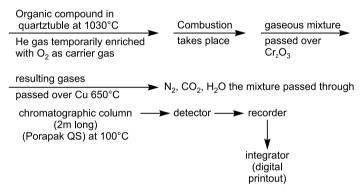
Final confirmation of individual components is done by spectral analysis for each components. For this purpose, the GC instrument is attached to an IR instrument, UV spectroscopy instrument, NMR instrument or a mass spectrometer. In other words, each fraction as it leaves the column is analysed using various spectral techniques. The information gathered is helpful for the final confirmation of the various constituents.

- (ii) Analysis of Polymers: Polymers can be analysed by pyrolysis gas chromatography (PGC). For details see section 6.3.2. This procedure is also useful for the analysis of compounds having high molecular mass and are not volatile.
- (iii) Quantitative Analysis of Metals: Some metals can be estimated by converting them into their chelates, which are volatile and so can be subjected to gas chromatographic analysis. The β-diketones, e.g., acetylacetone and its fluorinated derivatives like fluroacetylacetone (TFA) and hexafluoro acetylacetone (HFA) are used as chelating agents. Metals like beryllium, aluminium, chromium etc. can be analysed by this procedure (see also section 6.3.2).
- (iv) Elemental Analysis: A most important application of gas chromatography is the estimation of carbon, hydrogen, nitrogen, oxygen and sulphur in organic compounds. Such an analysis is extremely helpful to final the molecular formula of an organic compound.

#### Estimation of Carbon, Hydrogen and Nitrogen

The procedure consist in dropping a weight amount (about 1 mg) of a organic compound contained in a dry tin container (a tin foil can be used) into a vertical quertz tube which is heated at 1030°C and through which a stream of helium gas temporarily enriched with oxygen is passed. Thus, the compound is combusted. The mixture of gases thus obtained is passed over  $Cr_2O_3$  (to achive quantitiative combustion) and then over copper at 650°C. By this procedure excess oxygen is removed and the oxides of nitrogen are reduced to nitrogen ( $N_2$ ). The mixture of gases ( $N_2$ ,  $CO_2$ ,  $H_2O$ ) are passed through a chromatographic column (about 2m long) of Parapark QS (heated to about 100°C). By this procedure, the individual components ( $N_2$ ,  $CO_2$ ,  $H_2O$ ) are separated and passed on to conductivity detector and potentiometric recorder in parallel with an integrator and digital printout. It is necessary to calibrate the instrument by combustion of a standard compound like cyclohexanone-2, 4-dinitrophenyl hydrazone.

The procedure is summarised in scheme 6.1. Now ready made chromatographs are available incorporating the above procedure for the estimation of C, H and N.



Scheme 6.1 Estimation of C, H and N)

For the analysis of water, it is necessary to determine total organic carbon (TOC) is order to monitor the quality of water, which gives an indication of the total organic pollutants. The procedure consists in acidifying a sample of water and purging to remove  $CO_2$  from any carbonate or hydrogen carbonate present. After this treatment, a small amount of measured volume of water is injected into the gas stream and passed through a heated packed tube where the organic matter is oxidised to  $CO_2$ , which is determined by IR absorption or converted into  $CH_4$  and determined by gas chromatography using a flame ionization detector.

### **Estimation of oxygen**

For the estimation of oxygen, a weighed amount of organic compound (weighed in a clean dry silver container) is dropped into a reactor (heated at 1060°C). The oxygen is quantitatively converted into CO by passing over a layer of nickel-coated

carbon. The resultant gases flow into the chromatographic column (about 1 m long) packed with molecular sieves (5  $\times$  10<sup>-8</sup> cm) heated to 100°C. The CO is separated from  $N_2$ ,  $CH_4$  and  $H_2$  and is measured using a thermal conductivity detector. The decomposition of the oxygen containing compound can be enhanced by addition of vapour of a chlorohydrocarbon.

#### **Estimation of Sulphur**

The procedure for the estimation of sulphur is essentially similar to that used for the estimation of C, H and N. The sulphur is oxidised to SO<sub>2</sub> by passing the combustion gases over tungston (VI) oxide WO<sub>3</sub>. Excess oxygen is removed by passing the gases through a heated tube containing copper. Finally the mixture of gases is passed through a Porapak chromatograhic column (heated at 80°C). Here SO<sub>2</sub> is separated from other gases of combustion and measured by a thermal conductivity detector.

#### **Miscellaneous Applications**

Besides the applications mentioned above, the GC is also used for the following:

- Detection of steroid drugs (used by sports persons).
- Detection of hazardous pollutants like CO<sub>2</sub>, formaldehyde, benzene, acrylonitrile, trichloroethylene etc.
- Analysis of dairy products for the presence of aldehydes and ketones which are responsible for rancidity.
- Analysis of bood, urine and stomach excreta.
- Analysis of polyaromatic hydrocarbons, chlorinated pesticides (e.g., DDH, BHC), organophosphorus and organic sulphur compounds, phenols, amines, organo lead and organomercury compounds

#### 6.6 GAS CHROMATOGRAPHIC SEPARATIONS

#### 6.6.1 Estimation of Sucrose

Since sucrose decomposes at the high temperature in the chromatographic column, it is converted into its trimethyl silyl derivative, which being volatile can analysed by GC.

#### **Procedure**

All reagents should be pure and dry.

Sucrose (10 mg) is treated with anyhdrous pyridine (1 mL) (pyridine is purified by refluxing over KOH followed by distillation; it is stored over dry KOH), hexamethyl disilazane (HMDS, Me $_3$  Si–NH–Si Me $_3$ ) (0.2 mL) and trimethyl chlorosilane (TMCS, Me $_3$  SiCl) (0.1 mL) in a plastic stopper vial. The mixture is shaken vigorously (30 – 45 seconds), allowed to stand at room temperature (10 min). In case, sucrose still remains insoluble in the reaction mixture, the vial is warmed at 75 – 80°C for about 2 – 3 min. The resulting solution (0.3  $\mu$ L) is injected in the gas chromatograph and analysed by the usual procedure.

#### 6.6.2 Estimation of Aluminium in Water

Aluminium is estimated as its tris (acetylacetonato) complex.

The aquous solution (5 mL) containing aluminium (about 15 mg mL) is adjusted to pH 4 – 6 and equilibrated (10 min) with two successive 5 mL solution of pure distilled acetylactone – CHCl $_3$  (1:1). The organic extract is combined. The organic extract (0.30  $\mu$ L) is introduced into the gas chromatograph and analysed.

The chromatograph is equipped with a flame isonisation detector and data handling system. Pure  $N_2$  ( $O_2$  free) (flow rate 40 mL min<sup>-1</sup>) is used as a carrier gas. The chromatographic column is made of glass (length 1.6 m and out side diameter 6 mm) is packed with 5 wt percent SE 30 on carbomosob W as the stationary phase and is maintained at 165°C.

**Note:** The presence of fluoride ion interferes with the extraction of aluminium with acetylacetone –CHCl<sub>2</sub>. It must be removed.

The GC apparatus is calibrated as follows: A series of aqueous aluminium solutions (containing 5-25 mg Al in 5 mL) is extracted by the procedure described above.  $0.30~\mu\text{L}$  of the extract is injected into the chromatographic column and peak area on the chromatogram recorded. The procedure is repeated thrice with different concetrations of aluminium. A graph is plotted of peak area against concentration.

The amount of aluminium (present as its acetylacetonate) in the sample solution is determined by injecting  $0.30~\mu L$  into the column. The peak area is recorded and aluminium concentration determined from the calibration curve. The above procedure was developed by R.H. Hill and H. Gisser (J. Gas Chromatography, 1963, 1, 11).



- 1. What is gas chromatography? Discuss the principle involved.
- 2. Describe gas chromatography assembly and its various components.
- **3.** How will you confirm the structure of a compound using gas chromatography?
- **4.** Write a note on:
  - (a) Pyrolysis gas chromatograhy
  - (b) Separation of metals by GC.
  - (c) Porous layer open-tubular (PLOT) columns.
  - (d) Temperature programming in GC.
  - (e) Preparative gas chromatography.
- 5. Using GC how will you estimate C, H, N, O and S in organic compounds.
- **6.** How will you determine total organic carbon (TOC) in a water sample?
- 7. How will you estimate aluminium in water sample?
- **8.** Describe briefly the applications of GC.





Chapter 7

# High Performance Liquid Chromatography (HPLC)

#### 7.1 INTRODUCTION

We have known (Chapter 6) that gas chromatography can be used only for compounds which can be easily volatilized. However, a larger number of organic compounds are not sufficiently volatile and also decomposed under the gas chromatographic conditions (like high temperature of the column). Such compounds can be analyzed or separated by high performance liquid chromatography (HPLC), which is considered to be rival gas chromatography in performance. Using HPLC, separations can be achieved in a matter of few minutes. The successful separations in HPLC is attributed to small uniform particles to give small eddy diffusion and rapid mass transfer.

HPLC is used widely in pharmaceutical industry. As of date, the HPLC market for new instruments is much more than the GC market.

#### 7.2 PRINCIPLE OF HPLC

Conventional liquid chromatography is accomplished in large columns packed with large adsorbent particles; solvent for elution moved under the force of gravity and the fractions of eluants collected individually for measurements in a spectrophotometer. The performance of liquid chromatography increases considerably by using small particles under increased flow pressure; by this procedure, theoretically high plate numbers could be achieved. It was only in the 1970s that small silanized silica particles were produced to give high resolution performance. Fig. 7.1 illustrates schematic diagram of an HPLC components.

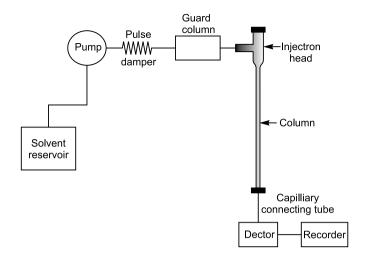


Fig. 7.1 Schematic representation of HPLC components

The column in HPLC is packed finity divided adsorbent, which has high spherical regularity for allowing optimum homogenity and packing density. Also the stationary phase should be in the form of a thin uniform film.

#### 7.3 HPLC INSTRUMENTS

A HPLC instrument consists of the following components.

#### 7.3.1 Mobile Phase

The choice of the solvent depends on the nature of the compound be separated. The solvent reservoirs are filled with different solvents of different polarities. HPLC grade solvents are used. These are degassed (made free of air bubbles) and filtered through  $0.2~\mu m$  filters. In some HPLC instruments on line vacuum degassing systems are incorporated. The solvent is pumped to the columns using a suitable pump.

# 7.3.2 Sample Injection Systems

Special injection systems are available for injecting a few microlitres of the sample solution at pressure up to 6000 psc'. Samples can also introduced manually into the valve with a micro syringe.

#### **7.3.3 Column**

Straight columns of stainless steel (diameter 3.9 or 4.6 mm) are the best. The length of the column depends on the amount and nature of the compound to be separated. The adsorbent are particles of high purity silica (low in trace metal content)  $(5-10\mu\text{m})$  in diameter). For high speed HPLC, diameter of silica about  $3\mu\text{m}$  is used. The pore size of the adsorbent is in the range 60 to 100Å. Liquid stationary

phases are coated on the particles or are chemically bonded. A typical well-packed 4.6 mm column of  $5 \mu \text{m}$  diameter particles give a plate count of the order of 60,000 to 90,000 plates at a flow rate of 1 mL/min. Though wide bore short columns are used for fast chromatography, but narrow columns give better resolution.

A small guard column (3 to 10 cm) is placed between the injector and the main column. The guard column contains the same adsorbent as the main column. It is helpful in retaining pump seal fragments and particulate matter.

#### 7.3.4 Detector

In HPLC detectors with high sensitivity are used. Some of such detectors include refractometer detector and UV detectors. The former, *i.e.*, differential refractometer detector is called 'universal detector'. It measures changes in refractive index of the eluant as it leaves the column. The UV detector has much better sensitivity (about  $10^{-8}$  g/mL, 0.01 ppm).



- 1. What do you understand by HPLC? On what principle does it work?
- 2. Describe schematically an HPLC instruments and the various components.
- 3. What type of column and adsorbent is used in HPLC?





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Chapter 8

# **Gel Chromatography**

#### 8.1 INTRODUCTION

It is also known as **molecular chromatography** or **size exclusion chromatography**. In this type of chromatography, molecular sieve is used as a stationary phase. The size of the adsorbent is of the order 50-3000Å. The organic molecules penetrate the pores of the adsorbent according to their size. The net result is that there is a difference in the rates at which different molecules pass down the column. This technique is also called **Gel Filtration** or **molecular sieve filtration** and can be used in open column (gravity form for preparative separations) and also in high performance separations.

#### 8.2 PRINCIPLE OF GEL CHROMATOGRAPHY

The sieves used in gel chromatography are usually polymeric materials (carbohydrates and acrylates). These are hydrophilic and can easily absorb water resulting in swelling of the sieves. The size of the 'holes' in the swelled sieves depend on the degree of crosslinking in the polymeric materials. Larger molecules which are larger than the pores of the swollen gel are incapable of penetration in the gel particles and so pass through the column *via* the spaces between the individual particles. On the other hand, smaller molecules penetrate to a varying degree depending on their size and shpae. Such molecules will be eluted in order of decreasing molecular size. In general for the separation of high molecular-weight substances gels with a high degree of swelling are used and for the separation of low molecular weight substances gels (denser gels) with lesser degree of swelling are used.

#### 8.3 TYPES OF GELS

Before use the gels are equilibrated for few hours (5 - 24 hrs) with the solvent to be used. Longer periods of soaking is necessary for loose crosslinking gels which

are used for high molecular weight substances. Different types of gels are available. These include sephadex, biogel and styragel.

Sephadex is the name given to a popular molecular-sieve, which is used for the separation of proteins. It is a polymeric carbohydrate substance and contains hydroxyl groups along the polymer matrix. It absorbs water, since it is fairly polar. Different types of sephadex gels are available depending on their swelling ability with water. As an example, sephadex G-10 can absorb 1mL water per g dry gel and sephadex G-200 can absorb 20 mL water per gram. These sephadex gels are insoluble in water and are stable in weak bases and acids (see Table 8.1).

Bio-gel is an polyacrylamide gel and is chemically more inert. These gels are soluble in common organic solvents and water and are used in the pH range 2 to 11. In the case of bio-gel, the adsorption is variable resulting in changes in chromatographic behaviour (Table 8.1).

A polystyrene gel, called stragel, is useful for non-aqueous separations in solvents like methylene chloride, toluene, trichlorobenzene, THF, cresol, DMSO etc. It cannot be used with water, alcohols or acetone. For such gets, the exclusion limit is molecular weight 1000 to 40 million.

Each gel has an exclusion limit. Exclusion limit refers to the molecular weight of that molecule which can just permeate the gel and so can be retarded. Depending of the type of the gel, the exclusion limit can vary from 1000 to several million. The separations are based on the molecule's size and configuration. Normally, molecules having smaller molecular mass than the exclusion limit can be fractionated.

Sephadex Type	Functional Range for peptides and globular proteins (MW)	Bio-gel Type	Fractionation range (MW)
G – 10	∠ 700	P – 2	100 – 1,800
G – 15	700 – 1500	P – 4	800 – 4,000
G – 25	1,000 – 5,000	P-6	1,000 – 6,000
G – 50	1,500 – 30, 000	P – 10	1,500 – 20,000
G – 75	3,000 – 70,000	P – 30	2,500 – 40,000
G – 100	4,000 – 1,50,000	P – 60	3,000 – 60,000
G – 150	5,000 – 4,00,000	P – 100	5,000 — 1,00,000
G – 200	5,000 - 8,00,000	P – 150	15,000 – 1,50,000
		P – 200	30,000 – 2,00,000
		P – 300	60,000 - 4,00,000

Table 8.1 Fractionating range of sephadex and biogels

#### 8.4 APPLICATIONS OF GEL CHROMATOGRAPHY

The important applications of gel chromatography are

- Determination of molecular weight distribution of polymers.
- Separation and purification of proteins, enzymes, peptiles, nucleic acids, hormones, polysaccharides etc.
- Determination of molecular weight of proteins.



- 1. What is gel chromatography explaining the principle involved?
- 2. What types of gels are used in gel chromatography?
- 3. Discuss the applications of gel chromatography.





Chapter 9

# Ion Exchange Chromatography

#### 9.1 INTRODUCTION

Various types of chromatographic techniques which have been discussed so far are used basically for the separation of organic substances. Ion-exchange chromatography is useful for the separation of inorganic anions and cations. This type of chromatography is based on the exchange of ions in the stationary phase. A special use of ion chromatography is in the separation of amino acids, since these contain both cationic and anionic components.

The stationary phase in ion chromatography consists of beads of polymer, e.g., a polystyrene polymer cross-linked with divinyl benzene. The resin (cross-linked polymer) having a free phenyl group attached to a chain can be suitably treated to add ionic functional groups. In analystical chemistry, four types of ion exchange resins (two cation exchange and two anion exchange) are used. (see Table 9.1). As in the case of gel chromatography (Chapter 8), ion exchange chromatography can also be used in open tube packed column (gravity form) and also in high performance procedure.

 Ion Exchanger
 Functional group
 Examples (Trade names)

 Anion Exchanger
 Quaternary ammonium
 Dowex<sup>a</sup>.1, Amerlite<sup>b</sup> IRA400, Ionac<sup>c</sup> AGA, 542 Rexgn<sup>d</sup> 201; Permutite<sup>e</sup>S1

Table 9.1 Ion exchange resins

Ion Exchanger	Functional group	Examples (Trade names)
Weak base	Amine group	Dowex-3, Amberlite IR 45, Ionac AGA – 316 Rexyn 203, Permutit-W
Cation Exchanger		
Strong acid	Sulfonic acid group	Dowex <sup>a</sup> 50, Amberlite <sup>b</sup> IR 120, Ionac <sup>c</sup> CGC-240, Rexyn <sup>d</sup> 101, Permutit <sup>e</sup> Q.
Weak acid	Carboxyl group	Amberlite IRC 50, Ionac CGC-270, Rexyn 102, permutit H-70

**Note:** The super scrips *a*, *b*, *c*, *d* and *e* refer to trade names of Dow chemical company, Mallinckrodt chemical works, J.T. Baker chemical company, Fisher Scientific company and Matteson coleman and Bell respectively.

#### 9.2 DIFFERENT TYPES OF RESINS

Typical examples of some ion exchange and their method of preparation are given below:

### 9.2.1 Anion Exchange Resins

As already stated (Table 9.1), the anion exchange resins contain basic groups on the resin, in which the hydroxyl anion is exchanged with other anions. These can be strongly basic (contain quaternary ammonium groups) and weak basic groups (contain amine group). These are represented as

n-Resin/
$$\mathring{N}$$
R $_3$ O $\overset{+}{H}$  + A $^{^{\leftarrow}}$  anion exchange resin

Resin/ $\mathring{N}$ H $_3$ O $\overset{+}{H}$  + A $^{^{\leftarrow}}$  anion exchange resin

(Resin/NH $_3$ ) $_3$ A + nOH $^{-}$  anion resin

The use of strong-base exchangers and weak base exchangers can be used over different pH range (0 - 12 for strong base exchangers and 0 to 9 for weak base exchangers). The weak base exchangers will not remove very weak acids but are used for strong acids.

$$\begin{array}{c} \text{CH}_2 & \text{CH}-\text{CH}_2\text{CI} + \text{NH}_3 \\ \text{epichlorohydrin} & \begin{array}{c} \text{Conduction} \\ \text{CH}_3 - \text{CHOH}-\text{CH}_2... \\ \\ \text{CH}_2 - \text{CHOH}-\text{CH}_2 - \text{N}-\text{CH}_2 - \text{CHOH}-\text{CH}_2... \\ \\ \text{...N}-\text{CH}_2 \text{ CHOH}-\text{CH}_2 - \text{N}... \\ \\ \text{CH}_2 - \text{CHOH}-\text{CH}_2 - \text{N}... \\ \\ \text{CH}_2 - \text{CHOH}-\text{CH}_2... \\ \\ \text{an anion exchange resin} \end{array}$$

## 9.2.2 Cation Exchange Resins

Cation exchange resins contain sulfonic acid as the functional group (Table 9.1). The weak acid cation exchangers contain  $-CO_2H$  group as the functional group. The protons on these groups ( $SO_3H$  or  $CO_2H$ ) can be exchanged with other cations.

n Resin/SO<sub>3</sub> H<sup>+</sup> + M<sup>n+</sup> 
$$\longrightarrow$$
 (Resin/SO<sub>3</sub>)<sub>n</sub> M + nH<sup>+</sup>  
n Resin/CO<sub>2</sub> H<sup>+</sup> + M<sup>n+</sup>  $\longrightarrow$  (Resin/CO<sub>2</sub>)<sub>3</sub> M + nH<sup>+</sup>

Usually, the cation exchange resins are available in hydrogen ion form. These can be converted into sodium form by treating with a sodium salt. The sodium ion subsequently undergo exchange with other cations. The strong acid cation exchangers can be used in the pH range 1 to 14, but weak-acid cation exchange resins can be used in the pH range 5 to 14

#### 9.3 PRINCIPLE OF ION CHROMATOGRAPHY

The principle of ion chromatography is based on the fact that different ions (cations or anions) have different capacity to undergo exchange on the ion exchanger. The extent of ion exchange has been found to depend on the size and charge of the hydrated ion in solution. The exchange capacity has been found to increase by the valency of the ion but decreases with the increase in the size of the hydrated ion. In fact ionic charge plays an important role in determination of their capacity to undergo exchange reaction. On this basis the capacity of following cation for exchange is of the order

$$Th^{4+} > A1^{3+} > Ca^{2+} > Na^{+}$$

In a similar way the capacity of the following anions (of same size) decreases in the order

$$PO_4^{3-} > SO_4^{2-} > I^-$$

For ions carrying the same charge, the exchange depends on the size of the hydrated ions. As an example, for univalent cations, the capacity of exchange is of the order

$$H^{+} > Rb^{+} > K^{+} > NH^{+}_{4} > Na^{+} > Li^{+}_{4}$$

For divalent cations, the capacity is found to be of the order

$$Ba^{2+} > Pb^{2+} > Sr^{2+} > Ca^{2+} > Ni^{2+} > Cu^{2+} > Co^{2+} > Zn^{2+} > Mg^{2+}$$

Amongst univalent anions, the capacity is the order:

$$I^- > NO_3^- > Br^- > CN^- > Cl^- > OH^- > F^-$$

#### 9.4 PROCEDURE FOR ION CHROMATOGRAPHY

As already stated, the ion chromatography involves separaton of the ions of a mixture. This is attributed to the difference in selectivity coefficients for the resin. This leads to different migration rates of ions on an ion exchange column. The apparatus used consists of a glass column fitted with a glass wool plug or a sintered glass disc at the lower end of the tube (Fig. 9.1)

The resin to be used should be of small particle size. A slurry of the resin (in distilled water) in made. Any fine particles suspended are removed by decantation. The slurry is poured slowly is the column containing some water. Adequate precautions be taken to exclude any air bubbles in the column. This is ensured by 'back washing' the column with distilled water or deionised water. Back washing is carried out by running water up from the bottom of the column. The excess water is then drained off.

Different fractions are collected and the presence of ion detected by conductivity detector. The use of conductivity detector is limited due to high background conductance (millimhos) of the eluting agent. This problem is overcome by using a second ion exchange

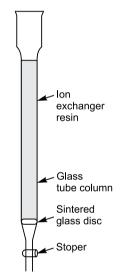


Fig. 9.1 Ion exchange column

column (called the suppressor column). For anion analysis, the suppressor column is the acid form and for cation analysis the suppressor column is the base form. For ion chromatography, weak exchange resins are normally used.

The overall separation procedure using two columns is explained as follows:

Let us suppose the salt MA of an anion  $A^-$  is to be separated on an anion exchange resin and elution carried out with NaOH. The eluting from the first chromatographic column will be a mixture of MA and NaOH. On passing this mixture through suppressor column containing cation exchange resin, the following reaction takes place.

```
Resin/SO_3^-H^+ + M^+ + A^- → Resin/SO_3^-M^+ + H^+ + A^- cation exchange resin
Resin/SO_3^-H^+ + Na^+ + OH^- → Resin/SO_3^-Na^+ + H_2O
```

In the above procedure NaOH is converted to  $\rm H_2O$  and the analyte ion is converted into the corresponding acid HA, which is detected by the conductivity detector.

In a similar way, for the cation analyte, a mixture of MA and HCl from the first column react in the suppressor anion exchange column as shown below:

```
Resin/N<sup>+</sup> R<sub>3</sub>OH<sup>-</sup> + M<sup>+</sup> + A<sup>-</sup> \rightarrow Resin/NR<sub>3</sub><sup>+</sup> A<sup>-</sup> + M<sup>+</sup> + OH<sup>-</sup> anion exchange resin
Resin/N<sup>+</sup> R<sub>3</sub>OH<sup>-</sup> + H<sup>+</sup> + Cl<sup>-</sup> \rightarrow Resin/NR<sub>3</sub><sup>+</sup> Cl<sup>-</sup> + H<sub>2</sub>O
```

In this case HCl is converted into H<sub>2</sub>O and the analyte cation is converted into the corresponding base MOH, which is detected by conductivity detector.

The anion analysis and cation analysis are digramatically represented as shown in Fig. 9.2.

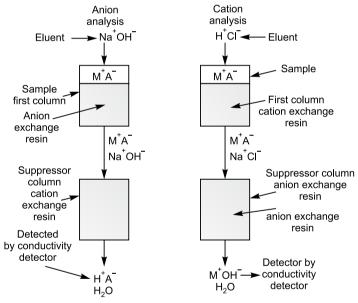


Fig. 9.2 Schematic representative of anion and cation analysis

The resin in the suppressor column gets exhausted and has to be regenerated (with NaOH for anion exchanger and with HCl for cation exchanger).

#### 9.5 APPLICATIONS OF ION CHROMATOGRAPHY

## 9.5.1 Determination of Anions

The elution is carried out with a mixture of Na HCO<sub>3</sub> and Na<sub>2</sub>CO<sub>3</sub>. By this procedure the anions are converted into low-conducting carbonic acid. Using this procedure,

anions like F<sup>-</sup>, Cl<sup>-</sup>, Br<sup>-</sup>, I<sup>-</sup>, NO $_2^-$ , NO $_3^-$ , SO $_4^{2-}$ , PO $_4^{3-}$ , SCN<sup>-</sup>, IO $_3^-$ , and ClO $_4^-$  can be determined in few minutes at parts per million.

Fig. 9.3 shows a typical anion analysis using ion chromatography. Such analysis is extremely difficult to perform by other methods.

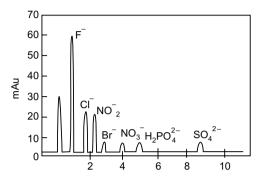


Fig. 9.3 Ion chromatography of some anions

## 9.5.2 Separation of Li<sup>+</sup>, Na<sup>+</sup> and K<sup>+</sup> ions

Similar ions like Li<sup>+</sup>, Na<sup>+</sup> and K<sup>+</sup> can be separated by passing their solution through a cation exchanger. The column is subsequently eluted with 0.1 N HCl.

## 9.5.3 Removal of Phosphate (Interferening Radical)

Phosphate is known to interfere during the estimation of  $Ca^{2+}$  or  $Ba^{2+}$  by the oxalate or sulphate method. Removal of phosphate is achieved by passing a solution of  $Ca^{2+}$  and  $Ba^{2+}$  containing  $PO_4^{3-}$  through a sulfonic acid cation exchanger. By this procedure  $Ca^{2+}$  and  $Ba^{2+}$  get exchanged with  $H^+$  but  $PO_4^{3-}$  passes through the column. The process is repreated 2-3 times for complete removal of  $PO_4^{3-}$ .  $Ca^{2+}$  and  $Ba^{2+}$  are held by the resin and are removed by suitable eluant and estimated by the usual methods.

# 9.5.4 Softening of Hard Water

Hardness of water is due to the presence of Ca<sup>2+</sup>, Mg<sup>2+</sup> and other divalent ions. These ions are removed by passing hard water through a column packed with cation exchange resins (sodium form). Following reaction take place

Thus,  $Ca^{2+}$  and  $Mg^{2+}$  are retained by the resin and  $Na^+$  passes into the solution. The water so obtained is soft water (contains  $Na^+$  ions) and can be used for washing purposes.

With time, the resin gets exhausted. It can be revived by percolation of a conc. solution of NaCl when the following reaction takes place

#### 9.5.5 Demineralised Water

It is used in laboratories for all types of analytical work. The process used for demineralisation of water consist in passing the water first through a cation exchanger (which replaces cations with H<sup>+</sup>)

Subsequently the water is passed through an anion exchanger resin (which replaces anions with OH<sup>-</sup>)

The H<sup>+</sup> ions obtained in the cation exchange process combine with OH<sup>-</sup> ions formed in the anion exchange process to produce H<sub>2</sub>O

When the resins are exhausted, they are regenerated by treatment with acid (in case of exhausted cation exhange resin) or alkali (in case of exhausted ion exchange resin)

In practice, hard water is first passed through a chamber (column) containing cation exchange resin and subsequently through another chamber (column) containing anion-exchange resin. When the resins are exhausted, the supply of hard water is out off, stopper in between the two chamber closed and acid passed through the first column containing cation exchange resin and NaOH solution passed through the column containing anion exchange resin. After the regeneration is complete, both columns are washed with demineralised water. Subsequently hard water is passed to make it soft (mineral free). The process is diagramentally represented as shown in Fig. 9.4.

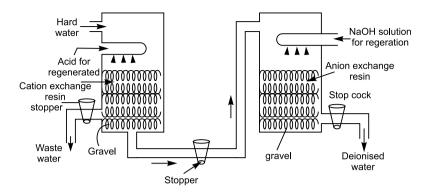


Fig. 9.4 Preparation of deionised water

## 9.5.6 Separation of Amino Acids

A mixture of amino acids is obtained by acidic or alkaline hydrolysis of proteins. The mixture can be separated by ion chromatography. Amino acids may be positively or negatively charge or neutral (isoelectric point) species. These three forms can be separated by a combination of cation and anion exchange resins.

$$R-CH-CO_{2}H \xrightarrow{+H^{+}} R-CH-CO_{2}^{-} \xrightarrow{-H^{+}} R-CH-CO_{2}^{-}$$

$$\downarrow \\ NH_{3}^{+} \qquad NH_{3}^{+} \qquad NH_{2}$$

$$(1) \qquad (2) \qquad (3)$$

Out of the three forms (1, 2 and 3) of amine acids, the form (2), called a Zwitter ion, is present as dominant form at a pH corresponding to the isoelectric point of the amino acid. The isoelectric point, as we know is the pH at which the net charge on the molecule is zero. In more acidic solution than the pH at the isoelectric form, the  $-CO_2^-$  gets protonated to form a cation (form 1), and in more alkaline solution, the  $-NH_3^+$  group looses a proton to form an anion (form 3). The isoelectric points are different for different amino acids. Thus, by the control of pH, it is possible to separate different amino acids.

The amino acids can be separated by passing through an anion and a cation exchange columns. The Zwitterionic form will pass through both columns, while the positively and negatively forms (1 and 3) will be retained by one of the columns.

Based on ion exchange columns, automatic amino acid analysers are commercially available. The resolution of each amino acid is recorded automatically by measuring the colour formed between the amino acid and ninhydrin as it is eluted from the column. A mixture of number of amino acids can be separated by operating at pressure of several hundred psi.



- 1. What is ion exchange chromatography? Discuss the principle involved.
- 2. What are anion exchange and cation exchange resins? How do they function?
- 3. Describe the procedure for ion chromatography.
- **4.** In ion exchange chromatography, the presence of ions is detected by conductivity detector. Such conductors do not give satisfactory results due to background conductance. How this problem is overcome?
- 5. Using ion exchange chromatography how will you detect the presence it various anions and cations in water?
- **6.** Give some interesting application of ion exchange chromatography.
- 7. How is demineralised water obtained?





Chapter 10

# **Electro Chromatography**

#### **10.1 INTRODUCTION**

Electro chromatography works on the principle of **electrophoresis**. Electrophoresis is a procedure of separation involving the migration of charged substances under the influence of electric current. In this technique, the movement of charged substances can be stopped anywhere along their path to the electrodes. However, in electrolysis, the charged species reach the electrodes. In this way electrophoresis differs from electrolysis.

The technique of electrophoresis is routinely used to separate substances based on their charge to mass ratios using the effect of electric field on the charge of these moieties. Using this technique charged colloidal particles or macromolecules including proteins, nucleic acid etc can be separated. Different types of electrophoresis techniques are available. The most common is the **zone electrophoresis**. In zone electrophoresis the macromolecules (*e.g.*, proteins) are supported on a solid. The separation depends not only on electric migration forces but also on conventional chromatographic forces. In view of this zone electrophoresis is also called **electro chromatography**. A number of supports can be used in zone electrophoresis. These include starch gels, polyarcrylamide gels, polyurethane foam and paper.

#### 10.2 PAPER ELECTROPHORESIS

In this technique paper is used as supporting medium. Only electrovalent compounds can be separated by this process. A solution of electrovalent compounds like NaCl, KNO<sub>3</sub>, BaCl<sub>2</sub> etc in water are dissociated into positive and negative ions. Due to the developed polarity, the ions migrate towards opposite electrodes.

In electrophoresis. Whatman filter paper is used (as in the case of paper chromatography). The filter paper is washed with 0.1 M HCl or 0.01 M EDTA solution and then with water. The usual size of paper used is  $1 \times 20$  cm. The electrodes used are carbon or platinum electrodes. Buffer solution containing weakly dissociated acids and bases is used as electrolyte.

Following procedure is followed for paper electrophoresis.

On a Whatman paper strip (1  $\times$  20 cm), a line is drawn perpendicular to the length of the strip. The stip is spotted with the sample (to be analysed) at positions marked on the line. A similar spot of a standard compound is also applied to the base line. The strip is then moistured with the electrolytic solution (a buffer solution which can affect separation) and placed in a horizontal position, the two ends of the strip dipping into the vessel of the same buffer solution (Fig. 10.1).

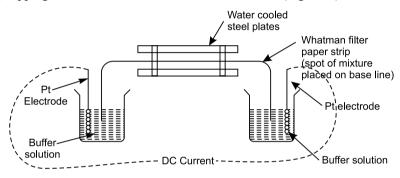


Fig. 10.1 Paper Electrophoresis technique

The Whatman strip is surrounded by steel tubes (water cooled) in order to prevent the evaporation of the electrolyte. On passing the current, the electrolyte on the strip acts as a bridge between the two containers of the electrolytic solution. The net result is the movement of the charged components towards anode or cathode. After some time (20-25 min) the paper strip (now called electrophoretogram) is removed and allowed to dry. The separated coloured components are visible on the strip and components which are colourless by spraying with a suitable reagent as in paper chromatography.

In place of Whatman filter paper strips, strips of cellulose acetate can also be used; the procedure is called **cellulose acetate electrophoresis**. The ability of cellulose acetate for the separation of very small amounts of substances makes it a valuable technique. Compared to Whatman paper, cellulose acetate is almost transparent, has more uniform structure and smaller pore size. This technique is well suited for clinical diagnosis like analysis of blood, serum protein, haemoglobins, enzymes, nucleic acids, alkaloids and drug extracts.

#### 10.3 GEL ELECTROPHORESIS

This technique is based on the differential electrophoretic mobilities of molecules (like proteins, amino acids etc.), which in turn are due to the difference in their

isoelectric points. The most commonly employed electrophoretic method is the SDS (Sodium Dodecyl Sulphate) gel electrophoresis. In this method, SDS—a surfactant denatures the protein and binds to it. Each molecule of the bonded SDS contributes a negative charge to the protein, different proteins acquire different charges depending on their size. These are separated in presence of electric field on the basis of charges on them.

A typical application of gel electrophoresis is the separation of amino acids. For example, a mixture of three amino acids (lysine, alanine and aspartic acid) spotteted in a gel buffered at pH 6.01 gets separated on application of electric potential at the ends. The schematic representation of the set-up is shown in Fig. 10.2.

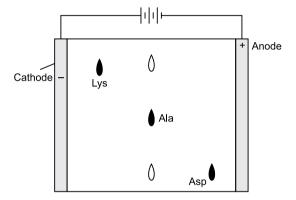


Fig. 10.2 Separation of a mixture of lysine, alanine and aspartic acid. The black spots indicate the position of lysine and aspartic acid before applying electric field. In case of alanine the position is the same before and after application of the field.

It is found that aspartic acid spot moves towards anode implying that this amino acid has a net negative charge. Similarly, the spot moving towards cathode is lysine having a net positive charge. The spot of alanine does not move indicating that the amino acid is at its isoelectric point, *i.e.*, it is the Zwitterionic form.

An improved version of gel electrophoresis is **capillary gel electrophoresis**. This trchnique is used for the separation of macromolecules based on the size. In this technique, the capillary is filled with a porous polymer gel. As the molecules move through the gel, the separation is affected; it is based on both the electrophilic mobility and molecular size. An improvement in the above process is to fill the capillary with a liquid gel matrix (pumpable gel solution like derivatized cellulose dissolved in a suitable buffer). This technique is used commonly for the separation of nucleotides in deoxyribonuclic acid (DNA) sequencing .

# 10.4 CAPILLARY ELECTROPHORESIS (CE)

It is a new separation technique, which in used for the separation of minute quantities in much shorter time with high resolution. The procedure uses a fused silica capillary

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(internal diameter 200-400  $\mu$ m) filled with an aqueous buffer electrolyte. The two ends of the capillary dip into containers of the electrolyte, one having the anode and the other having the cathode (Fig. 10.3).

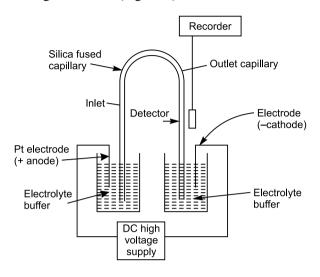


Fig. 10.3 Electrophoresis

The sample to be analysed is introduced by inserting anode end of the capillary into the vial containing the sample and then applying an electric field to the sample vial (electrokinetic injection). Alternatively, using a pressure on the vial to produce hydrodynamic injection procedure can be used. After a few nanolitres of the sample is in the capillary, the end of the capillary is introduced into an electrolyte container. Application of high voltage power supply (voltage between 20 and 100 kV) brings about the separation. Automatic sample injectors are now available for handling a large number of samples one after another. The nature of the elecrolyte (buffer solution) should be monitored constantly, since it is affected by the migration of solutes.

The most common detector employed is a uv absorbance detector.

The features of capillary electrophoresis and HPLC can be combined to give a hybrid technique; called **capillary electrochromatography (CEC)**. In this technique longer capillary is used and is filled with 1.5 to 3  $\mu$ m C<sub>18</sub> silica particles. A polar solvent mobile phase is driven by electrosmotic flow. The buffer generally contains 40 to 80 per cent of organic solvent like methanol or acetonitrile. The separation depends on the distribution of solutes between the mobile and stationary phase. The capillary electrochromatography exhibits more than twice the number of plates as in HPLC for the same size of the particles and same length of the column.

# **EXERCISES**

- 1. What is electrochromatography?
- 2. Describe paper electrophoresis technique.
- 3. Write notes on:
  - (a) Zone electrophoresis
  - (b) Gel electrophoresis
  - (c) Capillary electrophoresis
  - (d) Capillary electrochromatography.
- **4.** How will you separate and identify the components of an amino acid mixture containing lysine, alanine and aspartic acid?
- 5. Discuss some important application of electrochromatography.



# Part-III

#### **Thermal Methods**

- Thermogravimetric Analyses
- Differential Thermal Analysis
- Thermometric Titrations
- Miscellaneous Thermal Methods
- Derivative Thermogravimetric analysis
- Thermobarography
- Differential Scanning calorimetry
- Thermomechanical Analysis (TMA)
- Electric Thermal Analysis (ETA)



Chapter 11

# **Thermogravimetric Analysis**

#### 11.1 INTRODUCTION

Thermogravimetric analysis is a kind of thermal analysis. In thermal analysis, a substance is heated using a wide range of temperature. This results in the substance undergoing physical or chemical changes. All changes are accompanied by the absorption or release of energy in the form of heat. The changes may involve gain or loss of weight or changes in thermomechanical or electrical conductivity. These changes are characteristic of the material being examined. In fact, on the basis of the temperature at which such changes occur and the heat energy involved in the reaction, the compounds present in the material can be characterised. The term 'thermal analysis' includes a number of techniques in which some physical parameters of the system are recorded as a function of temperature. These techniques involve thermogravimetric analysis, differential thermal analysis, derivative thermogravimetric analysis, thermometric titrations (Table 11.1). The last technique, *viz.*, thermometric titration involves changes in temperature of the solution, which are plotted against volume of titrant added.

Table 11.1 Some Thermal Analysis Techniques

Technique (abbreviation)	Characteristic property measured	Instrument used	Graph plotted
Thermogravimetric analysis (TG)	Change in mass	Thermo balance	Mass versus temperature or time
Derivative thermo- gravimetric analysis (DTGA)	Rate of change of mass	Thermo balance	Rate of change of mass versus temperature

(Contd...)

Technique (abbreviation)	Characteristic property measured	Instrument used	Graph plotted
Differential thermal analysis (DTA)	Heat absorbed or evolved	DTA apparatus	Rate of change of temperature versus temperature
Calorimetric DTA	Heat absorbed or evolved	Calorimeter	DH/dt versus temperature
Thermometric titrations	Change of temperature	Titration calorimeter	Temperature versus titrant volume

#### 11.2 THERMOGRAVIMETRIC ANALYSIS

In this technique, the substance under investigation is weighed while it is being heated at a predetermined rate. The weighted substance is recorded as a function of time or temperature. There are three different types of thermogravimetric analysis. These are:

- (i) Isothermal or Static Thermogravimetry

  In this technique, the sample is heated at a constant temperature and its mass is recorded as a function of time.
- (ii) Quasistatic Thermogravimetry

  In this technique, the sample is heated to constant weight at each step in a series of increasing temperatures.
- (iii) Dynamic Tehrmogravimetry

  In this technique, the sample is heated at a uniform rate. With the advent of automatic recording instrument (1923) most of the studies are carried out using this technique.

#### 11.3 THERMOGRAVIMETRIC ANALYSER

A thermogravimetric analyser consists of a balance for measurement of weight of the sample, arrangement for heating and measurement of temperature, a cup for holding the sample and device for atmospheric control.

# 11.3.1 Measurement of Weight

The weight of the sample is determined using a thermobalance. A number of different types of thermobalances are available. These include beam type balance, helical type, centilever beam type, torsion wire type and null point balances; the later types are most commonly used. The basic requirement of a thermobalance is

- easy to operation
- easy response to changes in weight

- should be very sensitive and accurate
- should have good mechanical and electronic stability.

## 11.3.2 Heating Arrangement and Temperature Measurement

Different types of furnaces are available for purpose of heating the sample. The basic requirement is that there should be smooth heat rate so that the temperature could be either a linear heating mode (10°C to 500° C per hour) or a fixed temperature. Most of the instruments use the usual thermocouples or resistance thermometers. Commonly, platinum and tungsten windings are used. Temperatures as 1000°C or 1450°C are reached using nichrome or Pt-Rh windings.

The furnace size should be appropriate. Small furnaces cool rapidly and control of temperature is difficult. Larger furnaces maintain temperature and are used for linear heating rate. However, the required temperature takes more time to reach. Normally the heating rate is kept at 4-5°C per minute.

#### 11.3.3 Sample Holders

Depending on the nature, weight of the sample and maximum temperature range to be employed, the sample holder is chosen. Thus, in case volatile products are produced during heating, shallow pans are used. Such pans are used for the study of thermal behaviour of polymers. For calcinations and surface area studies, deep crucibles are used. Covered crucibles are used in case there is self-generated atmosphere. For boiling point studies, retro cups are used.

# 11.3.4 Atmospheric Control

In TGA, since there is weight changes in the sample due to the gaseous products formed during heating, atmospheric control is very necessary. A convenient way for maintaining atmospheric control is by flushing the balance with an inert gas. Helium is preferred due to its low thermal conductivity and very low density. The pressure of the gas must be regulated in TGA.

#### 11.3.5 Recorders

Different types of recorders are available. In automatic recorders (X-Y recoders), the curves having plot of weight directly against the temperatures are recorded.

A schematic diagram of a thermogravimetric analyser is given in Fig. 11.1.

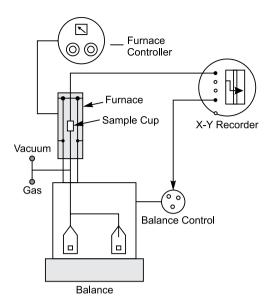


Fig. 11.1 A schematic representation of a thermogravimetric analyser

# 11.4 THERMOGRAVIMETRIC CURVE (TG CURVE)

An automatic recording (X-Y recorder) directly gives a plot of weight changes versus temperature or time. This is known as the thermogravimetric curve (TG curve). In this curve, the weight is plotted on the ordinate with weight decreasing downwards and temperature (T) or time (t) on the abscissa increasing from left to right (Fig. 11.2)

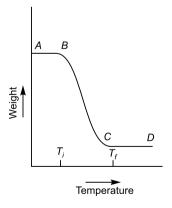


Fig. 11.2 A typical TG curve

In the TG curve, the horizontal portions (Plateaus) (curve AB) indicate that there is no weight change in this region. This is helpful to know about the thermal

stability of the material (Thermal stability is the ability of a substance to maintain its properties unchanged on heating). In Fig. 11.2 the curve AB is horizontal meaning that the compound is stable upto  $T_i$ °C

The curved portion of the TG curve (BC) in Fig. 11.2 indicate losses in weight. It is also possible to know that how much weight is lost by heating a particular sample to a given temperature. The temperature at which the completion of reaction is indicated is  $T_f$  (Fig.11.2). The difference between  $T_f$  and  $T_i$  is called the reaction interval.

The plot of weight against temperature in therogravimetric analysis is called a **thermogram**. The thermogram of calcium oxalate monohydrate is shown in Fig. 11.3. Loss of water occurs slightly above 100°C. A break is obtained (at about 250°C) in the curve at the stoichiometry corresponding to the anhydrous salt. Definite weight plateous for the carbonate are obtained on further heating (500°C to 600°C). The oxide is finally obtained at temperature above about 870°C. The exact locations of the weight plateous depend on the heating rate. Thus, a slower heating rate shifts values to lower temperature.

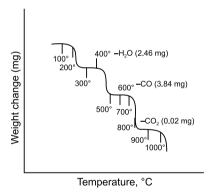


Fig. 11.3 Thermogram of calcium oxalate manohydrate (20 mg) heating rate 6°C/min)

Thermogravimetric analysis is also helpful to determine the loss in weight when a sample is heated to a fixed temperature. For example, CaCO<sub>3</sub> on heating at 850°C loses 44% of its weight. Also the gas can be collected and idenfied as CO<sub>2</sub>

$$CaCO_3 \rightarrow CaO + CO_2$$

# 11.4.1 Factors Affecting Thermogravimetric Curves

The thermogravimetric curves can be affected by the instrumental factors and the characteristic of the sample to be analysed. Deviations due to instrumental factors include the type of furnace (atmosphere, size and shape) sample holder, material of the sample holder and its resistance to corrosive attack, heating rate, response of the

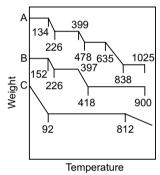
recording equipment and location of the thermocouples in the sample and reference chambers. The characteristics of the samples also affect the thermogravimetric curves. These characteristic include, thickness of the layer, particle size, packing density, amount of the sample, thermal conductivity of the material and heat capacity.

#### 11.5 APPLICATIONS OF TGA

TGA is mostly used in qualitative and quantitative studies and in thermal stability determinations. Following are given some of the important applications of TGA.

## 11.5.1 Determination of Thermal Stability of Salts

Using TGA curves it is possible to know the stabilities of salts. As an illustration TGA curves of calcium oxalate, magnesium oxalate and silver chromate are given in Fig. 11.4.



**Fig. 11.4** TGA curves of calcium oxalate monohydrate (A), magnesium oxalate dihydrate (B) and silver chromate (C)

As seen, TGA curve (A) of calcium oxalate monohydrate gives a number of platuex corresponding to monohydrate ( $100^{\circ}$ C), anhydrous calcium oxalate ( $226^{\circ}$ C to  $390^{\circ}$ C), calcium carbonate ( $460^{\circ}$ C to  $635^{\circ}$ C) and calcium oxide (> $840^{\circ}$ C). Thus, on heating to  $500 \pm 25^{\circ}$ C, the amount of CaCO $_{3}$  can be determined by heating a fixed amount of calcium oxalate monohydrate. In a similar way the TGA curve (B) of magnesium oxalate dihydrate revels that the up to  $160^{\circ}$ C, the dihydrate is stable and the anhydrous salt exist at  $220^{\circ}$ C to  $400^{\circ}$ C. The oxide is stable from  $480^{\circ}$ C to  $1000^{\circ}$ C. Therefore, the weighing at temperature > $480^{\circ}$ C gives the amount of oxide.

In the case of TGA curve (C) of silver chromate, it is found that up to 92°C, excess water is removal, the weight subsequently becomes constant and remains so up to 812°C. After 812°C, oxygen is lost to give silver. As seen, silver chromate is stable in the range 90-800°C. So in gravimetric analysis the precipitate of silver chromate can be dried in the region 90-800°C.

In a similar way, mercurous chromate (Hg<sub>2</sub>CrO<sub>4</sub>) is stable over the temperature range 52-256°C; this range is used to weigh the amount of mercurous chromate formed.

## 11.5.2 Analysis of Mixtures

TGA curves have also been used to analyse mixtures. As an example a mixture of calcium oxalate and magnesium oxalate can be analysed (refer to curves *A* and *B* in Fig. 11.4). As seen, there is significance difference between the oxalates of Ca and Mg. It is seen that magnesium oxalate does not pass through the carbonate stage.

$$CaC_2O_4 \rightarrow CaCO_3 + CO$$
 ...(i)

$$CaCO_3 \rightarrow CaO + CO_2$$
 ...(ii)

$$MgC_2O_4 \rightarrow MgO + CO + CO_2$$
 ...(iii)

Due to the difference in their thermal behaviour [equation (i), (ii) and (iii)], it is possible to simultaneously determine their amount by TGA. Thus, the mixture on ignition at 500°C (at which CaCO<sub>3</sub> and MgO are stable) while at 900°C, both metals exist as simple oxides. Finding the weight of a mixed precipitate at both the temperatures (500°C and 900°C) permits calculation of both Ca and Mg in the original mixture.

#### 11.5.3 Determination of Curie Temperature

Ferromagnetic substances (e.g., Fe<sub>2</sub>O<sub>3</sub>) show magnetism even in the absence of magnetic field. Such substances remain permanently magnetised, once they have been magnetised. In this type of magnetism there is spontaneous alignment of magnetic moments due to unpaired pair of electrons in the same direction (Fig. 11.5).

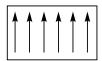


Fig. 11.5 Ferro magnetism

On heating, ferromagnetic solids change into paramagnetic (such substances are attracted by external magnetic field). For example,  $\mathrm{Fe_3O_4}$  on heating to 550°C becomes paramagnetic. This is due to greater alignment of spins in one direction on heating. Each ferromagnetic substance has a characteristic temperature above which no ferromagnetism is observed. This is known as curie temperature.

Ferromagnetic substance on keeping in a thermobalance with the poles of a magnet above the sample (Fig 11.6) the TG balance shows weight, which is less than the actual weight. The ferromagnetic substances loose their magnetism on heating. The temperature on which this happens is reproducible.

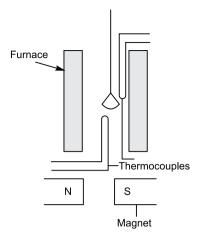


Fig. 11.6 Ferromagnetic material placed in pan of TG balance with magnet placed below the pan

A number of substances (metals or alloys) are available with curie temperatures between 150°C and 1000°C. If these substances are placed in the pan of TGA balance (as shown in Fig. 11.6) the sample experiences a downward attraction resulting in increase in weight. At the curie temperature, the loss of ferromagnetism is reflected by an apparent loss of weight (Fig. 11.7). This enables the determination of curie temperature for the particular substance.

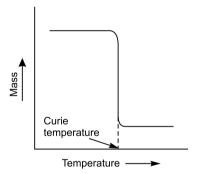


Fig. 11.7 Determination of curie temperature

This technique is used to study the effect of heat on ferromagnetic substances.

# 11.5.4 Organic Compounds

A number of organic compounds have been studied by thermogravimetric analysis. As an example, decomposition of malonic acid on subjecting to thermogravimetric analysis, a phase transition occured at about 70°C with melting at 140°C. This is followed by decomposition above 150°C. The whole process may be put as:

COOH 
$$CO_2 + CH_3COOH$$
  $COOH$ 

# EXERCISES ‡

- 1. What do you understand by Thermal analysis?
- 2. What are the important thermogravimetric methods?
- 3. What factors affect thermogravimetric analysis?
- **4.** Discuss the applications of thermogravimetric analysis.
- **5.** Describe the salient features of a thermogravimetric analysis.
- **6.** What is a thermogravimetric curve?
- 7. Explain the term curie temperature, What are its uses?





# Chapter 12

# **Differential Thermal Analysis**

#### 12.1 INTRODUCTION

Differential thermal analysis (DTA) is complementary to thermogravimetric analysis (TGA). However, DTA is much more versatile. The data obtained from DTA are of more fundamental in nature. In DTA, a smaple and a reference material are heated (or cooled) at a linear heating rate. The difference in temperature between the substance and the reference material is recorded against either time or temperature as the two specimens are subjected to identical heating rate. Using this technique, qualitative identification of a compound can be done by determination of the temperature at which endothermic or exothermic reaction takes place. A plot of the difference in sample and reference temperature (differential temperature,  $\Delta T$ ) as a function of time (t), sample temperature ( $T_s$ ), reference temperature ( $T_r$ ) or furnace tempemperature ( $T_r$ ) is known as a differential thermogram. A typical DTA curve is shown in Fig. 12.1.

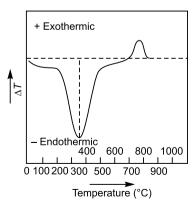


Fig. 12.1 A typical DTA Curve (Defferential thermogram)

Information about the nature of the test sample can be obtained from the shape and size of the peak. Normally, sharp endothermic peaks give ideas of fussion process or changes in crystallinity. On the other hand, broad endothermic peaks indicate dehydration reactions. It can be said that endothermic curves refer to physical changes and exothermic curves refer to chemical reactions.

#### 12.2 DIFFERENTIAL THERMAL ANALYSER

The instrument normally consists of the following parts:

- (i) Furnace: Its function is to heat the samples. Different types of furnaces are available. Usually, a tubular furnace is preferred. It is made of appropriate material wound on a retractory tube. The furnace to be selected should have good temperature regulation and programming.
- (ii) Sample Holder: The sample holder of various shapes and designs are available. These can be made of metallic (e.g., nickel, stainless steel (up to 1000°C) and platinum) and non-metallic materials (e.g., glass, silica, sintered alumina etc.). Normally, sample holder made of platinium is used
- (iii) Furnace Temperature Controller: For furnace temperature control, there are three basic elements. These are sensor, control element and heater. Normally, proportional controllers are used. In this the heat input to the system is progressively reduced as the temperature near the desired value. Besides these controllers, electronic controllers can also be used.

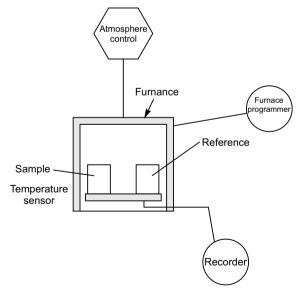


Fig. 12.2 Schematic diagram of a DTA apparatus

- (iv) **Recording Device:** These are the most sensitive part of DTA instrument. It should record the  $\Delta T$  as accurately as possible. Normally a recorder consists of a galvanometer; in this the reflected beam of light falls on a twin cathode photocell, the unbalanced beam is fed onto two thyratron tubes and the signal is recorded by a pen driver.
- (v) **Thermocouples:** In DTA instruments, the thermocouples are the temperature sensors. A suitable thermocouple is used.

A schematic diagram of a DTA instrument is shown in Fig. 12.2.

#### **Procedure for DTA Studies**

The powered sample (0.1 to 10 mg) (grounded to pass through a 100-mesh sieve) is taken in a sample tube (2 mm diameter) and placed in the sample holder. In another sample tube is taken an reference material (like alumina, quartz etc.) and placed in the reference holder. The furnace is heated uniformally at a constant rate and the temperature difference between the sample and reference material is continuously recorded against the reference material temperature.

The DTA analysis involves heating of the sample. Endothermic changes are more likely to occur than exothermic changes.

#### 12.3 FACTORS AFFECTING DTA

It is found that the differential thermograms are not always reproducible. This is due to Environmental factors, instrumental factors and characteristic of the sample.

Gaseous environment around the sample affects the DTA results. At times, the gaseous atmosphere may react with the sample to produce extra peaks in the DTA curves. As an example, the presence of oxygen in the atmosphere may cause oxidation reaction and give additional exothermic peak.

Since the atmosphere surrounding the sample is constantly changing in concentration due to convention currents, the results are not precise. Best results are obtained in dynamic atmosphere where the gases are swept across the sample in a controlled way. In case the swept gases contain one of the gaseous product, the decomposition is affected due to Le Chatelier effect.

Instrumental factors, which normally result in incorrect results include size and shape of the furnace, material of the sample holder, geometry of the sample holder, heating rate, location of the thermocouple (in sample chamber) and the sensitivity of the recorder.

DTA curves are also affected by the characteristic of the sample to be analysed. These characteristic include amount of the sample, particle size, packing density, degree of crystalinity, thermal conductivity and heat capacity. Besides these, swelling or shrinkage characteristic of the sample also affect the curves.

In order to get reproducible results, it is best to use the same instrument with all components of the necessary specifications. A standard DTA curve should be

obtained with a known sample and results compared with respect to unknown sample.

## 12.4 APPLICATIONS OF DTA

In DTA technique, any change in the substance (physical or chemical change) is indicated by a signal. Due to this DTA is more useful than TGA. Changes like melting point, crystallinity, specific heat, adsorption and also the chemical changes can be studied by DTA. Some of the important applications of DTA and described below:

#### 12.4.1 Heat of Reaction

It has been found (Borchardt and Daniels) that the area of a peak in DTA is invariably a linear function of the heat of a reaction, when the rate of heat of production (or consumption) is a function of time. The heat of the reaction,  $\Delta H$ , is given by

$$\Delta H = \frac{kA}{N_0}$$

where A is the peak area,  $N_o$  is the initial number of moles of the specimen and K is a constant, a callibration coefficient, which is characteristic of the instrument used. The value of K varies with change in temperature; its value is determined by using suitable methods.

A number of standard materials used for calibration purpose are available (Wendlandt). The calibration curves are used in calculations and are obtained by plotting the value of  $\Delta H N_0/A$  against  $T_{\rm max}$ .

# 12.4.2 Specific Heat

The procedure for determination of specific heat by DTA was developed by David. The apparatus used consisted of two identical holders on which small cups are suspended for the sample and reference material. Thermocouples are permanently fixed to the cups. Samples in the range 0.1 to 10 mg were used and DTA curves recorded in vacuum or in helium atmosphere. Following steps are followed:

- DTA curve of the sample (e.g., naphthalene) is recorded over the temperature interval so that the sample does not undergo any thermal effect except the normal change in specific heat. The area 'a' between the horizontal at  $\Delta T = 0$  and the curve recorded with the sample is measured.
- As a next step, the DTA curve is recorded without the sample and the area 'b' measured between the horizontal at  $\Delta T = 0$  and the curve recorded without the sample.
- Finally to specific heat  $(\overline{C}_p)$  it calculated using the formula

$$\overline{C}_p = \frac{KK(b-a)}{dm_s \ dT}$$

where k and K' are constants determined from calibration curves of standard substances. Their values depend on the heat transfer between the sample and reference material and the walls of the cup.

d is the density of the sample

 $m_s$  is the mass of the sample

dT is the temperature interval over which  $\bar{C}_n$  is measured.

#### 12.4.3 Identification of Substances

We know that DTA curves of two substances are not identical. In fact these serve as finger prints (as in IR spectroscopy) for various substances. This technique is particularly used for identification of clays.

#### 12.4.4 Identification of the Products of a Reaction

The product (or products) obtained by reaction of a substance with another substance are identified by their DTA curves.

## 12.4.5 Purity of the Compound

DTA is helpful to determine the melting point of a compound, which in turn is helpful to determine its purity.

#### 12.4.6 QUANTITATIVE ANALYSIS

The area of a DTA peak is proportional to the total heat of a reaction. This also implies that the total heat of a reaction is also proportional to the weight of the sample. In view of this, using standard curves of peak area vs. weight, quantitative analysis is possible. DTA is also helpful to defect the decomposition or volatilization of a sample; this is because DTA is based on changes of heat flow in the sample. Besides this, DTA is also helpful to detect changes such as crystallisation, changes in phase, and homogeneous reactions in solid state etc. that do not involve weight changes. In such changes there is heat flow between the sample and its surroundings due to endothermic or exothermic transitions.

In fact, DTA technique is widely used for the quality control of a number of substances like polymers, cement, glass etc.

#### 12.5 MISCELLANEOUS APPLICATIONS

- DTA is useful to study the thermal stability of a number of inorganic compounds and complexes.
- Determination of purity and identification of organic compounds is possible by DTA.

# **EXERCISES**

- 1. Explain differential thermal analysis. How it is carried out?
- **2.** What factors are responsible for affecting DTA curves?
- **3.** Discuss the application of DTA.

Closely related to DTA is differential scanning calorimetry (DSC). For details *see* chapter Chapter 14, Section 14.3.





# Chapter 13

# **Thermometric Titrations**

#### 13.1 INTRODUCTION

Thermometric titrations are based on measuring the heat generated in a chemical reaction which usually takes place at room temperature under adiabatic conditions. Normally all chemical reactions are accompanied by a heat change. The course of the reaction is followed by observing the heat liberated. The titration is carried out manually in a Dewar flask. Thus, addition of hydrochloric acid from a thermostated burette to solution of sodium hydroxide, which is kept in a thermally insulated vessel following reaction takes place.

$$HCl(aq) + NaOH(aq) \rightarrow NaCl(aq) + H_2O$$

The liberated heat (called the heat of neutralization or heat of reaction,  $-\Delta H$ ) is measured by a very sensitive thermometer calibrated in tenths and hundredths of degrees. Alternatively, the temperature can also be measured by a thermocouple. A plot of the solution temperature against the volume of titrant is shown in Fig. 13.1. A sharp break in the curve occurs at the end point.

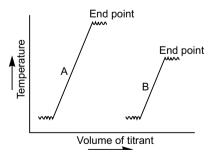


Fig. 13.1 Thermometric titration curves

(A-Titration HCl against NaOH and B-Titration of HCl against Boric acid)

Thermometric titrations are **not** based solely on the equilibrium constant of the reaction. Also, the dielectric constant of the solvent does not influence the thermometric titrations. These can also be performed in non-aqueous solvents.

A essential condition for a thermometric titration is that there should be a change in free energy at the end-point region and the change in free energy ( $\Delta G$ ) is related to enthalphy change ( $\Delta H$ ) by the expression.

$$\Delta H = \Delta G + T \Delta S$$

The enthalphy change is

$$\Delta H = \frac{A.\Delta T}{N}$$

Wher  $\Delta H$  is in Kcal per mol.  $\Delta T$  (°C) is the chance in temperature corresponding to the formation of N moles of the product. A is the apparatus constant and is equal to the effective heat capacity of the titration cell plus reactant solution (K Cal/°C).

#### 13.2 THERMOMETRIC TITRATION APPARATUS

The essential parts of a thermometric titration apparatus are:

- (i) A motor driven burette: It is contained in a thermostated water Jacket.
   This enables the temperature of the titrant to ramain constant within ± 0.05 °C.
- (ii) **Dewar flask** or a thermally insulated beaker. This prevents any loss of heat to the surroundings. The containers mouth is closed with a stopper which has holes for a motor driven stirrer, a themistor and for the burette nozzle.
- (iii) Thermistor bridge assembly: It measures the temperature changes in the solution. Alternatively, a Backmann thermometer can also be used.
- (iv) **Recorder:** It plots the temperature in the flask against the volume of titrant.

A schematic diagram of a thermometric titration apparatus is given in Fig. 13.2.

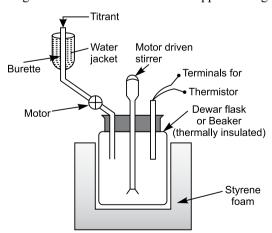


Fig. 13.2 Schematic diagram of a thermometric titration apparatus

#### 13.3 TITRIMETRIC PROCEDURE

Following procedure is followed:

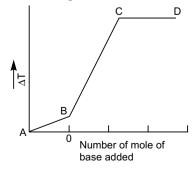
- (i) The solution (known volume) to be titrated is taken in the titrating vessel (insulated beaker or Dewar flask).
- (ii) The thermister is connected to one arm of the Wheatstone bridge.
- (iii) Titrant is filled in the motor driven burette. The concentration of the titrant is normally kept 10-100 times larger than that of the titrating solution. This avoids any volume correction and also to maintain the temperature variations between the solution and the titrant. In fact, higher concentration of titrant is helpful to use only about 3 mL of the titrant for each titration.
- (iv) The titratnt is added automatically from the motor driven burette at a constant rate (appoximately 600 μL per minute) and the motor stirrer helps a thorough mixing.
- (v) The heat evolved (or absorbed) in the reaction is measured with a thermometer, calibrated in tenth and hundredth of degree. Use of thermistor is a better method. Thus, a continuous record of temperature is obtained when the titratant is added continuously from the burette in the titration vessel. The heat liberated or absorbed in the reaction unbalances the bridge potential, which in turn is plotted by the recorder. A unbalanced potential of 15.7 mV corresponds to a change of 1°C.
- (vi) The end point is indicated by a sharp break in the curve.

#### 13.4 APPLICATIONS

Thermometric titrations can be performed successfully in cases where the end point in difficult to infer due colour of the solutions. These titrations have been carried out in neutralisation titrations, precipitation titrations, complexation titrations and redox titrations.

#### 13.4.1 Neutralisation Titrations

Thermometric titrations have been used for the titration of a strong acid with strong base or strong acid against weak base. A thermogram of a strong acid (HCl) and a strong base (NaOH) is shown in Fig. 13.3.



**Fig. 13.3** Thermogram of a neutralisation titration (strong acid/strong base)

100

In Fig. 13.3, the portion AB represents the situation before titrant is added. In fact, the increase in  $\Delta T$  along AB is attributed to the fact that there is exchange of heat with the surrounding from the titration vessel and its contents. At B, the base (titrant) is added, which neutralises the acid in the titration vessel evolving heat of neutration. The titration is complete at C and at this point the end point is reached.

CD represents heat changes due to heat of dilution and the differences in temperature (if any) between the titrant and the solution of the titrating vessel.

Some acid-base titrations, such as boric acid –NaOH, which depend on pH, cannot be titrated with accuracy. Such systems can be titrated using thermometric titrations.

$$HBO_3 + OH^- \Longrightarrow H_2O + H_2BO_3^-$$

A thermometric titration curve of titration of HCl against boric acid is shown in Fig.13.1.

### 13.4.2 Precipitation Titrations

Precipitation titrations, in which heat is evolved or absorbed can be performed using thermometric titration technique. A typical example is the titration of AgNO<sub>3</sub> with HCl.

$$AgNO_3 + HCI \longrightarrow AgCI \downarrow + HNO_3 + \Delta H$$

Following precipitation reactions have also been studied.

$$Ca^{2^{+}} + (NH_{4})_{2} C_{2} O_{4} \longrightarrow CaC_{2} O_{4} \downarrow + 2NH_{4}^{*}$$
 $Hg^{2^{+}} + 2KI \longrightarrow HgI_{2} \downarrow + 2K_{4}^{*}$ 
 $Ni^{2^{+}} + 2KCN \longrightarrow NC(CN_{2}) \downarrow + 2K_{4}^{*}$ 

# 13.4.3 Complexation Titrations

An important application of thermometric titrations is the complexation titration. Using the technique of thermometric titration, it is possible to titrate  $Pb^{2+}$ ,  $Cd^{2+}$ ,  $Cu^{2+}$ ,  $Ni^{2+}$ ,  $Ca^{2+}$ ,  $Zn^{2+}$ ,  $Co^{2+}$  and  $Mg^{2+}$  individually with tetrasodium salt of EDTA, which forms a complex  $M(EDTA)^{2-}$  where M is the metallic ion as stated above.

A very interesting application of thermometric titration is the estimation of  $Cu^{2+}$  and  $Mg^{2+}$  together in a mixture using EDTA. [such a titration is not possible by the usual volumetric procedure using Eriochrome Black T; this is because the stability constants of the chelates do not differ much]. Since the  $\Delta H$  values of the two chelates (+5.5 Kcal/mole for  $Mg^{2+}$ —EDTA and —5.7 Kcal/mole for  $Ca^{2+}$  — EDTA) differ greatly from each ofter, it is possible to determine these ions ( $Cu^{2+}$  and  $Mg^{2+}$ ) in presence of each other by thermometric titrations. In the thermometric titration curve (Fig 13.4), the points for  $Ca^{2+}$  and  $Mg^{2+}$  are shown at  $C_1$  and  $C_2$  respectively.

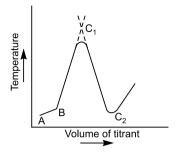


Fig. 13.4 Thermometric titration of a mixture of Ca<sup>2+</sup> and Mg<sup>2+</sup> (0.25 m mole both) with EDTA

#### 13.4.4 Redox Titrations

Only few redox titrations have been studied thermometrically. The examples include oxiadation of  $Fe^{2+}$  ions, potassium ferrocyanide ion, titanium (III) ion and peroxide by ceric ions ( $Ce^{4+}$ ). The oxidation of oxalic acid and ferrocyanide ions by permanganate ion has also been carried out.

$$MnO_4$$
 + OH + 5e  $\longrightarrow$   $Mn^{2+}$  +  $4H_2O$ 
 $Fe^{2+}$   $\longrightarrow$   $Fe^{3+}$  +  $Ie^{-}$ 
 $COO^{2-}$ 
 $COO^{2-$ 

- 1. What are thermometric titration?
- 2. Describe a thermometric titration apparatus giving essential parts.
- 3. Describe thermometric titration procedure.
- **4.** Give applications of thermometric titrations.
- **5.** How will you estimate Ca<sup>2+</sup> and Mg<sup>2+</sup> in a mixture using thermometric titration? Why such an estimation is not possible by the usual volumetric procedure?





Chapter 14

# **Miscellaneous Thermal Methods**

A number of miscellaneous thermal methods are available for analysis. These include:

- Derivative thermogravimetric analysis (DTA)
- Thermobarography.
- Differential scanning calorimetry (DSC)
- Themomechanical Analysis
- Electrical Thermal Analyser

# 14.1 DERIVATIVE THERMOGRAVIMETRIC ANALYSIS (DTA)

The derivative thermogravimetric analysis (as in thermogravimetry) involves the measurement of the rate of change of weight of the substance as it is heated at a slow uniform rate. In derivative thermogravimetric analysis, the derivative of mass change with respect to time, i.e.,  $d_m/d_t$  is recorded as a function of time or temperature

$$\frac{d_m}{dt} = f(T \text{ or } t)$$

#### 14.2 THERMOBAROGRAPHY

This technique consists in heating a sample at a programmed rate and the changes in pressure are measured using a suitable detector.

# 14.3 DIFFERENTIAL SCANNING CALORIMETRY (DSC)

Differential scanning calorimetry (DSC) is closely related to Differential thermal analysis (DTA). It is found that from conventional DTA, it is difficult to obtain quantitative data. This is attributed to uncontrollable factors or and instrumental

parameter like specific heat and thermal conductivity of the sample before and after the transition and some other factors. Much better results are obtained by differential scanning calorimetry (DSC), which works on a different principle. In this procedure, two sample pans (one each for the substance under investigation on the other for reference) are provided with their own heating coils. The current in the heating coil is regulated by a control unit.

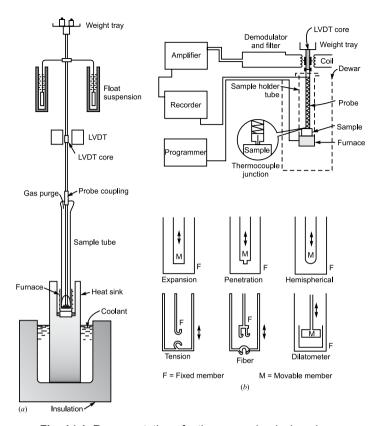
In DSC, the energy necessary to maintain a zero temperature difference between a substance and a reference material is recorded as a function of temperature or time when both the substance and reference are heated or cooled at a predecided rate. The DSC curve is recorded with transition temperature on abscissor. The total energy transfer to or from the sample is obtained from the area of the peaks.

In a DSC instrument, the sample and an inert reference are heated separately. The power supply to the sample heater is variable so that temperature difference can be maintained at zero when an reaction (endothermic or exothermic) take place. The difference in power supply to the two heaters is monitored as a signal ( $\Delta E$ ).

DSC can be used for all the applications of conventional DTA. In DSC, the advantage is in using small size of sample.

### 14.4 THERMOMECHANICAL ANALYSIS (TMA)

It is based on the measurement of mechanical properties (like expansion, contraction, extension or penetration of materials) as a function of temperature. Thus, this technique is useful to assess the mechanical properties of materials. A typical apparatus is shown in Fig. 14.1. It consists of a probe, which is mechanically connected to the core of a linear variable differential transformer. Any change in the sample is recorded by the transformer core, which in turn results in an output that is proportional to the change in the probe. The sign of the change indicates the direction of the movement.



**Fig. 14.1** Representation of a thermomechanical analyser (a) and (b) with configurations of the probe

TMA is mostly used in the study of polymers and for the assessment of their mechanical utility.

# 14.5 ELECTRIC THERMAL ANALYSIS (ETA)

In is useful to measure the electrical conductivity of a material as a function of temperature. In this procedure, two electrodes are immersed in a sample and selected voltage applied. The resulting current is converted into a linear signal. The recorder plots a curve. This method is mostly used for the thermal analysis of electrical insulating materials.

# EXERCISES +

- 1. What is the principal involved in derivative thermogravimetric analysis?
- **2.** What is thermobarography?
- **3.** For what purpose differential scanning calorimetry used? How it is better than conventional DTA?
- **4.** Explain thermomechanical analysis. What are its main applications?



# Part–IV

### **Electrochemical Methods**

- Coulometric method of Analysis
- Polarography
- Amperometric Titrations
- Potentiometric Titrations
- Conductometric Titrations
- Spectrophotometric Titrations
- High Frequency Titrations
- pH Measurements
- Calorimetry



# Chapter 15

# **Coulometric Method of Analysis**

#### 15.1 INTRODUCTION

Coulometric method of analysis is an electrochemical method. It is well-known that in electrolysis, a chemical change is brought about when an electric current is passed through a solution (electrolyte). Let us consider if two metal plates are dipped in an electrolyte. On connecting the two plates to the two terminals of a battery, a current starts flowing through the solution because of the movement of ions in the solution. The negatively and positively charged plates are called cathode and anode respectively and the ions moving towards the cathode and anode are called cations and anions respectively. As soon as the ions reach the two electrodes (cathode and anode), a chemical reaction takes place at each electrode; oxidation at the anode and reduction at the cathode. The reaction that occurs by passing electric current through a solution of CuSO<sub>4</sub>, into which two copper electrodes are dipped is described as:

At anode :  $Cu \rightarrow Cu^{2+} + 2e^{-}$  (oxidation) At cathode :  $Cu^{2+} + 2e^{-} \rightarrow Cu$  (reduction)

It was Michael Faraday (a Britist scientist) who first discovered that the reaction taking place (oxidation and reduction) depends on the quantity of electricity passed.

According to Faraday's law of electrolysis, the chemical change during electrolysis is proportional to the amount of electric current passed through an electrolyte. Mathematically, the law can be stated as

$$W = \frac{M \times q}{n \times F}$$

where W is the mass of an element deposited by passage of q coulomb of current, n is the number of moles of electrons required for the reaction and F is a universal constant, the Faraday having a value of roughly 96500.

Using Faradays law, it is possible to determine quantitatively any substance which can be made to undergo an electrochemical reaction. This method is known as **coulometric analysis**. An electrolytic cell, which is designed to measure the quantity of electric current that passes through the solution is called a **coulometer**.

In coulometry, constant current is passed till the reaction is complete. The magnitude of the current and the time for which the current has been passed help us to calculate the amount of electricity required to attain end point. This is referred to as **coulometric titration**.

Let us consider the oxidation of Fe<sup>2+</sup> to Fe<sup>3+</sup> in coulometric titration, for each Faraday of electricity the chemical change corresponds to one equivalent of the substance of interest (Fe<sup>3+</sup>). Initially, all current is involved in the oxidation of iron (II) at a platinum electrode reaction as shown below.

$$Fe^{2+} \longrightarrow Fe^{3+} + e$$
 ... (i)

As the concentration of  $Fe^{2+}$  decreases, a competing reaction involving the decomposition of water starts

$$2H_2O \longrightarrow O_2 + 4H^+ + 4e$$
 ...(ii)

Thus, the current, which is required for the completion of the oxidation of Fe<sup>2+</sup>(equation (1)) is more than the theoretical amount. Thus, an error is caused. However, this error can be overcome by adding excess at cerium (III) before the starting of electrolysis. The oxidation potential of Ce<sup>3+</sup> being lower than that of water, the following reaction occurs predominantely

$$Ce^{3+} \longrightarrow Ce^{4+} + e$$
 ...(iii)

The Ce<sup>4+</sup> generated (eqn. *iii*) oxides equivalent amount of Fe<sup>2+</sup>

$$Ce^{4+} + Fe^{2+} \longrightarrow Ce^{3+} + Fe^{3+}$$
 ...(*iv*)

In this way, the electrohemical reaction of  $Fe^{2+}$  to  $Fe^{3+}$  proceeds quantitatively.

#### 15.2 COULOMETER

As already stated, a coulometer is a device, which measures the quantity of electricity by determining the amount of change which is affected by the current. Different types of coulometers are available. The most common is a silver coulometer. It consists of a silver anode, which is suspended in a platinum crucible, which acts as cathode. The two electrodes (silver anode and platinum cathode) are separated with a porous cup, in which any metallic particles of silver, which may drop off are collected. The platinum crucible is filled with the electrolyte (pure solution of silver nitrate) (Fig 15.1).

The platinum crucible is washed carefully, dried and weighed before and after the electrolysis. The increase in weight gives the amount of silver deposited in the coulometer. It is known that 96500 coulombs (or one Faraday) of electricity deposits

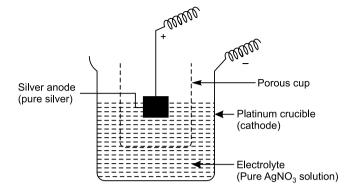


Fig. 15.1 Silver coulometer

107.88 gm. of silver. From this, the quantity of electricity used to deposit the determined amount of silver in the coulometer can be calculated.

#### 15.3 COULOMETRIC ANALYSIS

The coulometric analysis is of two types. These are constant current coulometric analysis and controlled potential coulometric analysis

### 15.3.1 Constant Current Coulometric Analysis

In this analysis, constant current is used and the time for which the current is passed is determined accurately. The amount of electricity is obtained as the product of current and time.

Amount of electricity passed = current (amp)  $\times$  time (sec)

This technique is accurate and simple. This type of analysis is further divided into two types, *viz.*, primary constant current coulometry and secondary constant current coulometry.

### 15.3.1.1 Primary constant curent coulometry

In this type of analysis, the element to be estimated undergoes reaction at the electrode. As an example, ferrous ion is oxidised into ferric ion at the anode. There is quantitative conversion.

### 15.3.1.2 Secondary constant current coulometry

In this type of analysis, the titrant is produced on the electrode, which in turn reacts with the ion to be estimated. A typical example is the oxidation of  $Fe^{2+}$  into  $Fe^{3+}$  by  $Ce^{4+}$ .

Initially Ce<sup>4+</sup> ion is produced by the oxidation of Ce<sup>3+</sup> ion at the anode

$$Ce^{3+} \longrightarrow Ce^{4+} + e$$
 (at anode)

The  $Ce^{4+}$  produced reacts with  $Fe^{2+}$  to give  $Fe^{3+}$ .

$$Ce^{4+} + Fe^{2+} \longrightarrow Ce^{3+} + Fe^{3+}$$

The Ce<sup>3+</sup> is used again and the reaction goes to completion.

### 15.3.2 Controlled Potential Coulometric Analysis

In this type of analysis constant potential is maintained at anode or cathode. This permits only a single reaction to occur at the electrode. The completion of the reaction is indicated until zero current is reached. The amount of species of interest is found by the total quantity of electricity produced using the Faraday's law.

#### 15.4 COULOMETRIC TITRATIONS

# 15.4.1 Principles of Coulometric Titrations

In coulometric titration, the primary standard and the titrant is the electron, which is generated by an electrochemical reaction. The substance under investigation may be oxidised or reduced in case of primary coulometric analysis (*see* section 15.3.1.1). Alternatively, the substance under investigation may react quantitatively with a product of electrolysis as in the case of secondary coulometric analysis (*see* section 15.3.1.2).

During titration, the current as already stated is maintained constant at a fixed value. The amount of electricity passed is obtained from the product

As the concentration of the substance under investigation decreases, polarisation sets in. In order to maintain constant current, the electrode potential must rise. This is achieved by the addition of cerium (1V) (as already explained in section) (15.3.1.2).

# 15.4.2 Advantages of Coulometric Titrations

- (i) These titrations are more accurate than the usual volumetric procedures.
- (ii) There is no need to use burette and preparation of standard solution. In these methods the electron is the primary standard and titrant.
- (iii) Even minute quantities of the titrant can be accurately generated, which is not possible in volumatric procedures.
- (iv) The method can be easily adapted for automatic operation.

#### 15.4.3 Errors in Coulometric Titrations

- (i) Error in the measurement of current.
- (ii) Variation in current during titrations.
- (iii) Error in the measurement of time.
- (iv) As in the case of volumetric titration, error may arise due to difference in equivalence point and end point.

# 15.4 NATURE OF ELECTRODES USED IN COULOMETRIC TITRATIONS

The nature of the electrode used depends as the naturature of titration. Following Table (15.1) gives the nature of titration and the electrodes used.

Nature of titration	Electrode use	
Neutralisation titrations	Platinum anode	
Redox Titrations	Platinum or gold anode	
Complexometric Titrations	Mercury electrode	
Precipitation Titrations	Silver or mercury electrode	

**Table 15.1** Types of electrodes used in different titrations

#### 15.5 APPLICATIONS OF COULOMETRIC TITRATIONS

All titrations which were earlied performed volumetrically can now be conducted coulometrically.

#### 1. Neutralisation titrations

Neutralisation titrations or acid-base titrations are carried out using electro generated OH<sup>-</sup> ions. A convenient method is the generation of OH<sup>-</sup> ions at a platinum cathode within the soultion. The end point is determined potentiometrically by using an appropriate indicator.

For strong base and weak bases, H<sup>+</sup> ions are generated at a platinum anode

$$H_2O = 2H^+ + \frac{1}{2}O_2 + 2e^-$$

The generation of H<sup>+</sup> ions can be carried out internally or externally.

For external generation of the reagent (H<sup>+</sup> or OH<sup>-</sup>), a procedure deviced by De Ford, Pitts and Johns is used. The flow sheet is shown in Fig. 15.2.

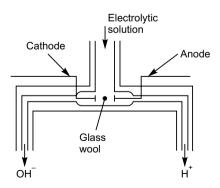


Fig 15.2 Flow shut for generation of reagent

At anode 
$$H_2O \longrightarrow \frac{1}{2}O_2 + 2H^+ + 2e$$

At cathode 
$$2H_2 + 2e \longrightarrow H_2 + 2OH^-$$

In coulometric neutralisation titrations, it is necessary to remove CO<sub>2</sub> from the solution to be analysed before analysis.

#### 2. Precipitation Titrations

Individual halide ions can be determined accurately by precipitation with anodically generated silver ions. The end point is determined by potentiometric or indicator method. It is also possible to analyse mixture of haldes. Electrogenerated mercury (I) is used to titrate halides (Cl<sup>-</sup>, Br<sup>-</sup>, I<sup>-</sup>) individually and in mixture.

#### 3. Complexometric Titrations

Coulometric titration of a number of cations can be performed by means of EDTA ion (Hv<sup>3-</sup>) generated at mercury anode. The EDTA ion is generated by introducing excess of Hg (II) complex of EDTA into ammonical solution of the sample

$$HgNH_3y^{2-} + NMH_4^+ + 2e^- \longrightarrow Hg + 2NH_3 + Hy^{3-}$$

The formed Hy<sup>3-</sup> in turn reacts with the cation under investigation. Thus, in case of Ca<sup>2+</sup>, the reaction is

$$Ca^{2+} + Hy^{3-} \longrightarrow CaHy^{2-} + NH_4^+$$

#### 4. Redox Titrations

It is well-known that titration which incorporate both oxidation and reduction are called redox titrations. These titrations are conveniently conducted by coulometric methods. For these reactions, a number of reagents (including halides, Ce<sup>4+</sup>, U<sup>4+</sup> etc.) have been used. Some of the important coulometric titrations involving redox reactions are given in Table 15.2.

Reagents	Generator electrode reaction	Substances determined
Br <sub>2</sub>	2Br <del>←</del> Br <sub>2</sub> + 2e <sup>-</sup>	As (III), Sb(III), U(IV) I⁻, SCN⁻, Phenol, aniline
Cl <sub>2</sub>	2Cl⁻ <del>←</del> Cl <sub>2</sub> + 2e⁻	As (III), I⁻
l <sub>2</sub>	2l <del>←</del> l <sub>2</sub> + 2e <sup>-</sup>	As (III), Sb(III), S <sub>2</sub> O <sub>3</sub> <sup>2-</sup>
Ce <sup>4+</sup>	Ce <sup>3+</sup> <del>←</del> Ce <sup>4+</sup> + e <sup>-</sup>	Fe(II), Ti(III)
Fe <sup>2+</sup>	Fe <sup>3+</sup> + e <del>←</del> Fe <sup>2+</sup>	Cr <sup>6+</sup> , Mn <sup>7+</sup> , Ce <sup>4+</sup>
Ag⁺	$Ag^+ \rightleftharpoons Ag^{2+} + e^-$	Ce <sup>5+</sup> , V <sup>4+</sup> , As <sup>3+</sup>

Table 15.2 Some Coulometric Redox Titration

### 5. Organic Compounds

Organic compounds can be determined by controlled-potential coulometric procedure. It has been shown that trichloroacetic acid and picric acid are reduced quantitatively at mercury cathode whose potential is suitably controlled.

OH O<sub>2</sub>N 
$$\rightarrow$$
 NO<sub>2</sub> + 18 H $^{+}$  + 18e  $\rightarrow$  H<sub>2</sub>N  $\rightarrow$  NH<sub>2</sub>  $\rightarrow$  NH

# EXERCISES #

- 1. Explain the term coulometric titration.
- 2. Discuss the principle involved in coulometric titration.
- **3.** Which of the two, *viz.*, primary constant current coulometry or secondary constant current coulometry gives better results. Explain.
- **4.** Give the reactions involved in the conversion at  $Fe^{2+}$  into  $Fe^{3+}$  in presence of  $Ce^{4+}$ .
- 5. Discuss the advantages and possible errors in coulometric titrations.
- **6.** Give applications of coulometric titrations.
- 7. Write notes on:
  - (a) Silver coulometer (b) Controlled potential coulometric analysis.
- **8.** How coulometric titration can be performed using external generation of reagents?





Chapter 16

# **Polarography**

#### 16.1 INTRODUCTION

It is an extremely sensitive electochemical method of analysis. The method involves the use of a dropping mercury electrode and is particularly useful for determining even small concentrations of metals in solution. The method was developed by Czechoslovakian Chemist, Jarosal Heyrovsky, who was awarded Nobel Prize in chemistry in 1959 for the discovery and development of polarography.

Polarography finds used in the qualitative and quantitative analysis of substances, which can undergo cathodic reduction or anodic oxidation. Subsequently, this technique was used for the determination of composition and stability of inorganic complexes. The method also finds use in the study of coordination compounds.

In polarographic analysis, current is passed between two electrodes, both of mercury. One of the electrodes is large in area in the form of a pool of mercury at the bottom of the cell. The other electrode is a drop of mercury, which comes out of a fine capilliary tube and is known as dropping mercury electrode. The electrolyte is a dilute solution of an electroactive material to be analysed; a different electrolyte base or supporting electrolyte is also used along with the electrolyte. The supporting electrolyte carries bulk of the current and also increases the conductivity of the solution. A steady increasing voltage is applied to the cell and a current-voltage curve drawn. Such a curve is reproducible. From this curve, information about the concentration and nature of the material can be obtained. The current flowing through the cell at any applied voltage is found with the help of an instrument called polarograph and the curves obtained are known as polarograms.

#### **16.2 THE INSTRUMENT**

A mannual polarograph is shown in Fig. 16.1. It contains a dropping mercury electrode (in the form of mercury reservoir from which mercury drops down in the

form of small drops through a capilliary tube), which acts as a cathode. Mercury in the form of mercury pool is at the bottom of the cell; it acts as a reference electrode. Since its area is large, it is not polarised.

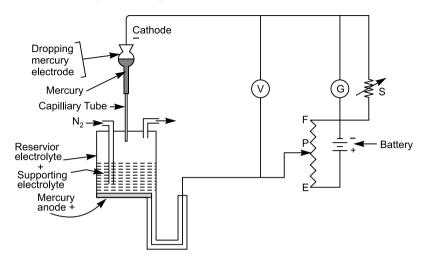


Fig. 16.1 Manual polagraph P. - Potentiometer G-Galvanometer S-shunt

The cathode and anode are connected at the appropriate end of a battery. The applied voltage may be adjusted by sliding contact along the potentiometer wire (EF). The potentiometer (P) can read E.M.F. up to three volts. The galvanometer (G) measures the strength of the current. The shunt(s) adjusts the sensitivity of the galvanometer. The dissolved oxygen in the electrolyte is removed by bubbling in nirogen. Through a number of micro electrodes can be used, but the dropping mercury electrode is the best, which has the advantages given below:

- For any capillary size, the surface area is reproducible.
- The reduction potential of the metal formed is lowered due to formation of amalgams (Hg-metal)
- The surface area can be calculated from the weight of the drop.

However, dropping mercury electrode has certain disadvantages. These include

- The area of the micro electrode (anode) changes as the size of the drop changes
- Mercury is easily oxidised thereby limiting the feasible range of the electrode.
- The capillary may be plugged, thus disrupting the whole process.

Let us consider that the electrolyte is a solution of cadmium chloride. When an external E.M.F. is applied, the positively charged cadmium ions (Cd<sup>2+</sup>) are attracted at the cathode (droping mercury cathode) by an electrical force and by a diffusive

force which results from the concentration gradient formed at the surface of the electrode. The total current flowing through the cell is the sum of electrical and diffusive forces. A graph as shown in Fig. 16.2 is obtained as the applied voltage is increased and the current recorded.

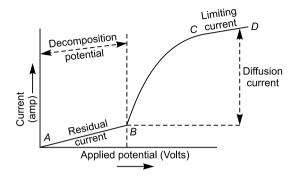


Fig. 16.2 Plot of applied potential vs current

As seen, only small current flows from A to B. This current is called the **residual current** and is carried by the supporting electrolyte. At point B, the potential of the electrode equals to the decomposition potential of  $Cd^{2+}$  ion. Subsequently, the current increases along the curve BC. At point C the current does not increase linearly with applied voltage and reaches a limiting value at point D. After these, there is no increase in current. The current corresponding to the curve CD is known as **limiting current**.

The difference between the limiting current and residual current is called **diffusion current**. (denoted by  $i_d$ ) (Fig. 16.2).

The polograph so far described (Fig. 16.1) is a manual polarograph. However, now recording polographs are available, which automatically record current voltage curves.

#### 16.3 FACTORS AFFECTING CURRENT-VOLTAGE CURVES

The current-voltage curve, also known as polarograms are the outcome of the polarography. A number of factors affect the current-voltage curves. These include

- Residual current
- Migration current
- Diffusion current.
- (i) Residual Current: During electrolysis of an electroactive species, initially small current flows, called residual current (see line A-B in Fig. 16.2).

When negatively charged mercury drop (from droping mercury cathode), the anion  $(K^+)$  in the supporting electrolyte (e.g., KCl) are attracted to the drop; this results in the formation of electrical double layer. As more mercury drops fall through

the capillary, a continuous flow of electric current takes place, which increases as the potential of the drop increases. In case, the electrolyte contains traces of impurities, a small faradic current is also superimposed on the current generated.

- (ii) Migration Current: The electroactive material reaches the electrode surface either by migration of charged particles in the electrical field due to potential difference between the surface of the electrode and the solution, or by diffusion of particles. The current required by both the above two processes is called migration current. According to Jaroslav Heyrovsky, the migration current can be eliminated if a supporting electrolyte be added in large concentration to the solution so that the ions of the suppoting electrolyte carry practically all the current. As an illustration, suppose a solution it 0.1 M KCl (supporting electrolyte) and 0.005 M an eletroactive material (e.g., Cd<sup>2+</sup> ions), all the ions present help for the migration of current through the cell; the fraction of current carried by each ion (K<sup>+</sup> or Cd<sup>2+</sup>) depends on their concentration and transference number. Thus, about 90% of the current will be carried by K<sup>+</sup> ions and only 10% by Cd<sup>2+</sup> ions. If, however, the concentration of K<sup>+</sup> ions is increased to more than 99 percent of the total ions present), almost all the current will be carried towards the cathode by K<sup>+</sup> ions.
- (iii) Diffusion Current: As already stated (Fig. 16.2), the difference between the limiting current and residual current is known as diffusion current. The diffusion current is directly proportional to the substance being reduced or oxidised at the dropping mercury electrode.

The diffusion current is given by the Ilkovic equation

 $i_d = 607 \ nCD^{1/2} \ m^{2/3} \ t^{1/6}$ 

where

 $i_d$  = diffusion current in microamperes,

n = number of electrons transferred per ion,

c =concentration in millimoles per litre,

 $D = \text{diffusion coefficient of the reducible ion in cm}^2 \text{sec}^{-1}$ ,

m =mass of mercury flowing per seond is milligrams,

t =drop time in seconds.

On the basis of Ilkovic equation, it is found that the diffusion current is directly proportional to the concentration of the electroactive material. In fact, this is the basis of quantative polarographic analysis.

Ilkovic equation does not take into account of the curvature of the electrode. This was taken care by the Lingane and Loveridge equation (given below):

$$i_d = 607 \ nCD^{1/2} \ m^{2/3} \ t^{1/6} \ (1 + 39D^{1/2} \ m^{-1/3} \ t^{1/6})$$

The figures in brackets correspond to the difference between linear and spherical diffusion. This correction has values 2-5% of the total current. Only for very accurate work, the above modified equation is used.

#### **16.4 HALF WAVE POTENTIALS**

In polography, the half wave potential is a characteristic property of the electroactive material. It is represent by  $E_{1/2}$  and is the potential found on the rising portion of current voltage current and is one half of the distance between the limiting and residual current (Fig. 16.3). The half wave potential is the basis for the qualitative and quantative polarographic analysis.

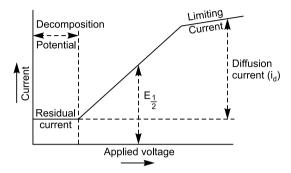


Fig. 16.3 Plot of current versus applied voltage

We know that in a chemical reaction, oxidation and reduction take place simultaneously at the anode and cathode respectively. The reduction occurring at the dropping mercury cathode is represented as

Oxidant + 
$$n$$
 electrons  $\Longrightarrow$  Reductant

The equation can also be represented as

$$OX + ne \Longrightarrow red$$
 ...(i)

The polarogram records the reversible potential of the system as it exists at the electrode-solution interface of the drop of mercury. The electrochemical equilibrium is given by the following equation

$$E = E^{0} + \frac{0.0591}{n} \log \frac{[O_{x}]_{e}}{[\text{red}]_{e}}$$
 ...(ii)

where E is the electrode potential of the above reversible reduction [equation (i)] at dropping mercury electrode,  $E^{\circ}$  is the electrode potential of the reaction against the reference electrode,  $[O_x]_e$  and  $[\text{red}]_e$  are the concentrations of the oxidant and reductant at the surface of the electrode.

Let us assume that just before the start of the current-voltage curve, the solution at the electrode interface consist of only oxidant. As and when the E.M.F. becomes large enough to reduce it, the concentration of the oxidant at the electrode surface starts decreasing. To take care of the concentration, some ions from the bulk of the solution moves in by diffusion. The average current, *i* flowing through the cell, at any point of the wave is given by the equation.

$$i = K[(Ox) - (Ox)_o]D_{ox}^{1/2}$$
 ...(iii)

where K includes capillary characteristics,  $(O_x)$  is the concentration of the reducible ions in the bulk of the solution and  $(O_x)_e$  is the concentration of the oxidant at the electrode-solution interface.

As and when the current attains the limiting value, *i.e.*, the concentration of the oxidant at the elecodes solution interface becomes zero. Thus, the equation (*iii*) becomes

$$i_d = K[Ox]D_{ox}^{1/2} \qquad \dots (iv)$$

From equations (iii) and (iv), we get

$$(Ox)_o = \frac{i_d - i}{KD_{ox}^{1/2}} \qquad \dots(v)$$

For metals, which form amalgams with the dropping electrode, the concentration of the metal is directly proportional to the current on the current voltage curve. So

$$i = K[\text{red}]_o D_{\text{red}}^{1/2} \tag{vi}$$

Substituting equations (v) and (vi) in (ii), we get

$$E = E^{0} + \frac{0.0591}{n} \log \frac{id - i}{i} + \frac{0.0591}{n} \log \left[ \frac{D_{\text{red}}}{D_{ox}} \right]^{1/2} ...(vii)$$

We have known earlier that, the half wave potential is a point, where

$$i = i_{d-i}$$
 or  $i = \frac{1}{2}i_d$ 

So the equation (vii) becomes

$$E^{1/2} = E^0 + \frac{0.0591}{n} \log \left[ \frac{D_{\text{red}}}{D_{ox}} \right]^{1/2}$$
 ...(viii)

From the above equation (*viii*), it is clear that the polographic half wave potential  $(E_{1/2})$  is related to standard reduction potential of oxidation-reduction system.

### 16.5 APPLICATIONS OF POLAROGRAPHY

(*i*) Estimation of Cations and Anions: Polarography has been successfully used for the estimation of cations and anions. For estimation of cations, a suitable supporting electrolyte has to be used. Best results are obtained in  $10^{-2}$  to  $10^{-6}$  M concentration range.

Lithium can be estimated in presence of other alkali metals provided the concentration of alkali metals is not very high.

Alkaline earth metals can be determined by using alkaline solutions as supporting electrolytes.

Simultaneous determination of two or more constituents in the same solution can be achieved provided these constituents are present in very different concentrations.

Anions like borate, iodate, vanadate, selenite, nitrite and dichromate can be determined by polarographic analysis provided the central atom has two or more oxidation states. However, in this case strong buffering of the solution is necessary.

Polarographic technique has also been used for the analysis of alloys.

(ii) Estimation of Organic Compounds Containing Reducible Functional Groups: Polographic studies were earlier confined to quinones and some other functional systems like phenylene diamines which resemble quinone in having resonating systems.

Polographic techniques has been used for the estimation of organic compounds containing reducible groups. Some of such group are given in Table 16.1.

Compound	Group	Example
Ketone	C = 0	$C_6H_5COR + e + H^+ \rightarrow C_6H_5C - R$
		   OH
Aldehyde	- CHO	$C_6H_5CHO + 2H^+ + 2e \xrightarrow{\text{basic}} C_6H_5CH_2OH$
		$2C_6H_5$ CHOH + $2H^+$ + $2e \xrightarrow{pH < 2} C_6H_5$ CH OH $C_6H_5$
Alkene	>c = c<	$C_6H_5CH = CH_2 + 2e + 2H^+ \longrightarrow C_6H_5 CH_2 CH_3$
Alkyne	- C ≡ C -	$C_6H_5C \equiv C.CHO + 2e + 2H^+ \longrightarrow C_6H_5 CH = CH CHO$
Nitrile	- C ≡ N	$C_6H_5CH_2COC \equiv N + 4e + 4H^+ \longrightarrow C_6H_5CH_2COCH_2NH_2$
Nitro	- NO <sub>2</sub>	$R - NO_2 + 4e + 3H^+ \longrightarrow R - NH OH + OH^-$
Azo	– N = N –	$C_6H_5 N = N C_6H_5 + 2e + 2H^+ \longrightarrow C_6H_5 NHNH C_6H_5$
Disulphide	-S-S-	$R - S - S - R + 2e + 2H^{+} \longrightarrow 2RSH$
Bromo	– Br	$RCH_2Br + e + H^+ \longrightarrow R - CH_3 + Br^-$

Table 16.1 Reducible functional groups

(iii) Polarography of Metal Complex: The polographic technique has been used for the determination of stability constants of metal complexes if the simple metal ion and complex of that metal ion are in the same oxidation state, give reversible electrode processes and involve the same number of electrons.

# EXERCISES

- 1. What do you understand by polarography? Discuss the principles involved.
- **2.** Explain the following terms:
  - (a) Residual current
  - (b) Migration current
  - (c) Diffusion current.
- 3. What is the function of dropping mercury electrode in polarography?
- **4.** Explain half wave potential. What are its applications?
- **5.** What are the applications of polarography?
- **6.** How do you come to the conclusion that the diffusion current is directly proportional to the concentration of electroactive material?
- 7. What is a supporting electrolyte? What are its functions?





Chapter 17

# **Amperometric Titrations**

#### 17.1 INTRODUCTION

The basis of amperometric titrations is polarography. In amperometric titrations, the voltage, which is applied across the indicator electrode and reference electrode is kept constant. The diffusion current (which we known is equivalent to limiting current minus residual current) passing through the cell is measured and plotted against the volume of the reagent added.

It is known that in polarographic technique (*see* chapter 16), the current is independent of the applied voltage on the dropping mercury electrode. In case supporting electrolyte is taken in excess, the migration current is eliminated (in such a case the ions of the supporting electrolyte carry practically all the current) and the limiting curent is the rate of diffusion of electroactive material from the bulk of the solution to the surface of the electrode. Thus, in this case, the diffusion current is proportional to the concentration of the electro active material in the solution. If some of the electro active material is removed by any method by reaction with a reagent, the diffusion current decreases. This forms the basis of amperometric titrations.

#### 17.2 APPARATUS FOR AMPEROMETRIC TITRATIONS

The apparatus used for amperometric titrations is simple in the sense that any apparatus for polarography can be used with simple modification and simplification. Appropriate care is taken that the temperature remains almost constant during the time of titration (10 - 12 min). A diagrammatic representation of a simplified apparatus is shown in Fig. 17.1.



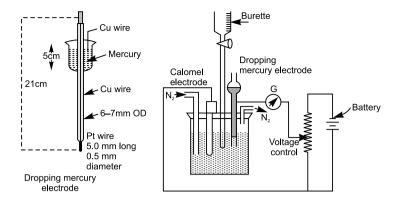


Fig. 17.1 Simplified apparatus for amperometric titrations

The titration may be performed either by the dropping mercury electrode or with rotating platinum electrode. Calomel electrode is used as a reference electrode. The current is measured by a galvometer (*G*). For titration, the voltage applied to the indicator electrode and the reference electrode is kept constant. The sensitive galvometer gives the value of the diffusion current after each addition of the titrant.

Following procedure is followed for amperometric titrations:

- The dropping mercury electrode (cathode) and saturated calomel electrode (anode) is connected to the battery.
- A known volume (about 25 mL) of the test solution is taken in the titration cell.
- Nitrogen is bubbeled through the solution for 10 15 min.
- The burette (micro or semimicro) is filled with the reagent of known concentration.
- The voltage is adjusted at the desired value and the initial diffusion current is noted.
- Known volume (0.02 mL) of the reagent is added from the burette, nitrogen bubbled through the solution for about 2 min in order to eliminate traces of oxygen from the added reagent and complete mixing.
- An inert atmosphere is maintained by slowly bubbling nitrogen.
- The burette reading (reagent added) and the corresponding current reading is noted.
- The process is repeated unit a number of readings have been obtained.
- Due to change in volume of the solution during titration, correction has to be applied using the formula.

$$i_{\text{corr}} = i_{\text{obs}} (V - v) V$$

Where V is the initial volume of the test solution and v is the volume of the titrating reagent added.

 Finally, the corrected current is plotted as ordinate and the volume of the reagent added as abscissae and a straigth line is drawn through the two brances of the curve. The point of intersection is the end point.

Some reference calomel electrodes are given in Table 17.1.

Table 17.1	Electrode potentia	il (in volts) of calome	l electrodes	
as function of temperature				

Temperature (°C)	0.1 M calomel	Saturated calomel	1.0 m Ag/AgCl
10	0.3362	0.2528	0.2314
15	0.3360	0.2508	0.2286
20	0.3358	0.2476	0.2256
25	0.3356	0.2444	0.2223
30	0.3353	0.2417	0.2190

As already stated, in place of a dropping mercury electrode, a rotating platinum micro electrode can be used. It consists of a glass tube (15-20 cm long and 6 mm diameter). A platinum wire (5-10 mm) extends from the wall of the glass tube (Fig. 17.2). The platinum electrode is mounted in the shaft of a suitable motor so that it rotates at a constant speed at 600 rotations per minute.

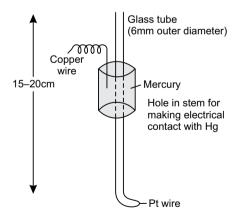


Fig. 17.2 Diagrammatic representation of a rotating platinum electrode

The rotating platinum electrode is simple to construct and increases the workable range on the positive voltage side up to 0.9V. It can be used at positive potentials whereas dropping mercury electrode cannot be used. Another advantage of rotating

platinum electrode is that the value of the diffusion current is increased 20 times the value in polarography.

#### 17.3 END POINT IN AMPEROMETRIC TITRATIONS

In amperometric titrations, either the reagent or the species being titrated should be electroactive. It is, however, not necessary for both to be electroactive. One of the following situations may be there:

(i) In case the metal is electroactive and the reagent is electroinactive (as in the titration of lead<sup>2+</sup> with  $SO_4^{2-}$  or  $C_2$   $O_4^{2-}$ ), the diffusion current decreases to the residual current when it has fully reacted with the reagent. In this case, the shape of the curve will be as shown in Fig. 17.3. The end point is obtained as shown in Fig. 17.3.

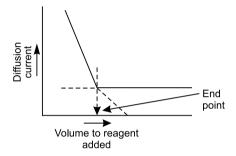


Fig. 17.3 Titration of electroactive material with electroinactive reagent

(ii) In case, the material is electroinactive and the reagent is electroactive (e.g.,  $SO_4^{2-}$  titrated with  $Ba^{2+}$  or  $Pb^{2+}$ ), residual current flows between the electrodes. The addition of electroactive reagent increases the diffusion current and the type of the curve shown in Fig. 17.4 will be obtained. The end point is obtained as shown in Fig. 17.4.

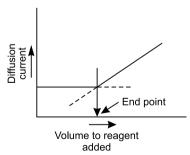


Fig. 17.4 Titration of electro inactive material with electroactive reagent

(*iii*) In case, both the material and the reagent are electroactive, they both contribute to the diffusion current and a V shape curve is obtained (*e.g.*, titration at  $Ni^{2+}$  with dimethylglyoxime). The end point in this case is the intersection of two lines (Fig. 17.5).

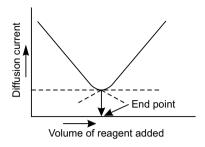


Fig. 17.5 Titration of electroactive material with electroactive reagent

In all the cases [(i), (ii)] and (iii) described above the end point is the point of intersection of two lines which give the current change before and after the equivalence point.

An interesting case [different from (i), (ii) and (iii)] is described below.

(*iv*) Redox titrations can also be carried out by amperometric method. In such a case, both the oxidising and reducing agents given diffusion current. The titration curve of this type is shown in Fig. 17.6.

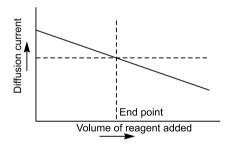


Fig. 17.6 Titration curve for Redox titration

As an illustration amperometric titration of ferric ions (Fe<sup>3+</sup>) against titanous ions (Ti<sup>3+</sup>). In this case the diffusion current linearly decreases with the addition of titrant (Ti<sup>3+</sup>) to ferric ions until it attains the zero value at the end point. At the end point, the ferric ions have completely reacted. After the end point, when more titrant is added, a diffusion current due to oxidation of titanous ion is set up. A change in the slope due to the difference in diffusion coefficients is evident as the lines cross the zero axis. The end point is the point of intersection of two lines.

Other examples of redox titration by amperometric procedure is the titration of iodide with mercury nitrate, and chloride ion with silver ion.

#### 17.4 ADVANTAGES OF AMPEROMETRIC TITRATIONS

(i) The apparatus used is simple and is more sensitive than potentiometric or conductometric titrations.

- (ii) The amperometric titrations are best for as low as 0.001M concentration with good results.
- (iii) The capillary characteristics and electrode characteristics do not interfere in the titrations
- (iv) As the temperature is kept constant through out the titration, there is no need to record the temperature.
- (v) The presence of foreign salts do not interfere in the titration. These are usually added as the supporting electrolytes.
- (vi) Reversible or irreversible reactions can be carried out equally well.
- (vii) Depolarising substances, which cannot be accurately determined by polarography, can also be determined by amperometric titrations. In such cases, a suitable titrant, which gives diffusion currents is selected.
- (viii) Systems, which do not possess measurable equilibrium can also be estimated by amperometry. In this case, redox characteristics of other reagents is used. As an example, magnesium (which does not give diffusion current curve) can be estimated with 8-hydroxyquinoline (oxine), which yields a diffusion current.
  - (ix) Amperometric titration is also applicable in precipitation titrations.

#### 17.5 DISADVANTAGES OF AMPEROMETRIC TITRATIONS

- (i) At times, due to coprecipitation, the results obtained are inaccurate.
- (ii) In the presence of higher concentration of foreign substances the results obtained are unreliable. This is because at higher concentration, the relative current becomes smaller.

#### 17.6 APPLICATIONS OF AMPEROMETRIC TITRATIONS

- (i) Determination of sulphate (using lead nitrate), which cannot be determined potentiometrically since no suitable indicator is available.
- (ii) Determination of Co, Cu and Pd using α-nitroso-β-naphthol,
- (iii) Determination of iodide with mercuric nitrate.
- (iv) Determination of nickel with dimethylglyoxime.
- (v) Determination of lead with dichromate ion.
- (vi) Simultaneous determination of Cl<sup>-</sup>, Br<sup>-</sup> and I<sup>-</sup>.

For simultaneous determination of Cl<sup>-</sup>, Br<sup>-</sup> and I<sup>-</sup>, the procedure developed by H.A. Laitinen and co workers (1946) is used. In this procedure, a rotating mercury electrode and Hg|HgI<sub>2</sub>|KI reference electrode (-0.23 V vs SCE) is used. A known volume of the mixture (containing Cl<sup>-</sup>. Br<sup>-</sup> and I<sup>-</sup>) is placed in the titration cell containing 0.1 - 0.3 N solution of ammonia (pH9). When silver solution is added from a micro burette, only iodide gets precipitated as AgI under these conditions [the Br<sup>-</sup> forms the complex [Ag (NH<sub>3</sub>)<sub>2</sub>] Br]. The current remains zero till all the iodide gets precipitated, which happens at the end point (Fig. 17.7). The titration is continued after the end point. The solution is a cell is acidified with HNO<sub>3</sub> to make it 0.8 N HNO<sub>3</sub>. Under these conditions, the Br<sup>-</sup> is released from the complex, [Ag (NH<sub>3</sub>)<sub>2</sub>] Br formed earlier combines with silver ions from the reagent and AgBr is precipitated. At this stage also, the current drops to zero and remains at zero till all the bromide ions have been consumed, which happens at the end point (Fig. 17.7). Finally, for the determination of Cl<sup>-</sup>, a solution of 0.1% gelatin is added (gelatin suppress the current due to AgCl). The titration is continued as for bromide or idodide ions. The third rise in current indicate the end point for chloride titration (Fig. 17.7).

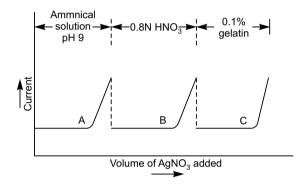


Fig. 17.7 Amperometric titration of a mixture of Cl<sup>-</sup>, Br<sup>-</sup> and l<sup>-</sup>
[A is the end point when all the l<sup>-</sup> is used up
B is the end point when all the Br<sup>-</sup> is used up and
C is the end point when all the Cl<sup>-</sup> is used up]

The amperometric titrations have been commonly used for precipication titration. Some of these are given below (Table 17.2).

Electrode	Reagent	Substance determined
Dropping Mercury electrode	K <sub>2</sub> Cr O <sub>4</sub>	Pb <sup>2+</sup> , Ba <sup>2+</sup>
	Pb (NO <sub>3</sub> ) <sub>2</sub>	F <sup>-</sup> , Cl <sup>-</sup> , SO <sub>4</sub> <sup>2-</sup>
	8-Hydroxy quinoline	Mg <sup>2+</sup> , Zn <sup>2+</sup> , Cu <sup>2+</sup>
	Cupfferon	Fe <sup>2+</sup> , Cu <sup>2+</sup>
	Dimethyl glyoxime	Ni <sup>2+</sup>
Rotating Platinium electrode	AgNO <sub>3</sub>	Cl <sup>-</sup> , Br <sup>-</sup> , l <sup>-</sup>

Table 17.2 Some precipitation titration using amperometric procedure

# 17.7 AMPEROMETRIC TITRATIONS WITH TWO INDICATOR ELECTRODES

This is a modification of the usual amperometric titration and was introduced by C.W. Foulk and A.T. Baldwen, who coined the word 'dead stop end point' for this procedure. In this method two similar platinum electrodes dipping in two different containers are used and are linked *via*. a salt bridge (Fig. 17.8). This procedure is applicable only when oxidation reduction system is involved before and after the end point.

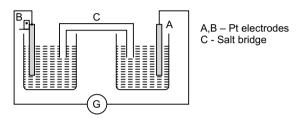


Fig. 17.8 Setup for amperometric titrations with two indicator electrodes

A small and constant voltage is applied to the two electrodes. In this procedure, the amount of oxidised form reduced at the cathode equals to that formed by the oxidation of the reduced form at the anode. At this stage, both the electrodes are depolarised equally until one of the components either oxidised or reduced is completely consumed by the titrant. After the end point has reached only one electrode remains depolarised in case the titrant does not involve a reversible system. The current becomes zero at the end point and shoots up again as shown in Fig. 17.9.

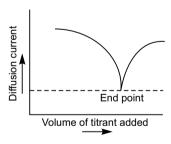


Fig. 17.9 End point in a amperometric titration with two electrodes

Following amperometric titrations have been performed by this technique:

- (i) Titration of iodine by thiosulphate
- (ii) Titration of Fe<sup>2+</sup> by Ce<sup>4+</sup> ion
- (iii) Titration of V<sup>4+</sup> by Fe<sup>2+</sup>
- (iv) Titration of hexacyanoferrate (II) with  $Ce^{4+}$ .

# EXERCISES :

- 1. Explain amperometric titrations. Discuss their advantages and disadvantages.
- 2. Discuss the principle of amperometric titrations.
- **3.** Give the procedure involved in amperometric titrations.
- **4.** What are the advantages of rotating platinum electrode versus dropping mercury electrode?
- **5.** Write a note on amperometric titrations using two indicator electrodes.
- **6.** Describe, how will you carry out the simultaneous determination of Cl<sup>-</sup>, Br<sup>-</sup> and I<sup>-</sup>.
- 7. Using amperometric titrations how will you determine  $SO_4^{2-}$ ,  $Pb^{2+}$ ,  $Cu^{2+}$  and  $Zn^{2+}$ .





# Chapter 18

# **Potentiometric Titrations**

#### 18.1 INTRODUCTION

In potentiometric titration, a suitable electrode is immersed in the solution to be titrated. This electrode acts as an 'indicator'. The **indicator electrode** is paired with a reference electrode (a calomel electrode). The two electrodes are connected to an electronic voltmeter. With the change in the concentration of ions (caused by the addition of a titrant from the burette), the e.m.f. of the indicator electrode changes. The equivalence point is indicated by a sharp change in the electrode potential.

The potential of a metal electrode in a solution of its own ion at  $25^{\circ}$ C is given by the equation

$$E_{25} = E^{\circ} + \frac{0.0591}{n} \log C_M^{n+} \qquad \dots(i)$$

where  $E^{\circ}$  is the standard potential of the metal, n is the valency of the metal and  $C_{M}^{n+}$  is its concentration.

The concentration in fact is  $-\log [M^{n+}]$  and this is equal to  $pM^{n+}$ 

$$E_{25} = E^{\circ} - \frac{0.0591}{n} p M^{n+}$$
 ...(ii)

For hydrogen electrode  $E_{25}$  will be

$$E_{25} = E_H^O - \frac{-0.0591}{n} pH^{n+}$$
 ...(iii)

Where  $E_H^{\circ}$  is the potential of the normal H electrode,  $E_H^{\circ}$  is arbitarily taken as zero at all temperatures. This is because  $E_H^{\circ}$  the normal potential of hydrogen ions of unit activity in equilibrium with hydrogen gas at one atmospheric pressure is taken arbitrarily as zero.

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So the equation (iii) becomes

$$E_{25} = -0.0591 \text{ H}$$
 ...(*iv*)

### 18.2 PRINCIPLE OF POTENTIOMETRIC TITRATION

As already stated, in a potentiometric titration the equivalence point (end point) is detected by measuring the potential change during the titration. Figure 18.1 shows the usual apparatus for potentiometric titration.

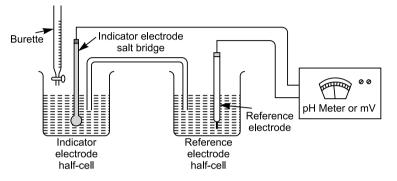


Fig. 18.1 Apparatus for potentiometric Titration

Notes: 1. Half-cell reaction: oxidation or reduction reaction occurring at an electrode

2. Electric potential (*E*) is measured in volts (*V*). The smaller unit of potential is millivolt (mV)  $\lceil 1 \text{m V} = 10^{-3} \text{V} \rceil$ 

The potential between the reference half-cell (whose potential is known) and the indicator half-cell (whose potential varies will the concentration of the solution) is measured at the start and after each addition of small amounts of titrant, and more closely near the equivalence point, when readings start to change by comparatively larger values. Appropriate precaution is taken to stir the solution after each addition (using a magnetic stirrer) so that the reactings becomes steady.

In potentiometric titration, for detecting the equivalent point, a graph is plotted between the potential and the volume of titrant (Fig. 18.2).

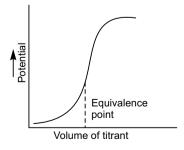


Fig. 18.2 Determination of equivalence point

Once a titration curve is plotted, the equivalence point is determined by inspecting the curve: the point where the curve is steepest (Fig. 18.3). At times, there is uncertainty in determination of the equivalence point. This uncertainty is overcome by change in potential with change in volume the reagent ( $\Delta E/\Delta V$ ) is plotted against the average volume of the reagent added (Fig. 18.3). The equivalence point corresponds to the maxima in the curve, which is obtained by extraploting the experimental points.

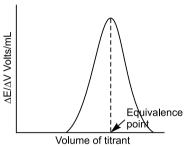


Fig. 18.3 A better procedure for determination of equivalence point

#### 18.3 INDICATOR ELECTRODE

For potentiometric titration, the appropriate indicator electrode has to be used. The main characteristic of an indicator electrode on which the choice of indicator electrode depends are:

- The potential of the indicator electrode should be related to the concentration of the species to be determined via the Nernst equation.
- Its response to various concentrations should reproducible be and rapid. A number indicator electrodes are available. These include hydrogen electrode. antimony electrode. quinhydrone electrode and glass electrode. Most commonly, glass electrode is used in potentiometric titrations. In glass electrode, the

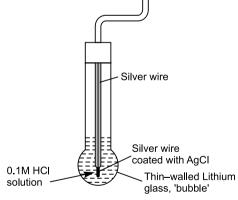


Fig. 18.4 Glass electrode

inner and the outer glass surfaces are bound to generate different potentials. We get a junction potential which is a symmetric and depends on the type of glass, age and usage of the electrode (Fig. 18.4).

#### 18.4 REFERENCE ELECTRODES

The potential of a reference electrode should remain constant during potentiometric titration. Most commonly, calomel electrode is used. It consists of metallic mercury and solid mercurous chloride (calomel) in contact with, and in equilibrium with aqueous solution of KCl. The half cell reaction of a calomel electrode is

$$Hg_2Cl_2(s) + 2e^ 2Hg(s) + 2Cl^-$$

The electromotive force is given as

$$E_{Hg_2Cl_2,Hg} \ = \ E_{Hg_2Cl_2,Hg} - \frac{0.0591}{2} \log \left[ Cl^- \right]^2$$

or 
$$E_{Hg_2Cl_2,Hg} \ = \ E_{Hg_2Cl_2,Hg}^{\circ} - 0.0591 \, log \, [Cl^{-}]$$

As seen, the potential of a reference calomel electrode is dependent only on the concentration of chloride ion. As mercurous chloride is insoluble, the concentration of chloride ion depends almost entirely on the amount of KCl used in the preparation of the electrode.

Three types of calomel electrodes and known. These are:

 Saturated Calomel Electrode (SCE). It consists of solid mercurous chloride with saturated potassium chloride solution with metallic mercury

$$E^{\circ} = +0.2415 \text{ volts.}$$

 Normal Calomel Electrode (NCE). It consists of aqueous medium containing 1F KCl.

$$E^{\circ} = 0.280 \text{ volts.}$$

Decinomal Calomel Electrode (DCE). It consists of 0.1 F KCl

$$E^{\circ} = +0.004 \text{ volts.}$$

The most commonly used is the saturated calomel electrode (SCE). Since the solubility of KCl depends on the temperature, it is necessary that temperature control is required for the operation of the electrode. Saturated calomel electrodes are commercially available (Fig 18.5). It consists of two concentric tubes, the inner tube containing an amalgamated platinum wire which is connected to external circuit. The inner tube is also filled with a paste of metallic mercury, mercurous chloride and potassium chloride. The outer tube contains saturated KCl solution having few crystals of KCl in order to maintain saturation conditions. There is a small pin hole in the inner tube which provides contact of the electrolyte with the outer tube. A porous asbestos fibre, which is sealed into the tip of the outer tube provides contact with the sample solution.

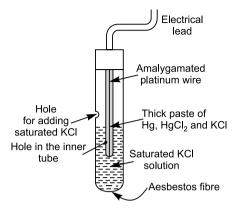


Fig. 18.5 Saturated calomel electrode

It has been suggested (Pinkhof and Treadwell) that in place of a reference cell, a compensation electrode can be used. Such an electrode has potential exactly equal to the E.M.F. of the indicator electrode at the end point. In such a setup, the end point is located by a sudden reversal of polarity. No potentiometric assembly is needed. A simple capillary electrometer is used in place of a galvanometer. When the titration starts, mercury in the electrometer moves in one direction till the end point is reached and mercury stops to move. The mercury moves in the opposite direction on further addition of the reagent. The disadvantage of this procedure is that for every titration a separate electrode system is needed.

#### 18.5 APPARATUS FOR POTENTIOMETRIC TITRATIONS

It has been stated (section 18.2) that in potentiometric titrations the indicator electrode and the reference electrode are placed in separate cells and that the two cells are connected by a salt bridge. It is more convenient to use a single cell (vessel) for housing both the electrodes. The vessel for titration is the type of a beaker. Indicator electrode and saturated calomel electrodes are suspended in the container. The standard titrant is added in small amounts using a burette. Each addition of the titrant should be preferably of the same volume. The solution in the vessel is stirred using a magnetic stirrer. The titration vessel should be kept closed and there should be arrangement for bubbling in inert gas and its outlet. This is necessary for some potentiometric titrations.

After each addition of the titrant, the solution is sturred to attain homogenity and electrode potential *vs* SCE measured. The end point is determined as shown in Figs. 18.2 and 18.3.

A diagrammatic representation of a simple apparatus for potentiometric titrations is shown in Fig. 18.6.

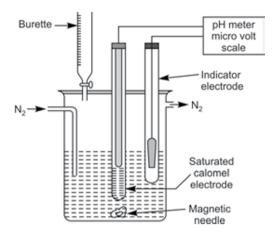


Fig. 18.6 A simple apparatus for potentiometric titration

#### 18.6 APPLICATIONS OF POTENTIOMETRIC TITRATIONS

We come across following types of potentiometric titrations:

**Neutralisation Titrations** 

Oxidation-Reduction Titration

**Precipitation Titrations** 

Complexometric Titrations.

#### 18.6.1 Neutralisation Titrations

Neutralisation titrations, commonly known as acid-base titrations are always accompanied by changes in the concentration of H<sup>+</sup> and OH<sup>-</sup> ions. The apparatus used for potentiometric neutralisation titrations is shown in Fig. 18.7.

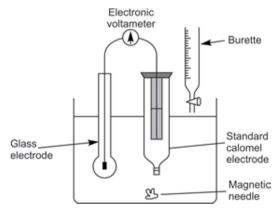


Fig. 18.7 Apparatus for potentiometric acid-base titrations

A glass electrode (or a hydrogen electrodes) is immersed in solution of acid whose strength is to be determined. The glass electrode is coupled with a standard calomel electrode.

The cell thus formed is connected to a potentiometer or electronic voltameter. On addition of alkali from the burette, the pH of the solution changes and the e.m.f. of the cell also changes with pH of the solution in accordance of the reaction.

$$E = E^{\circ} + 0.0591 \text{ pH}$$

The standard alkali solution is then added from the burette in small volumes. After each addtion, the solution is stirred and e.m.f. of the cell recorded. The e.m.f. is then plotted against the volume of alkali added. The shape of the curve for titration of a strong acid against strong alkali (HCl versus NaOH) is shown in Fig. 18.8 (a). The steepest portion of the curve indicates the equivalence point. However, in case solutions are very ditute, or weak acids or bases, are involed, the steepness of the curve is less market and it is difficult to judge the end point. In such a case, plotting the slope of the curve,  $\Delta E/\Delta V$  against volume of alkali is done. The end-point indicated by the maximum of the curve [Fig. 18.8(b)].

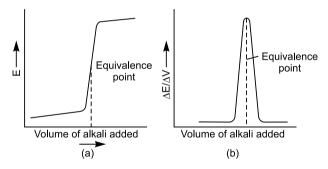


Fig. 18.8 Potentiometric titration curve of an acid and a base

The potentiometric titrations are particularly useful where visual end point detection (in the normal volumetric titration) is difficult in presence of turbidity or coloured species.

For acids and bases having dissociation constants greater than  $10^{-8}$ , aqueous medium give satisfactory results. However, for weaker acids and bases, the reactions are incomplete resulting in sufficient changes near the end point. In such cases, non-aqueous solvent is used. For example, phenol ( $K_a = 10^{-10}$ ) can be titrated against a base in non-aqueous medium.

#### 18.6.2 Oxidation-Reduction Titrations

Oxidation-reduction titrations are also known as **redox titrations**. A typical example of an redox titration is the titration of ferrous ions ( $Fe^{2+}$ ) with ceric ions ( $Ce^{4+}$ ). In this case  $Fe^{2+}$  is oxidised to  $Fe^{3+}$  ion, while  $Ce^{4+}$  is reduced to  $Ce^{3+}$  ion.

$$Fe^{2+} + Ce^{4+} \rightarrow Fe^{3+} + Ce^{3+}$$

The titration is carried out by using the apparatus as shown in Fig. 18.9. A strip of shining platium dipping in the solution of  $Fe^{2+}$  ions is the indicator electrode, which is connected to a standard calomel electrode. A burette is used to add  $Ce^{4+}$  solution and the cell potential, E, is recorded after each additon.

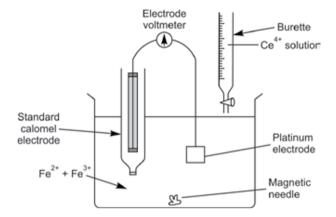


Fig. 18.9 Apparatus for potentiometric titration of Fe<sup>2+</sup> with Ce<sup>4+</sup>

The potential of the platinum electrode depends on the ratio  $[Fe^{3+}]/[Fe^{2+}]$ . Also, the potential of the cell, E changes with the change of the ratio  $[Fe^{3+}]/[Fe^{2+}]$ . In fact, the cell potential changes with the addition of  $Ce^{4+}$  ions from the burette. Fig. 18.10 shows, how the potential of the cell changes as the titration proceeds. There is a sharp rise in potential at the equivalence point, which indicates the end point.

Potentiometric titrations are particularly useful for coloured solutions in which an indicator cannot be used.

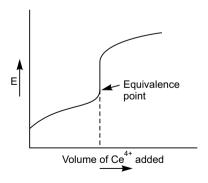


Fig. 18.10 Potentiometric titration curve of Fe<sup>2+</sup> ions and Ce<sup>4+</sup> ions

#### 18.6.3 Precipitation Titrations

A typical precipitation titration is that of sodium chloride against silver nitrate solution. The apparatus for this purpose is shown in Fig. 18.11. A silver electrode dipping in unknown sodium chloride solution is coupled with a calomel electrode through a KNO3 salt bridge, so that KCl solution does not diffuse into the sample. In case the calomel electrode comes in direct contact with a solution containing excess of silver ions, there is a possibility that may seep through the sintered base of the calomel electrode and react to form an insoluble layer of silver chloride.

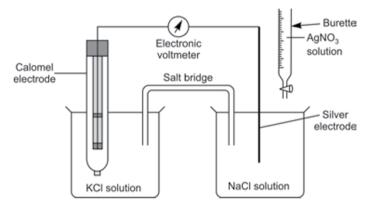


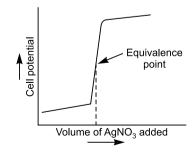
Fig. 18.11 Potentiometric Titration of sodium choride against silver nitrate solution

Any change in the cell potential is due to changes in the concentration of Ag<sup>+</sup> ions around the silver electrode

$$Ag^+ + e \rightarrow Ag$$

The concentration of Ag<sup>+</sup> is intially zero. However, as AgNO<sub>3</sub> is added from the burette, silver choride gets precipitated. At this stage, the solution will contain

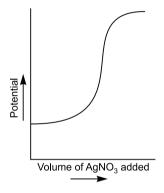
a small concentration of Ag<sup>+</sup> ions formed due to slight dissociation of AgCl. This concentration increases slightly as Clions are removed in order to maintain the solubility product,  $K_{sp} = [Ag^+]$ [Cl-]. After the equivalence point, the concentration of Ag<sup>+</sup> ions and therefore, the silver electrode potential rises sharply owing to the presence of excess of Ag<sup>+</sup> ions. The volume of AgNO<sub>2</sub> solution used to reach the equivalence Fig. 18.12 Potentiometric titration curve point is shown in Fig. 18.12.



for NaCl and Ag NO<sub>3</sub>

#### 18.6.4 Complexometric Titrations

Most of the potentiometric complexometric titrations have been carried out with EDTA. A typical example is the determination of calcium ions in an unknown sample by potentiometric EDTA titration. A simple apparatus (Fig. 18.6) for potentiometric titration is used. It consists of a mercury indicator electrode and a saturated calomel reference electrode suspended in a titration vessel containing Ca<sup>2+</sup> ions. A small concentration (10<sup>-4</sup>M) of mercury (II) EDTA complex is introduced in the titration vessel. Finally, the mixture is titrated against standard EDTA solution. The potential of the mercury indicator electrode is measured after each addition of titrant. The potential of the indicator electrode against the volume of the titrant is plotted (Fig. 18.13).



**Fig. 18.13** Titration curve for the titration of Ca<sup>2+</sup> ions with EDTA. Ammonia buffer (pH 10) is used

Using potentiometric EDTA titrations, Mg<sup>2+</sup> and Zn<sup>2+</sup> can be determined.

#### 18.7 DIFFERENTIAL TITRATIONS

We have seen earlier (Fig 18.2) that a graph plotted between the potential and the volume of titrant possesses a distinct maxima near the equivalence point. Such a curve can also be obtained using differential titration. The procedure (developed by Cox) required two identical electrodes, one of which is shielded from the bulk of the solution (Fig. 18.14). One electrode is housed in a long test tube with side arm. There is a small hole (about 1mm) in the bottom of the tube to provide contact with the bulk of the solution. Around the shielded electrode, the composition of the solution changes due to restricted acess on addition of the titrant.

As stated, there is a very small difference in composition of the solution around the two electrode, but this gives rise to the difference in potential ( $\Delta E$ ) between the electrodes. This difference in mangaged and the solution homogenised by sqeezing the rubber bulb. At this stage  $\Delta E$  again becomes zero. At equivalence point  $\Delta E$  is

maximum. An advantage of differential titration is that there is no need to use a reference electrode and salt bridge. Normally, the end point is sharp.

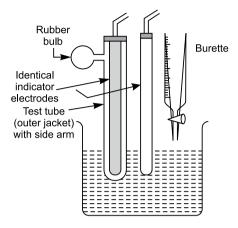


Fig. 18.14 Differential titration (apparatus)

#### **18.8 AUTOMATIC TITRATIONS**

A potentiometric titration is a time consuming procedure. This problem has been overcome by using automatic equipment for performing the titration and also recording the titration curve. A typical instrument for automatic titration provides a titration curve of potential vs. reagent volume or, in some cases  $\Delta E/\Delta V$  or  $\Delta^2 E/\Delta V^2$  against volume. On examination of the curve, it is easy to determine the end point. The titration automatically stopped when the potential reaches a predetermined value. The instrument employes a burette system with a selenoid operator value which controls the flow. The titrant is added very slowly so that the end point is not over run.

#### 18.9 ADVANTAGES OF POTENTIOMETRIC TITRATIONS

- (i) The apparatus empolyed for potentiometric titrations is readily available and is dependable.
- (ii) It is easy to find the end point by referring to the titration curves.
- (iii) This method is particularly used for coloured solutions where indicator does not work.
- (*iv*) A distinct advantage is that two or more constituents can be determined in a single operation. For example, Br<sup>-</sup> and I<sup>-</sup> may be titrated together.

## EXERCISES

- 1. What are potentiometric titrations?
- 2. Explain the principle involved in potentiometric titrations.
- **3.** What are the advantages of potentiometric tiration over indicator methods?
- 4. Write notes on:
  - (a) Indicator Electrode
  - (b) Reference Electrode.
- **5.** Describe the apparatus for potentiometric titrations.
- **6.** What is a compensation electrode? Explain.
- 7. Discuss the applications of potentiometric titrations
- **8.** How will you determined the concentration of Fe<sup>2+</sup> using potentiometric titration?
- **9.** Explain the following:
  - (a) Differential Titrations
  - (b) Automatic Titrations.





# Chapter 19

### **Spectrophotometric Titrations**

#### 19.1 INTRODUCTION

Generally, in normal titrimetric methods, the equivalence point of a reaction is visually detected by the colour produced by an indicator (as in acid-base titrations etc.) or by the colour of the reactant (permanganate etc). Such titrations give results within one per cent accuracy. In titrations, if the colour change is gradual or the colour change does not contrast sharply, accurate results are not obtained. All such problems can be overcome if photometric or spectrophotometric titrations are carried out.

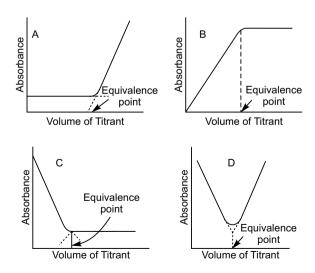


Fig. 19.1 Various titration curves. Determination of equivalence points.

In spectrophotometric titrations, a spectrophotometer is used to determine the equivalence point. The method consists in placing the titration vessel directly in the path of the light of the instrument. The absorbance of the solution is then determined after adding titrant and a plot of absorbance as a function of the volume of the tritrant is made. Typical titrations are given in Fig. 19.1.

When the titration is complete, the titration curve is made of two straight lines. The intersection of the two lines is the equivalence point. This is similar to amperometric and conductometric titrations. If on the other hand, the reaction is incomplete, there is an appreciable curvature in the region of the equivalence point. In such a case, extrapolation of the two linear segments of the curve to their intersection gives the equivalence point volume.

Explanations of the curves A to D in Fig. 19.1.

- (i) Curve A is typical of a case when only the titrant absorbs. A typical example is the titration of arsenic (III) with bromate-bromide. In this case, the absorbance readings are taken at the wavelength where bromine absorbs. As long as arsenic (III) remains in the solution, the absorbance remains unchanged as the product does not absorb in that region. When all the arsenic (III) has reacted with the titrant, there is increase in absorbance due to colour of the titrant (bromine).
- (ii) Curve B is typical of a case in which only the product of the reaction absorbs. An example is the titration of Cu (II) with ethylene diaminetetracetic acid (EDTA). The titration is carried out at a wavelength 745 nm, since at this position only the Cu-EDTA complex possesses a much greater absorption compared to copper (II) solution alone.
- (iii) Curve C is characteristic in case where only the substance being titrated absorbs. An example is the titration of p-toluidine in butanol with perchloric acid at 290 nm. At 290 nm p-toluidine absorbs sharply where as perchloric acid does not absorb in this region. As soon as the whole of p-toluidine has reacted with perchlore acid, the absorbance becomes constant after the equivalence point.
- (iv) Curve D is characteristic of cases when a coloured analyte is converted into a colourless product by a coloured titrant. On addition of titrant, the colour of the analyte starts fading because of the formation of a colourless product. After the equivalence point, the absorbance rises again due to the colour of the titrant.

#### 19.2 PROCEDURE OF TITRATION

For spectrophotometric titrations, a specially made titration cell (Fig. 19.2) of 5 to 100 mL capacity is used. The cell fits into the cell compartment of the spectrophotometer. The cell is made of perspex sheet (which is opaque to ultraviolet

light) in which there are two openings to accommodate circular quartz windows. These windows are arranged in such a way so that the beam of monochromatic light passes through their centres to the photoelectric cell. In the cell there are two small openings, one for inserting the tip of a microburette and another for a micro stirrer. The whole cell, except the quartz windows is covered with black paper in order to exclude any extraneous light.

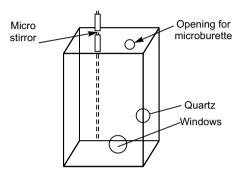


Fig. 19.2 Titration cell

The solution to be titrated is taken in the cell, which is then placed in the path of the light of the spectrophotometer, which in turn is adjusted to the wavelength at which experiment has to be carried out. The instrument is set to read zero absorbance (if the reactant is colourless) or to some other starting value in case the reactant is coloured. After the addion of a known volume of titrant to the stirred solution, the absorbance is noted. This is repeated at a number of points before the end point and several points after the end point. A graph is finally plotted of the absorbance against the volume of the titrant added. This graph is used to determine the equivalence point.

Since the absorbance depends on concentration, so the effect of dilution should also be taken into account. The error due to dilution can be overcome by the following pocedure:

- (i) It is best that the titrant should be more concentrated than the solution to be titrated. Thus, there is neligible change in volume.
- (ii) Alternatively, a simple correction factor in calculation be included.

$$A = A' - \frac{V + v}{V}$$

where A and A' are the corrected and measured absorbances respectively. V is the original volume of solution and v is the volume of titrant added.

#### 19.3 APPLICATIONS

(i) Acid Base Titrations: Titration of phenol with NaOH. The absorbance due to the formation of phenolate ion is followed.

followed.

- (ii) Redox Titrations: Titration of Ce (III) with Co(III). Formation of Ce (IV) is followed.
- (iii) Complexometric Titrations: Titration of Fe (III) with EDTA. Disappearance of Fe sulphosalicyclic acid complex is followed.Cu (II) can be titration with EDTA. Formation of Cu-EDTA complex is
- (iv) Precipitation Titrations: Titration of  $SO_4^{2-}$  ions with Ba(II) ions. Appearance of turbidity is followed.

## EXERCISES #

- 1. What type of plot of absorbance and volume of titrant is obtained in the following:
  - (a) Titration of As (III) with bromate-bromide
  - (b) Titration of Cu (II) with EDTA
  - (c) Titration of p-toluidine in butanol with perchloric acid.
- 2. Describe the procedure of spectrophotometric titrations.
- **3.** In spectrophotometric titrations how will you take into account the error due to dilution.
- 4. Give application of spectrophotometric titrations.





## Chapter 20

### **High Frequency Titrations**

#### 20.1 INTRODUCTION

In an electric field, the conductivity of a solution is due to the mobility of the ions. If alternating current is applied to the solution, the ions still move a short distance between alterations. The ions will have no time to gain full speed if the current applied exceeds 1 Mc/sec. At such high frequencies, molecular polarisation is more important.

On placing a molecule in an electric field, the electrons are naturally attracted towards anode and the nuclei are attracted towards cathode. This results in some type of distortion of the molecule and is called *distortion polarisation*. This is a temperory effect since it disappears on removal of the field. Molecules, which have dipole moments, like water, chloroform, nitrobenzene etc., tend to get oriented in such a way that the positive is towards negative and the negative towards positive. Thus, such molecules show orientation polarisation in addition to the distortion polarisation. During the flow of current under applied field for a short time, both types of polarisation take place. The duration of the polarisation current is very small and is virtually insignificant at low frequencies. However, at high frequencies (radio frequencies) both the polarisation and conduction current become of the same magnitude and so cannot be neglected.

#### 20.2 INSTRUMENT

The technique of High Frequency titration was developed by F.W. Jensen and A.L. Parrack (1946). The instrument used for this titration is quite different. In this technique, the electrodes are placed outside the vessel and there is no direct contact with the solution. The conductivity cell can be made of any glass or ceramic vessel. Alternating current at frequencies more than one Mc/Sec. affects

the conductance and also the capacitance of the solution placed within the field. The vessel containing the solution is placed between the plates of a cpacitor (carrying the high frequency current) or in the field of an induction coil. The induction coil or the capacitor being part of high frequency oscillator circuit, any changes in the composition of the solution will affect changes in the oscillator or change in the plate and grid currents and voltages.

Two types of cells are used for high frequency titrations. These are capactive type or induction type (Fig. 20.1).

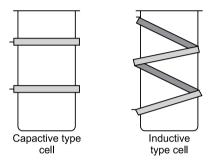


Fig. 20.1 The Cells

In the capactive type cell, capacitance is a variable parameter, which depends on the dielectric constant of the solution. However, in the induction type cell, inductivity of the cell is the variable parameter, which is almost independent of the nature of solution and its resistance.

Two methods are used for high frequency titrations. These are capacitative cell method and induction cell method. In the former viz., capacitative cell method, the cell containing the solution is placed between the plates of a capacitor having high frequency. However, the latter method, viz. induction cell method, two bands of silver or copper are put around the outer part of the vessel. These bands are connected to high frequency voltage supply. In this method the cell containing the solution is placed inside the coil in the tuned circuit of the oscillator.

The oscillator circuit has capacitance C, an inductor L and a resistive component R attributed to both (Fig. 20.2). On adjustment of the circuit capacitance a condition, known as resonance is attained. On discharge of the condenser through inductance, the current surges back and forth from the inductor to capacitor at the frequency (f)given by the expression.

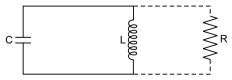


Fig. 20.2 Circuit of an Oscillator

$$f = \frac{1}{2\pi\sqrt{LC}} \qquad \dots(i)$$

The current decays rapidly, since the circuit always has some resistance. In case the resonance circuit (Fig. 20.2) is coupled to the input of an electronic amplifier (Fig. 20.3), the voltage impressed on the grid will alternate at the frequency given by equation (i). So the output voltage ( $\mu_{eg}$ ) at the plate will also alternate at the same frequency.

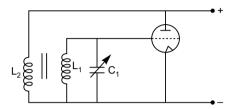


Fig. 20.3 Resonance Circuit Coupled to electronic amplifier

If an inductance  $L_2$  is placed in the plate circuit and coupled to  $L_1$  in the grid circuit (as shown in Fig. 20.3), amplified voltage will appear across  $L_2$ . The coil  $L_2$  can serve to convey output voltage back to the input grid circuit if the windings of the coils are suitably arranged. The resonant circuit can be placed in the grid or plate circuits or coupled via. a common inductance.

#### 20.3 HIGH FREQUENCY TITRATIONS

The diagrammatic representation of a high frequency titrimeter is shown in Fig. 20.4. The sample cell containing the solution is put in parallel with a calibrated capacitor. Alternatively, the cell may be a tube upon which the coil is wound, with the corresponding alternations in capacitance between the windings and in inductance. It is necessary to remove the exact amount of capacitance that was added to the sample.

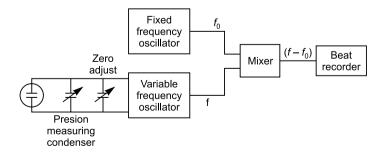


Fig. 20.4 Diagrammatic representation of a high frequency titrimeter

An alternative convenient method consist in measuring the output frequencies of two identical oscillator circuits. One of the oscillator circuit contains the sample cell as part of the oscillator capacitance (f) and the other serves as a reference unit  $(f_0)$ . The output from the two oscillator circuits is fed into a mixer unit. The low-frequency difference  $(f-f_0)$  obtained from the mixer unit is directly proportional to the change in the high frequency capacitance of the cells. Such instruments are more stable in practice than direct reading instruments.

#### 20.4 APPLICATIONS OF HIGH FREQUENCY METHODS

#### 20.4.1 Acid-Base Titrations

During the titration, the temperature must be kept constant. Before carrying out the high frequency titrations, it is helpful to determine the response of the instrument as a function of the electrolyte concentration. Each cell in any instrument has an optimum range for which maximum responce in the test solution will be obtained. Some high frequency conductance and capictance curves are shown in Figs. 20.5 and 20.6.

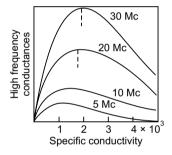


Fig. 20.5 High frequency conductance curves of some systems

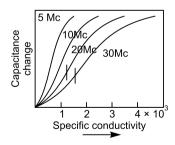


Fig. 20.6 High frequency capacitance curves of some systems

#### 20.4.2 Measurement of Dielectric Constant

Dielectric constant is a measure of a solvent's ability to insulate opposite charges from each other. The polarity of a solvent depends on its dielectric constant. In fact,

solvents with high dielectric constants are better solvents for ions than are solvents with low dielectric constant.

If a non-conducting liquid is placed between two metal plates (Fig. 20.7), the system behaves as two capacitors,  $C_g$  and  $C_g$  in series. The capacitance  $C_g$  will vary from fixed value Co, which is determined by the geometry of the cell filled with air to the value D dependent on the dielectric constant the sample; that is

$$Cs = CoD$$

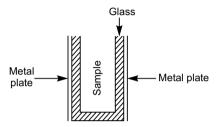


Fig. 20.7 Sample placed between two metal plates

The value of  $C_0$  is dependent on the thickness of the glass walls, the effective plate area, and dielectric constant of the container. Fig 20.8 gives a plot the cell response with solutions of different dielectric constants.

The Delectric constant of some comman liquids is given in Table 20.1. The test material is uniformly packed between two parallel plates which serves as a measuring cylinder.

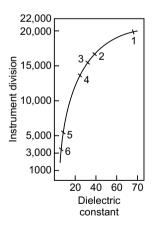


Fig. 20.8 Instrument response as a function of dielectric constant : 1. Water, 2. Methanol, 3. Ethanol, 4. Acetone, 5. Chloroform and 6. Benzene

Substance	Dielectric Constant	Substance	Dielectric Constant
Water	81	Benzol alcohol	13.1
Formic acid	58.5	Phenol	9.78
Acetonitrile	37.5	Aniline	6.89
Nitrobenzene	34.8	Acetic acid	6.15
Methanol	32.6	Chloroform	4.806
Ethanol	24.3	Benzene	2.379
Acetone	20.7	Carbontetrachloride	2.238

Table 20.1 Dielectric Constant of same liquids

#### 20.4.3 Analysis of Binary Mixtures

Binary mixtures can be analysed with the help of a calibration curve prepared from known solution.

#### 20.4.4 Complexometric Titrations

Using the method of high frequency titration ions like Mg<sup>2+</sup>, Ca<sup>2+</sup>, La<sup>3+</sup> on titration with EDTA give sharp end points. Similarly Ni<sup>2+</sup> is analysed by complexing with dimethyl glyoxime.

#### 20.5 ADVANTAGES OF HIGH FREQUENCY TITRATIONS

- (i) No special type of conductivity cell is required.
- (ii) Metal plates are not placed in the solution thus eliminating polarization of electrides.
- (iii) The variation of oscillator current can be plotted as a function of time by connecting the timer to a potentiometric recorder.
- (*iv*) The method is useful for organic compound, complexometric titration and precipitation titrations.



- 1. Discuss the principles involved in high frequency titrimetry.
- 2. Give the applications of high frequency titrations.
- Discuss the advantages and disadvantage of these titrations.
   Hints: The technique is complicated and tuning is difficult and the titrimeter less sensitive than the Wheatstone's bridge especially for quantitative work.
- 4. Using high frequency titration, how with you determine dielectric constant.

**Procedure:** The test-tube holder is connected to a high frequency instrument. A calibration curve of instrument response as a function of dielectric constant is prepared by successively measuring pure benzene, chloroform, acetone, ethanol, methanol and water. Finally determine the instrument reading of the unknown solution and record the dielectric constant by reference to the calibration curve.





# Chapter 21

### pH Measurements

#### 21.1 INTRODUCTION

All aqueous solutions, whether acidic, alkaline or neutral contain both hydrogen  $(H^+)$  and hydroxyl  $(OH^-)$  ions. The product of  $H^+$  and  $OH^-$  is invariably constant and is equal to  $1\times 10^{-4}$  at  $25^{\circ}$ C. The greater concentration of  $H^+$  or  $OH^-$  shows whether a particular solution is acidic or alkaline. In fact, the acidity or alkalinity of a solution is expressed by the concentration of hydrogen ions only. A scale for expressing the hydrogen ion concentration was suggested by Soreen (1909) and is called pH scale. The pH of a solution is defined as the negative logarithm of the hydrogen ion concentration  $(H^+)$ .

$$pH = -log \ [H^+]$$
 or 
$$[H^+] = 10^{-pH}$$
 For pure water,  $[H^+]$  is equal to  $1\times 10^{-7}.$ 

∴ pH pure water is 7.

Table 21.1 Relation of [H+] with pH

[H <sup>+</sup> ]	рН	Nature of solution	[H <sup>+</sup> ]	рН	Nature of solution
10 <sup>-1</sup> 10 <sup>-2</sup> 10 <sup>-3</sup> 10 <sup>-4</sup> 10 <sup>-5</sup> 10 <sup>-6</sup>	1.00 2.00 3.00 4.00 5.00 6.00 7.00	Strongly acidic  Weakly acidic Neutral	10 <sup>-8</sup> 10 <sup>-9</sup> 10 <sup>-10</sup> 10 <sup>-11</sup> 10 <sup>-12</sup> 10 <sup>-13</sup>	8.00 9.00 10.00 11.00 12.00 13.00	Weakly alkaline Strongly alkaline

The solution is acidic, if pH is less than 7 and is alkaline, if pH is more than 7. Thus, greater is the hydrogen ion concentration, the lower the pH (and the solution is acidic) Table 21.1 gives a comparison of the hydrogen ion concentration with pH and nature of the solution.

It is interesting to know the pH values of some acids and bases (Table 21.2).

Acids		Bases			
Solution	рН		Solution	рН	
Hydrochloric acid	1.0	I	Sodium bicarbonate	8.4	ı
Sulphuric acid	1.2		Borax	9.2	
Phosphoric acid	1.5	acidity	Ammonia	11.3	basicity
Acetic acid	2.9	ъ	Sodium carbonate	11.6	bas
Boric acid	5.2	7	Sodium hydroxide	13.0	♥

**Table 21.2** pH values of some acids and bases (0.1N solution at 20°C)

On the basis of pH determination, gastric juice is acidic (pH 1.0–2.0) and Blood and tears are slightly alkaline (pH 7.4 for both).

Any assembly of apparatus, which is used to measure pH determines the electromotive force. It has now been recognised that it is the activity rather than the concentration of hydrogen ion that is responsible for the determination of electromotive force. In view of this, the pH of a standard solution (pH<sub>s</sub>) is represented by the equation

$$pH_S = -log a_H$$

where, a<sub>H</sub> is hydrogen ion activity.

Also, 
$$a_H = f_H m_H$$

where,  $f_H$  is the activity coefficient of hydrogen ion and  $m_H$  is the molality of hydrogen ion. As the activity coefficient of single ionic species can not be determined, a scale based on certified pH values of a number of buffer solutions (Table 21.3) has been developed by National Bureau of standards (USA). All these buffers are assigned fixed pHs.

<b>Table 21.3</b> Standard buffers and th	eir pH values as per NBS standard

Solution (m = molality)	pH at		
(III – Molanty)	25°	30°	38°
Saturated potassium hydrogen tartarate at 25°	3.557	3.552	3.548
0.05 M potassium hydrogen phthalate	4.008	4.015	4.030

(Contd...)

Solution	pH at		
(m = molality)	25°	30°	38°
0.25 M potassium dihydrogen phosphate and 0.025 M disodium hydrogen phosphate	6.865	6.853	6.840
$0.008695~{ m M~KH}_2~{ m PO}_4~{ m and} \ 0.03043~{ m M~Na}_2{ m HPO}_4$	7.413	7.400	7.381
0.010 M Borox	9.180	9.139	9.081

## 21.2 DETERMINATION OF pH OF A SOLUTION BY POTENTIOMETRY

The procedure involves setting up of a half cell with the test solution as an electrolyte. The e.m.f. of the cell is dependent on the concentration of H<sup>+</sup> ions or pH of the solution. The e.m.f. of the half cell is determined by coupling it with another standard half cell and measuring the e.m.f. of the complete cell. Folllowing standard electrodes are commonly used.

- Hydrogen electrode
- Quinhydrone electrode
- Glass electrode.

#### 21.2.1 Determination of pH using Hydrogen Electrode

The hydrogen electrode is a standard for pH measurements. It consists of a small platinum sheet or wire coated with friely divided platinum black. A standard hydrogen electrode is coupled with another hydrogen electrode which contains the solution of unknown pH. In both the hydrogen cells hydrogen gas is passed at 1 atm pressure and 25° (Fig. 21.1).

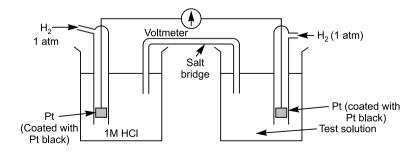


Fig. 21.1 Determination of pH with hydrogen electrode

The e.m.f. of the complete cell.

$$Pt | H_2 (1 atm) | H^+ (1M) | H^+ (unknown) | H_2 (1 atm) | Pt$$

is experimentally recorded.

The second electrode reaction is

$$H^+ + e^- \rightarrow \frac{1}{2} H_2$$

The electrode potential of the second half-cell is given by Nernst equation

$$E = E^{0} + \frac{2.303RT}{nF} \log \frac{[H^{+}]}{PH_{2}^{1/2}}$$

Since

$$pH_2^{\frac{1}{2}} = 1$$
 and  $E^0 = 0$ , we get

$$E = \frac{2.303 RT}{nF} \log[H^+]$$

Substituting the values R, T and n (charge number of  $H^+$ ) and F (Faraday constant), the above equation becomes

$$E = 0.0591 \log [H^+]$$
  
= -0.0591 pH ...(1)

The e.m.f.  $(E_{cell})$  of the cell is given as

$$E_{\rm cell} = E_{\rm right} - E_{\rm left}$$

The left hand electrode is the standard hydrogen electrode and its e.m.f. will be zero.

: 
$$E_{\text{cell}} = E_{\text{right}} = -(-0.0591) \text{ pH}$$

Using the equation (1), we get

$$E_{\text{cell}} = -(-0.0591 \times \text{pH})$$
$$= 0.0591 \times \text{pH}$$
$$\text{pH} = \frac{E_{\text{cell}}}{0.0591}$$

or

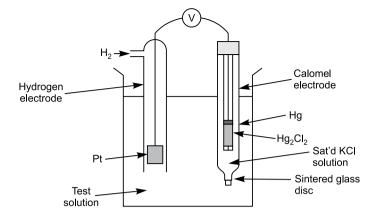
Knowing the observed value of e.m.f. of the cell, the pH of the test solution can be calculated.

In the above procedure (Fig. 21.1), in place of standard hydrogen electrode, a standard calomel electrode can also be used (Fig. 21.2).

The complete cell (Fig. 21.2) is represented as

Pt, 
$$\rm H_2$$
 (1 atm) |  $\rm H^+$ unknown || KCl Sat' d soln. |  $\rm Hg_2Cl_2$  |  $\rm Hg$ 

By noting the e.m.f. of the cell with the help of a voltmeter, the pH of the unknown solution can be calculated as shown below.



**Fig. 21.2** Determination of pH with hydrogen electrode coupled with calomel electrode

The e.m.f. of the cell is given by

$$\begin{split} E_{\rm cell} &= E_{\rm right} - E_{\rm left} \\ &= 0.2415 - (-0.0591 \times {\rm pH}) \qquad ... [{\rm from~eq.~}(i)] \\ E_{\rm cell} &= 0.0415 + 0.0591 \times {\rm pH} \\ \\ \therefore \qquad \qquad {\rm pH} &= \frac{E_{cell} - 0.0415}{0.0591} \end{split}$$

#### Advantages of Hydrogen Electrode

Hydrogen electrode is highly accurate, can be used over the entire pH range and is a fundamental standard pH electrode.

#### Disadvantages of Hydrogen Electrode

Hydrogen electrode cannot be used in presence of strong oxidsing agents, solutions having reducible ions or substances having positive electrode potential (*e.g.*, cu, Ag). Additionally, the hydrogen electrode is difficult to set up and is inconvenient to use in remote places.

#### 21.2.2 Determination of pH using Glass Electrode

A glass electrode provides one of the easiest method for measuring the pH of a solution. A simple type of glass electrode consist of a glass tube having a thin walled bulb at the lower end. The bulb contains 1M HCl solution. A silver wire coated with silver chloride is at its lower end. The lower end of this silver wire dips into HCl forming silver-silver chloride electrode. The glass electrode is represented as:

Ag, AgCl | 1M HCl : H<sup>+</sup> Test solution. It is represented as shown in Fig. 21.3.



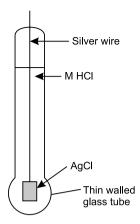


Fig. 21.3 A simple glass electrode

For determination of pH of a solution, the glass electrode is immersed in the solution of unknown pH and is coupled with a standard calomel electrode (SCE) (Fig. 21.4).

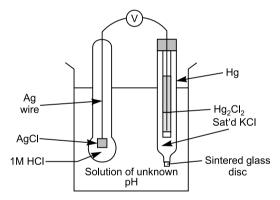


Fig. 21.4 A glass electrode coupled with a standard calomel electrode (SCE)

The e.m.f. of the complete cell is represented as can be determined experimentially.

Ag, AgCl | 1M HCl | glass | Solution of unknown pH 
$$\parallel$$
 SCE

The potential of the glass electrode,  $E_G$  at 25° is given by the equation

$$E_G = E_G^0 + \frac{2.303 RT}{F} \text{ pH}$$
  
=  $E_G^0 + 0.0591 \text{ pH}$ 

We know that

$$E_{\text{cell}} = E_R - E_L$$
 (R—Right cell, L—Left cell)

$$E_{\text{cell}} = E_{SCE} - E_G$$

Substituting the value of  $E_G$  from equation (i), we get

$$E_{\text{cell}} = E_{SCE} - E_{G}^{0} - 0.0591 \times \text{pH}$$

$$\therefore \qquad \text{pH} = \frac{E_{SCE} - E_{G}^{0} - E_{\text{cell}}}{0.0591} \qquad ...(ii)$$

 $E_{\rm cell}$  is found experimentally and the value of the potential of calomel electrode is known.

 $\therefore$  We can find pH of the given solution, if  $E_G$  is known. This can be determined by using a solution of known pH is the cell and measuring  $E_{\text{cell}}$ . The value of  $E_G^0$  is constant for a particular glass electrode and can be used for any subsequent determination of pH of unknown solutions will help of equation (ii).

#### **Advantages and Disadvantages of Glass Electrode**

A glass electrode is simple to operate, is not easily poisoned (unlike the hydrogen electrode which migh get poisoned by test solution) and the results are unffected by strong oxidising and reducing agent. In view of all this, a glass electrode is used universally.

However, a disadvantage, associated with glass electrode is that since  $E_G^0$ . depends on a particular glass electrode, it is not a universal constant. Also, glass electrode cannot be used in strongly alkaline solutions and fluoride solutions.

#### 21.2.3 Determination of pH using Quinhydrone Electrode

Quinhydrone electrode involves a redox reaction between quinone (Q) and hydroquinone (Q $H_2$ )

$$Q + 2H^{+} + 2e^{-} \Longrightarrow QH_{2}$$

OH

OH

OH

Hydroquinone

$$Q + 2H^{+} + 2e^{-} \Longrightarrow QH_{2}$$

The hydroquinone half cell consists of a platinum strip immersed in a saturated solution of quinhydrone at a definite  $H^+$  ion concentration (buffer solution). The quinhydrone is a molecular compound (Fig. 21.5), which gives equimolar amounts of quinone and hydroquinone insolution.

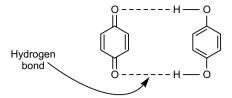


Fig. 21.5 Representation of quinhydrone

The electrode system is represented as

Pt/QH<sub>2</sub>, QH<sup>+</sup>

Quinhydrone electrode is represented as shown in Fig. 21.6.

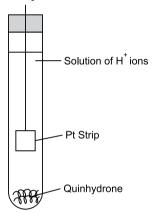


Fig. 21.6 Quinhydrone electrode

The pH of a solution in determined by suspending a platinum electrode in a solution whose pH is to be determined. The solution is saturated with quinhydrone. The platinum electrode is coupled with a standard calomel electrode (SCE) (Fig. 21.7).

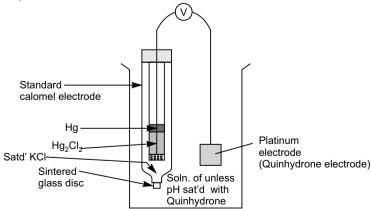


Fig. 21.7 Quinhydrone electrode coupled with a standard calomel electrode (SCE)

The complete cell is represented as

SCE||Solution of unknown pH saturated with quinhydrone||Pt

The e.m.f. of the complete cell  $(E_{cell})$  is determined experimentally.

The reduction half-cell reaction of quinhydrone electrode is

$$Q + 2H^+ + 2e^- \Longrightarrow QH_2$$

The potential  $(E_{\mathcal{Q}})$ , of the quinhydrone electrode depends on the concentration of the  $\mathrm{H}^+$  ions in solution.

On the basis or Nernst equation, the potential of hydroquinine electrode ( $E_{\it Q}$ ) is given by the equation

$$E_Q = E_Q^0 - \frac{2.302 \, RT}{F} \log [H^+]$$
  
 $E_Q = E_Q^0 + \frac{2.303 \, RT}{F} \text{pH}$ 

or

The standard reduction potential,  $E_O^0$ , of the quinhydrone electrode is 0.6996.

$$E_O = 0.6996 + 0.0591 \times \text{pH}$$
 ...(*i*)

It is known that

$$\begin{split} E_{\rm cell} &= E_{\rm right} - E_{\rm left} \\ E_{\rm cell} &= E_O - E_{SCE} \end{split}$$

Substituting the value of e.m.f. of standard calomel electrode,  $E_{SCE}$ , which is 0.2415, and the value of  $E_{O}$  form equation (i), we get

$$\begin{split} E_{\rm cell} &=~0.6996 + 0.0591 \times \rm pH - 0.2415 \\ 0.0591~\rm pH &=~0.4581 - E_{\rm cell} \\ \rm pH &=~\frac{0.04581 - E_{\rm cell}}{0.0591} \end{split}$$

where

٠.

#### Advantages and Disadvantages of Quinhydrone Electrode

The quinhydrone electrode is easily set up by simply immersing a platinum strip into the test solution (which is saturated with quinhydrone). Even in presence of oxidising agents, the pH values are accurate.

However, a disadvantage associated with quinhydrone electrode is that satisfactory results are not obtained for solutions whose pH is more than 8.5. This is due to ionisation or oxidation of hydroquinone.

#### 21.3 DETERMINATION OF pH USING A pH METER

Direct reading pH meters are commercially available and are used for finding the pH of a solution. Before using, the pH meter is first calibrated with a buffer solution of known pH and then the glass and reference electrodes are immersed in an unknown solution and pH directly red on the pH scale. Following procedure is followed to measure the pH.

- 168
- (i) In case the glass electrode has not been used for a long time, it should be immersed in water for 4-5 hrs before use. The glass electrode should not be allowed to dry.
- (ii) The pH meter has to be standardised. For this the glass electrode and calomel electrode are immersed in a buffer solution of known pH (see table 21.3) and the needle set at the pH of the buffer solution. The pH meter should be checked against a second buffer to ascertain that the pH meter has been standardised.
- (iii) The electrodes are washed with distilled water and rinsed the electrodes with the solution whose pH is to be determined.
- (*iv*) The electrodes are immersed into the solution whose pH is to be determined. The pH is read on the pH scale.

#### 21.4 DETERMINATION OF pH USING pH INDICATORS

A number of indicators are available, which change colour depending on the pH. Some of the indicators and their pH range are given in Table 21.4.

Indicator	pH range	Change corner
Acid cresol red	0.2-1.8	Red to yellow
Acid m-Cresol purple	1.2-2.8	Red to yellow
Acid Thymol blue	1.2-2.8	Red to yellow
Dimethyl amino azobenzene	2.0-4.0	Red to yellow
Bromophenyl blue	3.0-4.6	Yellow to blue
Bromocresol green	3.8-5.4	Yellow to blue
Methyl red	4.4-6.3	Red to yellow
Chlorophenol red	4.8-6.4	Yellow to red
Bromothymol blue	6.0-7.6	Yellow to blue
Phenol red	6.4-8.2	Yellow to red
Cresol red	7.0-8.8	Yellow to red
m-cresol purple	7.4-9.0	Yellow to purple
Thymol blue	8.0-9.6	Yellow to blue
Phenolphthalein	8.0-10.0	Colurless to red
Alizarin yellow	10.0-12.0	Yellow to lilac

Table 21.4 Indicators, their pH range and colour change

Most of the indicators are soluble in water or alcohol. Either their solutions are used as such or filter paper strips dipped in solutions and then dried in air are used.

A universal indicator, a mixture of several indicators given different colours over wide range of pH. A typical universal indicator in prepared by mixing 7.5 mL dimethyl aminoazobenzene, 10 mL bromothymol blue, 2.5 mL methyl red, 10 mL each of phenolphthalein and thymol phthalein. This indicator gives different colours at different pH as shown below.

рН	Colour	рН	Colour
2.0	Red-pink	7.0	Yellow-green
3.0	Red-orange	8.0	Green
4.0	Orange	9.0	Blue-green
5.0	Yellow orange		
6.0	Lemon yellow	10.0	Violet

Another universal indicator is prepared by dissolving 20 mg phenolphtholein, 400 mg methyl red, 600 mg methyl yellow, 800 mg bromothymol blue and 1g thymol blue in 96% alcohol and then adding 0.1 N NaOH solution until a pure yellow colour occurs. This indicator also given colour change at different pH values as shown below:

Colour	Red	Orange	Yellow	Green	Blue
рН	2.0	4.0	6.0	8.0	10.0

The pH indicator method gives an idea of approximate pH value.



- 1. Explain the term pH. Rain water has a pH 6.5 7.5. What inference you draw from this.
- 2. Write notes on:
  - (a) Hydrogen electrode
  - (b) Quinhydrone electrode
  - (c) Glass electrode.

What are their advantages and disadvantages?

- **3.** Which in your opinion is the best method for determing pH of a solution accurately.
- **4.** For determining pH using hydrogen electrode the pH is determined by the expression.

$$pH = \frac{E_{cell}}{0.0591}$$

How this relationship is derived?

#### 170 Instrumental Methods of Chemical Analysis

- **5.** What procedure is followed for determination of pH by using pH meter?
- **6.** Explain the uses of pH indicators.
- 7. The electromotive force of the following cell in 0.445V Pt, H<sub>2</sub>(1 atm)/H<sup>+</sup> (test solution)/KCl sat'd soln./Hg<sub>2</sub>Cl<sub>2</sub>/Hg Calculate the pH of the unknown solution.

(Given 
$$E_{\text{cell}} = 0.2415$$
)

**Ans.** 
$$pH = 3.44$$
.

**8.** Determine the pH of a solution placed in a hydroquinone cell which is coupled with standard calomel electrode. The e.m.f. of the combined cells as determined is 0.123 V at 25°.

**Ans.** 5.67.





# Chapter 22

### **Calorimetry**

#### 22.1 INTRODUCTION

Calorimetry is based on the measurement of the intensity of colour in order to find the concentration of a coloured solution. The intensity of the colour depends on the concentration of the species (which may be ions, molecules or a complex causing colour). The species to be determined may impart colour to the solution. Alternatively, the species may give a distinct colour on being complexed with a suitable reagent.

When light of an appropriate wavelength is passed through a coloured solution contained in a suitable cell, a fraction of the light is absorbed (depending on the concentration of the absorbing species and the thickness of the absorbing medium), and the rest of the light is transmitted. However, some light is reflected back from the solution, (its amount being negligibly small) but is eliminated by using a control. For all practical purposes, we may say

$$I_0 = I_a + I_t$$
  
 $I_0 = \text{Intensity of incident light}$   
 $I_a = \text{Intensity of light absorbed}$   
 $I_t = \text{Intensity of transmitted light.}$ 

The relationship between the intensity of incident rediation and that of the transmitted radiation is given by Lamberts law and Beer's law.

#### Lambert's Law

According to this law, when a beam of light passes through a solution/medium, equal fractions of incident light are absorbed by layers of equal thickness. It can be said that the differential decrease in the intensity with thickness of the absorbing medium is proportional to the intensity of the incident light. Mathematically,

$$-\frac{dl}{dI} = KI \qquad \dots(i)$$

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where, *K* is proportionality constant and *I* is thickness.

In rearranging equation (i), we get

$$-\frac{dl}{I} = K.dI \qquad ...(ii)$$

Integrating the above equation (ii), when I = 0 and  $I = I_0$ , we get

 $\log_e \frac{I}{I_0} = -KI$ or  $\log_{10} \frac{I}{I_0} = \frac{KI}{2.302}$   $\frac{K}{2.302} = K$ 

 $\log_{10} \frac{I}{I_0}$  is called 'absorbance' and K(K/2.303) is called absorption coefficient.

#### **Beers Law**

or

According to this law, the intensty of a beam of light decreases exponentially as the concentration of the medium decreases. It can be said that the differential decrease in the intensity of light as a function of concentration is directly proportional to the intensity of incident light.

$$-\frac{dI}{dc} = k'I \qquad \dots(iii)$$

Rearranging, we get,

$$-\frac{dI}{I} = kdc$$

on integration and putting the condition that when C = 0,  $I = I_0$ , we get

 $\log_e \frac{I}{I_0} = -kc$   $\log_{10} \frac{I_0}{I} = \frac{kc}{2.302} = kc$   $\log \frac{I_0}{I} = A, \text{ the absorbance}$ 

and k is absorption coefficient.

As can be seen K = k.

#### **Beer-Lambart Law**

The two laws (viz., Lambert law and Beers law) as explained above are combined to give in commonly known Beer-Lambart Law. According to this law the fraction of light absorbed by a given absorbing medium is directly proportional to the thickness of the medium and the concentration of the absorbing species. Solving in mathematical expression (i) and (iii) we get

$$A = \varepsilon C1$$
 ...(iv)

where l is thickness it till medium 1

c is concentration in mol dm<sup>-3</sup>,

ε is molar absorption coefficient.

ε the molar absorption coefficient is the absorbance of a solution having unit concentration, 1 M, placed in a cell of unit thickness, 1 cm. Absorbance is also called optical density (OD). On the basis of equation (*iv*), it can be said that the absorbance or OD of a solution in a container of fixed path is directly proportional to the concentration. A plot between absorbance and concentration is expected to be linear and a solution exhibiting such a behaviour is said to obey Beer-Lambert Law. Dilute solutions obey the law over a considerable concentration range. At higher concentration discrepancies occur; these are attributed to the changes occuring in coloured solutes which may undergo association at higher concentration. It is best to prepane calibration curve using a series of standards of known concentration.

For colorimetic determination, a number of instruments are availale.

#### 22.2 PRINCIPLE OF CALORIMETER

A calorimeter consists of a light source, a cell/cuvette for holding the solution, a pholoelectric cell to capture the radiation transmitted by the solution and a measuring device to detect the response of a photocell. A schematic representation of a colorimeter is given in Fig. 22.1.



Fig. 22.1 Schematic representation of calorimeter

A calorimeter has three light emitting diodes (LEDs), which emit light of different colours. The light to be used depends on the colour of the absorbing medium. The light from the source passes through a slit in order to get a thin ray, which falls on the cell cointaining the solution. As already stated, some of the light is absorbed and the rest transmitted. The transmitted light falls on the photo cell where a current is generated, whose magnitude is proportional to the intensity of the light falling on it. The amplified current signal is measured with the help of an ammeter. The deflection on the meter is directly proportional to the intensity of the light. The intensity of the incident light is determined by putting distilled water in the cuvette; in this case no light is absorbed and the whole of it falls on the photocell. If the solution is made in a solvent other than water, the reference sample is taken as the pure solvent. The difference of the two reading gives the amount of light absorbed.

## 22.3 PROCEDURE FOR THE ESTIMATION OF Cu<sup>2+</sup> IN A UNKNOWN SOLUTION

Following steps are followed:

#### 1. Calibration of the Calorimeter

A typical calorimeter is shown in Fig. 22.2. It has the following controls.

**Power Switch:** This is an SPST toggle switch and is located on the back panel of the calorimeter. It turns the instrument OFF/ON. When ON, the LED on the front panel glows on plugging to a 220 V supply.

**Colour Selector:** This is a 2 pole 3 way rotary selector switch. It is used to switch on a particular LED in the cell holder and also to bring into the amplifier circuit a particular parallel register. It has three positions. *R*, *Y*, and *G* relating to red, yellow and green colours.

**Set Zero:** By this knob the meter is set to zero when the reference solution in put in the cell holder.

**Sensitivity:** This knob adjusts the sensitivity of the meter.

**Six Pin Plug:** It is located at the right side of the back panel and is inserted into the six pin socket in the cell holder (Wires from the LEDs and the LDR are connected to the socket).

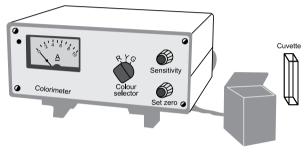


Fig. 22.2 Calorimeter

For the calibration of the calorimeter, the clean, dry cuvette in filled with distilled water or the reference solution. The cuvette has two plain sides and two striated sides. One of the plane side of the cuvette is marked with a pen and inserted in the cell holder with the marked side facing LEDs inside the holder (always insent in the same way). The cell holder is closed with the lid and meter reading adjusted to zero by using the set zero knob.

As a next step, the cuvette is removal, reference solution poured off, cuvette is rinsed and dried. Now the cuvette is filled with copper sulphate stock solution (8%  ${\rm CuSO_4}$  solution), the cuvette is inserted in the cell holder in the same orientatin as is done earlier) and the lid of the cell closed.

The colour selector knob is set on R (A  $CuSO_4$  solution has an absorption maximum in the red region). Using the sensitivity knob, the meter reading is adjusted to near to end of the scale (say 0.9).

The above two steps are repeated for settings the zero with distilled water or the reference solution.

In order to ascertain, whether the calorimeter has been properly caliberated follow the following steps:

- (i) To ten clean dry test tubes is added 10 mL, 9 mL, 8mL, 7 mL, 6 mL, 5 mL, 4 mL, 3 mL, 2 mL and 1 mL of copper sulphate stock solution (8% prepared earlier) in these test tubes respectively. Each solution is diluted with distilled water to make the total volume 10 mL in each case.
- (ii) Using the same cuvette (as for calibration) measure the meter readings in each of the above ten solutions. The cuvette should be properly rinsed before pouring the solution. It should be ascertained that the set zero and sensitivity knobs are not disturbed throughout the measurement.

(iii) The meter readings are plotted against the volume of the stock solution taken in each of the test tubes. A linear graph is expected since CuSO<sub>4</sub> solution is known to obey Beer-Lambert law in this concentration range. The linear graph shows that the calorimeter has been properly calibrated.

## 2. Preparation of Copper Nitrate Solution of Varying Concentration

For this purpose prepare 10% solution  $\text{Cu(NO}_3)_2$ .  $3\text{H}_2\text{O}$  and dilute it as shown below:

<b>Table 22.1</b>	Copper ni	trate soluti	on of var	rying co	oncentration.	

SI. No.	Volume of Cu(NO <sub>3</sub> ) <sub>2</sub> .3H <sub>2</sub> O stock solution	Volume of distilled water	%Cu(NO <sub>3</sub> ) <sub>2</sub> .3H <sub>2</sub> O
1	0	10	0
2	2	8	2
3	4	6	4
4	6	4	6
5	8	2	8
6	10	0	10

Thus, we will get six solutions where the concentration of  $\text{Cu(NO}_3)_2$ .  $3\text{H}_2\text{O}$  varies from 0–10% as given in table.

# 3. Plating a Calibration Curve

As a first step the cuvette is cleaned dried and filled with solution number 1 after rinsing it with the same solution. The cuvette is placed in the cuvette holder in the instrument and meter response recorded. In a similar way response from other solution is recorded by repeating the above procedure. The observations is recorded as shown below:

#### Observation

**Table 22.2** Meter response as a function of concentration of copper nitrate

SI.No.	Strength of Cu(NO <sub>3</sub> ) <sub>2</sub> .3H <sub>2</sub> O in % m/v	Meter Response
1	0	
2	2	
3	4	
4	6	
5	8	
6	10	
Unknown		

For the sake of convenience, a sample reading, Table II and a calibration curve is produced from the readings obtained (Fig. 22.3).

Table 22.3 Meter response as a function of concentration of copper sulphate

SI.No.	Strength of CuSO <sub>4</sub> 5H <sub>2</sub> O in % m/v	Meter response
1	2	1.8
2	4	3.9
3	6	6.10
4	8	8.05

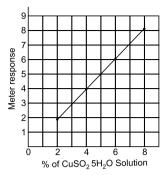


Fig. 22.3 Sample calibration curve drawn from the readings given in Table 22.3

# 4. Determination of the amount of Cu<sup>2+</sup> in unknown solution

Preparation of Cu<sup>++</sup> solution.

Cupper wire (properly cleaned by washing with dilute H<sub>2</sub>SO<sub>4</sub>, washing well with water and drying, amount used 1.7 g) is treated with 5-10 mL of 6 M HNO<sub>3</sub> in a conical flask. In case reaction is slow, few drops of conc. HNO<sub>3</sub> are added. If necessary copper wire is dissolving by warming in a hot water bath. After all the copper wire has dissolved, the solution diluted with about 50 mL water and boiled for about 10 min to removal oxides of nitrogen. Urea (1 g) is added and the solution boiled (5 min). The cooled solution is neutralised with ammonia solution (1:3) until a faint permanent precipitate of Cu(OH)<sub>2</sub> appears.

In case, the solution acquire deep blue colour on addition of ammonia, the solution is boiled. To the cooled solution is added dropisewise glacial acetic acid until the precipitate dissolves and a clear solution is obtained. The solution is made up to one litre in a volumetric flask with distilled water. The homogeneous solution thus obtained is used to determine the strength of  $Cu^{2+}$ .

Finally, the concentration of the unknown solution (as prepared above) is determined with the help of the calibrated curve (Fig. 22.3).

From the calibration curve the percentage of copper nitrate solution is X% = ...%.

$$\frac{X \times 63.5}{187.5} = Zg \text{ of Cu/100 mL}.$$

Mass of copper wire taken = 1.7g/100 mL

$$\therefore \qquad \% \text{Cu} = \frac{Z}{1.7} \times 100\% \,.$$
**EXERCISES**

- 1. Discuss the principle of calorimetry.
- 2. Write a note a Beer-Lambert Law.
- 3. How will you estimate the amount of  $Cu^{2+}$  in a unknown solution.
- **4.** Give examples of other ions or compounds which can be estimated calorimetry.

# Part-V

# **Instrumental Methods for Structure Determination of Organic Compounds**

- Infrared Spectroscopy
- Ultraviolet Spectroscopy
- Nuclear Magnetic Resonance Spectroscopy
- Electron Spin Resonance Spectroscopy
- Mass Spectrometry
- Polarimetry
- Optical Rotatory Dispersion and Circular Dichroism



# Chapter 23

# **Infrared Spectroscopy**

#### 23.1 INTRODUCTION

The infrared radiation refers to the part of electromagnetic spectrum between the visible and the microwave regions. An organic chemist is mostly interested between 4000 cm $^{-1}$  and 666 cm $^{-1}$  (2.5 - 15.0  $\mu$ ). The shorter wavelength region (12500 - 4000 cm $^{-1}$ ) is referred to as near infrared region and the region extending from 666 cm $^{-1}$  to 50 cm $^{-1}$  is referred to as far infrared region. The electromagnetic spectrum is shown in Fig. 23.1.

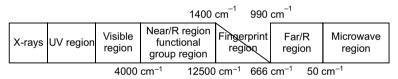


Fig. 23.1 The electromagnetic spectrum

Infrared spectroscopy (IR) provides a simple and rapid instrumental technique for the identification of functional groups in organic compound. It is equally important in providing information regarding functional groups which are absent.

The basis of IR spectroscopy is the absorption of infrared radiations. So, it is also called absorption spectroscopy. The extent of the absorption depends on the wavelength/frequency of the IR radiation passing through the molecule. The absorption of radiation leads to gain in energy of the molecule, which in turn is responsible for a variety of vibrations in the molecule (this aspect will be discussed subsequently). If a graph is plotted showing the variation of absorption (or transmission) of radiation by a compound with wavelength or frequency, we get the absorption curve known as **infrared spectrum** of the compound. In the IR

spectrum, the intensity of absorption (or transmission) is usually expressed along the ordinate while the wavelength or frequency is denoted along the abscissa. A typical IR spectra is shown in Fig. 23.2.

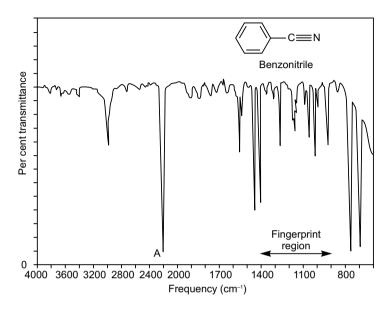


Fig. 23.2 IR spectrum of benzonitrile A 2210 cm<sup>-1</sup> (C = N stretch)

As seen (Fig. 23.2), in the fingerprint region (900 – 1400 cm<sup>-1</sup>), there are large number of peaks; this region has an extremely complex spectrum. Advantage is taken of this region and the spectrum of an unknown compound is matched against that of an authentic sample. A peak-by-peak correlation is excellent evidence for its identity. It is most unlikely that any two compounds (except the enantiomers) give the same IR spectrum.

Band positions in infrared spectra are represented either as wave numbers or wavelengths. The wave number unit (cm<sup>-1</sup>, reciprocal centimeters) is more often used, since it is directly proportional to the energy of vibration and also because most of the latest modern instruments are linear in the cm<sup>-1</sup> scale.

Wave number is inversely proportional to wavelength

$$\overline{v} = \frac{1}{\lambda}$$

which  $\overline{v}$  is the wave number (or frequency of radition) and  $\lambda$  is the wavelengh.

Only those vibrations that result in rhythmical change in the dipole moment of the molecules are observed in the infrared. For example, a molecule like  $\rm H_2$  does not display a hydrogen-hydrogen stretching vibration because the dipole moment does not change as the hydrogen-hydrogen bond is elongated or compressed.

It has been found that the frequency of vibration is inversely related to the masses of the atoms bonded to one another. Thus, heavier the atoms are, the lower is the frequency of vibration. Thus, C = O exhibits absorption at 1700 cm<sup>-1</sup> compared to C = S, which exhibits absorption at 1350 cm<sup>-1</sup>. Similarly the absorption due to C—H is 3000 cm<sup>-1</sup> compared to 2200 cm<sup>-1</sup> in case of C—D.

Also, the frequency of vibration is directly proportional to the strength of the bond. Thus, the stretching vibration of a triple bond (C = C) appear at a higher frequency (2150 cm<sup>-1</sup>) than that of either a double bond (C = C) (1650 cm<sup>-1</sup>) or single bond (C - C) (1200 cm<sup>-1</sup>).

# 23.2 BASIC THEORY

A molecule is known to contain various energy levels. These include electronic, vibrational and rotational energy levels. When a molecule absorbs radiation its energy increases in proportion to the energy of the photon and is given by the relation

$$E = hv = \frac{E_C}{\lambda}$$

where h is planck's constant, v is the frequency of radiation,  $\lambda$  is the wavelength and c is the velocity of light. Photon is a small unit of energy.

The increase is energy may be electronic, vibrational or rotational energy of the molecule. The energy required changes in the order:

electronic energy > vibrational energy > rotational energy

Since only small amounts of energy are associated with infrared radiation, they do not result in electronic excitations but result in transitions between vibrational and rotational energy levels of a molecule. In fact, the infrared spectrum of an organic molecule is due to transitions between vibrational and rotational energy levels.

#### 23.3 INSTRUMENTATION

The infrared spectrometers are of two types: Single beam and double beam. In a single beam infrared spectrometer, the radiations (from a source) pass through a cell containing the sample and then through a prism, which disperse the light. A main disadvantage associated with such instruments is that the bands due to solvent are also obtained. This makes the interpretation of the bands and identification of the compounds difficult. These problems have been overcome be using **double beam spectrometers**.

In double beam spectrometers, the light (from a source) is split into two beams, one passing through the sample and the other through the solvent for compensation. The two beams are subsequently combined.

Subsequents steps are same as in the case of single beam instrument. A schematic diagram (Fig. 23.3) of a double-beam infrared spectrometer and its essential features are given below:

(i) Radiation Source: Radiations are obtained from a ceramic rod, which is electrically heated to 1100 – 1800°C. This rod is made of silicon carbide or a Nernst filament (which is made of compressed mixture of sintered oxides of zirconium, thorium and cerium (using a binding material). The radiation from the source is divided into two beams, one passing through the sample, while the other functions as a reference beam. Both the reference and sample beams are–alternately passed into a monochromator at very short intervals using a rotating mirror (Fig. 23.3).

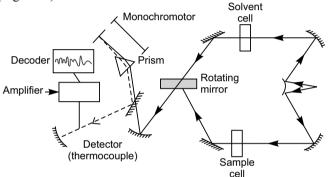


Fig. 23.3 Schematic diagram of a double beam infrared spectrometer

- (ii) **Absorption Cells:** The cells are made of rock salt or potassium bromide. Glass or quartz cells cannot be used since these absorb IR radiation.
  - The organic compound 0.5g is dissolved in a suitable solvent like CCl<sub>4</sub>, CHCl<sub>3</sub>, CS<sub>2</sub> etc. For more details *see* section 23.4.
- (iii) **Monochromator:** If consists of prism and grating. The monochromator separates source radiation wavelengths. The prism is made of sodium chloride (which is transparent to IR radiation in the range 4000 650 cm<sup>-1</sup>), lithium fluoride (better than sodium chloride but is not transparent below about 1000 cm<sup>-1</sup>), potassium bromide (transparent up to 400 cm<sup>-1</sup>) or cesium iodide (Transparent up to 200 cm<sup>-1</sup>).
  - The high resolution instruments use filters to reject unwanted radiations.
- (*iv*) **Detector:** The detector is of thermocouple type and changes the thermal radiant energy into electric energy. From detector the radiation is passed on to amplifier and recorder.

# 23.4 FOURIER TRANSFORM INFRARED (FTIR) SPECTROMETER

The conventional IR instruments have slow response. High speed IR instruments

are necessary for on line identification of constituents of a flowing system. In FTIR instruments, the IR spectras are rapidly and accurately recorded in a few seconds using the principle of interferometry (section 23.4(A)). FTIR spectroscopy makes use of the all frequencies from an IR source simultaneously. Since all wavelengths of IR spectra are simultaneously measured, the method is faster than conventional IR spectrometers. In this full IR energy is available and so even trace amount of the sample can be analysed. In fact FTIR provides complete spectral range without using movable parts like slits, grating, chopper etc. (which are usal in conventional IR spectrometers.

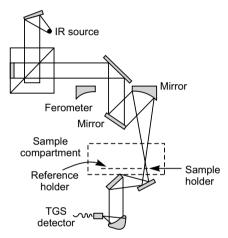


Fig. 23.3(a) Flow shut of an FTIR spectrometer

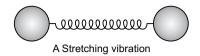
FTIR spectroscopy has been used for the identification and characterisation of both organic and inorganic compound. A typitcal diagram of a FTIR instrument in given in the Fig. [23.3(*a*)].

# 23.4(A) PRINCIPLE OF INTERFEROMETRY

Infrared radiation consists of all wavelengths (*e.g.*, 5000 – 400 cm<sup>-1</sup>). It is split into two beams, which subsequently recombine after the introduction of a path difference. One of the beams is of fixed length and the other is of variant path length. When the difference of the corresponding wavelength is an integral multiple of the invariant beam, the interference is constructive. However, when the difference is an odd integer multiple of the one quarter of the wavelength the interference is distructive. Thus, the result of a complete variation of wavelength in an oscillary series of destructive and constructive combinations. This is referred to as interferrogram.

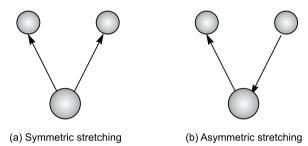
#### 23.5 MODE OF VIBRATIONS

The molecules can vibrate in a number of ways. Two atoms joined by a covalent bond can undergo a stretching vibration where the atoms move back and forth as if joined by a spring.



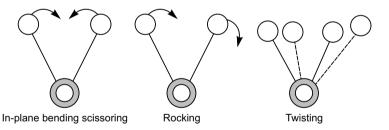
Three atoms can also undergo a variety of stretching and bending vibrations.

(i) Stretching: This may be symmetric or asymmetric.



For example, the two CO bonds present in the molecule of carbon dioxide can be shown in (a) and (b) type of stretch. The symmetric stretching, in the case of  $CO_2$  does not result in a change in the dipole moment and hence is IR inactive. The asymmetric stretch on the other band is IR active.

(ii) Bending: This may be in plane or out of plane bending. Scissoring, rocking, twsiting, etc., are some of the important types of bending vibrations.



# 23.5(A) NUMBER OF FUNDAMENTAL VIBRATIONS, SELECTION RULES

A molecule having n atoms is known to have 3n degrees of freedom. Three of these degrees of freedom are assigned each to rotational and translational (two in the case of linear molecules like carbon dioxide or acetylene). The remaining degrees of freedom (3n - 6) (3n - 5) is case of linear molecules) to vibrational motions. Thus, it can be said that number of vibrational degrees of freedom for non-linear polyatomic molecule is 3n - 6 and for a linear molecule it is 3n - 5. Thus, as an

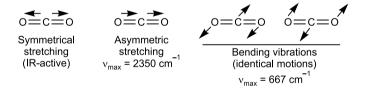
example, non-linear molecule like methane should theoretically have  $(3 \times 5-6) = 9$  degrees of freedom and benzene should theoretically have  $(3 \times 12-6) = 30$  degrees of freedom. However, a linear molecule like  $CO_2$  should theoretically have  $(3 \times 3-5) = 4$  fundamental absorptions.

It has already been stated that IR light is absorbed only when a change in dipole character of the molecule takes place. In practice it is found that the total number of absorption will be less than the 3n–6 or 3n–5 rule since there are certain structures which are symmetrical and no IR absorption takes place. Thus, some of the fundamental vibration are infrared active and others are infrared inactive. This is decided on the basis of the selection rules, which are as follows:

- In molecules having a centre of symmetry, some of the vibration which are symmetrical about the centre of symmetry are infrared inactive (such vibrations are active is Raman spectra).
- In molecules without a centre of symmetry, the vibrations are infrared active (such vibration are inactive in Raman spectra).

It is thus found that the IR spectra and Raman spectra are complementary. However, the IR is most informative for organic chemists since most of the functional groups do not have centre of symmetry.

As an illustration CO<sub>2</sub> does not have a centre of symmetry and can have the following four fundamental vibrations:



Thus,  $CO_2$  shows two bands at 2350 and 667 cm<sup>-1</sup> corresponding to asymmetric stretching and bending vibrations.

#### 23.6 RECORDING OF IR SPECTRA

Before recording an IR spectra, the following should be kept in mind:

- The compound whose spectra is to be recorded must be reasonably pure.
- The IR spectra must be adequately resolved and of adequate intensity.
- The IR spectrophotometer should be calibrated so that the bands are observed at proper frequencies or wavelengths. The calibration is normally done by using polystyrene film.

2.5 3.5 4.5 10 12 14 100 80 Fransmittance, 60 40 0 -3400 3000 2600 2200 2000 1800 1600 1400 1200 1000 800 4000 600

The IR spectra of polystyrene (thin film) is shown below:

IR spectra of polystyrene (thin film)

— The procedure of handling of sample must be specified. In case, a solvent is used its concentration and cell thickness must be indicated.

Wave numbers, cm<sup>-1</sup>

- Finally, the IR spectra may be recorded is one of the following ways:
  - (i) **KBr Pellet:** The solid compound (50 mg) is finely powdered with spectral grade potassium bromide (0.5 g). It is then covnerted into a pellet by applying high pressure to the mixture. The pellet is placed in the spectrophotometer and IR recorded.

Though this method is generally used for solids, special precaution should be taken to use dry potassium bromide and to exclude moisture during pellet formation. The presence of moisture interferes in the detection of functional groups like O—H, N—H, S—H, etc.

(ii) Solution Method: The dry solid compound is dissolved in anhydrous CCl<sub>4</sub>, CHCl<sub>3</sub> or CS<sub>2</sub> (20% solution) and the solution is placed in the sample cell. Pure solvent is taken in the solvent cell and the IR is recorded on a double beam spectrophotometer, which automatically cancels the absorption due to solvent. Small absorption peaks are always observed because of the unequal concentration of solvent in the two cells. These peaks can, however, be ignored.

**Note:**  $CS_2$  absorbs at 1500 cm<sup>-1</sup> and absorption due to C—C1 occurs between 600 and 800 cm<sup>-1</sup>.

(iii) Mull Method: A dispersion of the finely powdered organic compound in Nujol (a mixture of saturated hydrocarbons containing 20-24 carbon atoms) or perfluorokerosene (fluorinated kerosene containing C<sub>12</sub>—C<sub>18</sub> linear hydrocarbons) can be used to record the spectrum. (iv) NaCl Windows: The IR spectrum of pure (neat) liquid compound can be recorded using NaCl windows. A small amount of the liquid is placed on a moisture-free NaCl plate (windows); it is covered with another NaCl plate and the spectrum recorded. If the compound is a low boiling liquid, some of it tends to evaporate during the time taken to record the spectrum and a uniform concentration of the compound is not maintained throughout. Also a uniform thickness of the sample is not possible, if the spectra of two compounds is to be compared.

Some of the spectrophotometers in use have a fixed distance between NaCl plates and the evaporation of the liquid is prevented by using teflon covers.

(v) IR of a Gaseous Compound: When the IR spectrum of a gaseous organic compound is to be determined, the path length of the cell containing the sample is increased (10-20 cm) so as to have a high concentration of the substance for obtaining a good IR spectrum.

It is found that in a IR spectrum, the frequency of a given stretching vibration can be related to two factors. These are masses of the bonded atoms (light atoms vibrate at higher frequencies than the heavier ones) and the relative stiffness of the bond (for example, triple bonds being stiffer vibrate at higher frequencies than double bonds, and double bonds vibrate at higher frequencies than single bonds). The absorption range of various multiple bonds are shown below:

Bond	Frequency range (cm <sup>-1</sup> )
$C \equiv C$	2100–2260
$C \equiv N$	2220–2260
C = C	1620–1680
C = O	1630–1788

It is also found that the stretching frequencies of groups involving hydrogen (a light atom) such as C–H, N–H and O–H all occur at relatively high frequencies.

Group	Bond	Frequency Range (cm <sup>-1</sup> )
Alkyl	С—Н	2853–2962
Alcohol	О—Н	3590–3650
Amines	N—H	3300–3500

The IR spectra of even simple molecules may show absorption peaks. All molecules exhibit molecular vibrations by the absorption of IR energy. For a

vibration to occur with the absorption of IR energy, the dipole moment of the molecule must change as the vibration occur. Thus, in case of methane, when the four hydrogen atoms vibrate symmetrically, the molecule does not absorb IR energy. Symmetrical vibrations of the carbon-carbon double bond and triple bonds of ethene and ethyne do not result in the absorption of IR energy either.

The region of interest to an organic chemist  $2.5-15~\mu$  or  $4000-650~cm^{-1}$ . Broadly speaking, this region may be divided into the functional group region  $(2.5-7.0~\mu)$ , and the fingerprint region. The fingerprint region like the human finger is a characteristic of each molecule. Careful analysis of the IR spectrum in the functional group region provides a great deal of information concerning the presence or absence of different functional groups. For example, the C=O group absorbs in the region of  $1680-1800~cm^{-1}$ . Presence of a strong peak in this region indicates the presence of a C=O group and the absence of a strong absorption peak in this region indicates the absence of a C=O group and hence the compound cannot be a ketone, aldehyde, carboxylic acid, amide, ester, anhydride, or acid halide, etc. An aldehyde can be easily distinguished from a ketone because of the presence of two bands at 2760 and  $2850~cm^{-1}$ , which are due to the C—H stretch of the —CHO group and are absent in the spectrum of a ketone.

The absence of strong absorption in the 909-650 cm $^{-1}$  (11.0–15.4  $\mu$ ) region normally indicates a non-aromatic structure. Similarly, the presence of a broad absorption peak between 3200 and 3600 cm $^{-1}$  indicates the presence of O–H, N–H, etc.

Following three, features of an IR spectrum give information for the identification of structural elements:

- Position of the bands (in wave numbers)
- The intensities of the bands (weak, medium, strong)
- Shapes of the bands (sharp or broad).

Different kinds of bond vibrate at different frequencies. Thus, IR spectrascopy is used to know about different kinds of bonds and is possible to determine the presence of functional groups.

The frequency of bond vibration is derived from Hooke's law, which shows that the frequency of vibration depends on the atom mass and bond strength. Hooke's law is:

$$\overline{v} = K\sqrt{\frac{f}{m}}$$

where f is the force constant, m is the mass and k is a constant.

The force constant (f) is the strength of the bond. The greater the value of f, the stronger is the bond and higher is the  $\vec{v}$  of vibration. The mass (m) is the mass of the atoms. The smaller is the value of m, the higher is the  $\vec{v}$  of vibration.

# 23.7 MAJOR BANDS IN THE IR SPECTRA OF DIFFERENT TYPES OF ORGANIC COMPOUNDS

### **Hvdrocarbons**

Hydrocarbons in general have C—H bonds. Absorptions for bonds to H occurs on the left side of the spectrum (the high weve number region). All hydrocarbons show carbon-hydrogen stretching vibrations in the region 2800-3300 cm<sup>-1</sup>. These absorptions can be used in interpreting IR spectra, since the exact location of the peak depends on the strength (and stiffness) of the C—H bond; this in turn depends on the hybridization state of the carbon that bears the hydrogen. Thus, C—H bonds involving sp-hybridized carbon are strongest and those involving sp<sup>3</sup>-hybridized carbon are the weakest. The bond strength is of the order

$$sp > sp^2 > sp^3$$

This is also the order of bond stiffness.

The carbon-hydrogen stretching peaks of hydrogen atoms attahced to sp-hybridized carbon atoms occur at highest frequencies (about 3300 cm<sup>-1</sup>). Thus,  $\equiv$  C–H groups of terminal alkyne give peaks in the region.

The carbon-hydrogen stretching peaks of hydrogen atoms attached to  $sp^2$ hybridized carbon atoms occur in the region 3000-3100 cm<sup>-1</sup> region. Thus, the alkenyl C—H bond and the C—H bond of aromatic ring give absorptions in this region.

The carbon-hydrogen stretching peaks of hydrogen atoms attached to  $sp^3$ hybridized carbon atom occurs at lowest frequencies in the region 2800-3000 cm<sup>-1</sup>.

The carbon-hydrogen stretching bonds of various types can be summarized as:

C ( <i>sp</i> )–H	3300 cm <sup>-1</sup>
C ( <i>sp</i> <sup>2</sup> )–H	3000-3100 cm <sup>-1</sup>
C ( <i>sp</i> <sup>3</sup> )–H	2800-3000 cm <sup>-1</sup>

It should be noted that higher is the per cent s-character, the stronger the bond and higher the frequency of absorption.

(increasing frequency of absorption)

Besides the C–H stretching band, the hydrocarbons also give absorption peaks resulting from carbon-carbon bond stretching. We know that bond strength decreases in the order C = C < C = C < C. So the frequency of vibration decreases, *i.e.*, the absorptions for these bonds move further to the right side of the spectrum. The carbon-carbon single bonds usually give very weak absorptions, which are of little use in the assignment of structures. Carbon-carbon double bonds give absorptions peaks in the region of 1620-1680 cm<sup>-1</sup> and carbon-carbon triple bonds give absorptions peaks in the region of 2100-2260 cm<sup>-1</sup>. However, these absorptions are not strong and they are absent if the double bond or triple bond is symmetrically substituted (in such cases, no dipole moment change will be associated with the vibration). The carbon-carbon bonds of benzene rings generally give a set of characteristic sharp peaks in the region of 1450-1600 cm<sup>-1</sup>.

The carbon-carbon bond stretching in hydrocarbons can be summarized as given below:

C — C Very weak peak (not used in assignment of structures)

C = C 1620–1680 cm<sup>-1</sup> C = C 2100–2260 cm<sup>-1</sup>

C — C bonds 1450–1600 cm<sup>-1</sup> (sharp peaks) of benzene.

In case of alkenes, the absorptions arising from carbon-hydrogen bending vibrations occur in the region 600–1000 cm<sup>-1</sup>; the exact locations of these peaks is used for determination of the substitution pattern of the double bond and its configuration.

Monosubstituted alkenes, RCH =  $CH_2$ , give two strong peaks in the regions 905–920 and 985–1000 cm<sup>-1</sup>.

Disubstituted alkenes,  $R_2C = CH_2$ , give strong peak in the region of 880-900 cm<sup>-1</sup>.

cis-Alkenes give an absorption peak in the region of 675-730 cm<sup>-1</sup>.

trans-Alkenes given a peak in the region of 960-975 cm<sup>-1</sup>.

The absorption peak range given above for *cis*- and *trans*- alkenes are dependable for alkenes that do not have electron-releasing or electron-withdrawing substituent (other than an alkyl group) on one of the carbon atoms of the double bond. In case, electron-releasing or electron withdrawing substituents are present on a double-bonded carbon atoms, the bending absorption peaks may be shifted outside the regions given above.

It should, however, be noted that symmetrically *trans*-disubstituted and tetrasubstituted double bonds and internal alkynes do not show any absorption corresponding to C = C or  $C \equiv C$  stretching mode respectively.

As an example, ethylene  $(H_2C=CH_2)$  does not absorb infrared radiation. This is because there is no change in dipole moment on subjecting to IR radiation. So the C=C bond in ethylene does not show IR absorption.

Also, the symmetrical molecule like 2-butyne ( $CH_3 - C = C - CH_3$ ) does not exhibit IR absorption around 2250 cm<sup>-1</sup>. This is because the C = C is non-polar and there is no change in the dipole moment. This type of vibration is called IR inactive.

# Cycloalkanes (Cycloparaffins)

In unstrained cycloalkanes, the C—H stretching frequencies are the same as those for acyclic alkanes. However, increasing ring strain moves C–H stretching bands to higher frequencies.

The C-C stretching vibrations in cycloalkanes appear as weak bands in the region 1200-800 cm<sup>-1</sup> and are of little value for structure elucidation.

# **Aromatic Hydrocarbons**

These exhibit following absorption bands:

3100–3000 cm<sup>-1</sup> due to aromatic C—H stretching vibrations.

 $2000{-}1667~{\rm cm^{-1}}$  due to overtones and combination bands of C—H out of plane bending vibrations.

There are six equivalent C······C bonds in benzene and so there will be six C······C stretching vibrations. The exact position of the bands depends on the substitution pattern (see Table 23.1).

Following examples of hydrocarbons make the understanding of IR absorptions clear.

Alkane (*n*-hexane),  $CH_3CH_2CH_2CH_2CH_3$ . Its IR spectra shows  $C_{sp^3} - H$  absorption in the region  $3000 - 2850 \text{ cm}^{-1}$ .

Alkene (1-hexene),  $CH_2 = CH \ CH_2CH_2CH_2CH_3$ . Its IR spectra shows the following absorptions:

3150–3000 cm<sup>-1</sup> (m) due to 
$$C_{sp}^2$$
 – H  
3000–2800 cm<sup>-1</sup> (s) due to  $C_{sp}^3$  – H  
1650 cm<sup>-1</sup> (m) due to C = C

Alkyne (1-Hexyne),  $HC = CCH_2CH_2CH_2CH_3$ . Its IR spectra shows the following absorptions:

3300 cm<sup>-1</sup> (m) due to 
$$C_{sp} - H$$
  
3000–2850 cm<sup>-1</sup> (s) due to  $C_{sp}^3 - H$   
2250 cm<sup>-1</sup> (m) due to  $C \equiv C$ .

## **Functional Groups**

The functional groups are generally identified by looking for absorption bands in the following four regions:

Region	Frequency Range (cm <sup>-1</sup> )	Type of Bonds	Functional Groups
1	3500–3200	0—Н	Alcohol, Phenol
		N—H	Amine, Amide
2	2300–2100	C≡C	Alkyne
		C≡N	Nitrile
3	1800–1650	C = O	Aldehyde, Amide, Anhydride (2 bands), Carboxylic acid, Acid chloride and Ester
4	1650–1500	C = C	Alkene
		C = C	Arene
		C = N	Imine
		N = O	Nitro compound

Following are given some of the functional groups whose presence may be inferred on the basis of IR absorption.

## **Carbonyl Groups**

Carbonyl group gives a prominent absorption peak in the IR spectra. This group is present in aldehydes, ketones, esters, carboxylic acids, amides, etc. The carbonoxygen double bond stretching frequency in all these groups give a strong peak in the region of 1630–1789 cm<sup>-1</sup>. The exact absorption depends on the nature of the compound as given below:

The stretching frequencies of C = O group in cycloalkanones increase with decrease in ring size as shown below:

The aldehydes, besides showing carbonyl absorption show characteristic C–H stretching bands at about 2740 and 2855 cm<sup>-1</sup>.

[For more details see Hint given for Question No. 20].

The following compounds (I and II) have carbonyl groups.

Predict their IR absorptions.

**Ans.** Both I and II show the peaks in the region 1680-1750 cm $^{-1}$  due to the presence of carbonyl group and another peak in the region 3000–2800 cm due to  $C_{\rm sp}^3$  – H.

Since both I and II are different components, the absorptions in the fingerprint region will be quite different.

## **Hydroxyl Group**

IR spectra is very helpful to recognize the hydroxyl groups of alcohols and phenols. In case an alcohol or a phenol is present as a very dilute solution in carbon tetrachloride, O—H absorption occurs as a very sharp peak in the region of 3590-3650 cm<sup>-1</sup>. In very dilute solution or in the gas phase, formation of intermolecular hydrogen bonds does not take place because the molecules are too widely separated. The sharp peak in the region of 3590-2650 cm<sup>-1</sup> is attributed to 'free' (unassociated) hydroxyl group. Increasing the concentration of the alcohol or phenol causes the sharp peak to the replaced by a broad band in the region of 3200-3550 cm<sup>-1</sup>. This absorption is attributed to OH groups that are associated *via* intermolecular hydrogen bonding.

# **Carboxylic Acids**

Carboxylic acids normally exist as dimers in the solid state and as monomers in dilute solution.

In the monomeric form, carbonyl stretching band appears at about 1750 cm<sup>-1</sup>, whereas the dimeric form, carboxylic acids show the stretching band at about 1700 cm<sup>-1</sup>.

As the dimeric form of carboxylic acids has strong hydrogen bonds, the O—H stretching band shift to lower frequency at about 2700–2500 cm<sup>-1</sup>. However, in the monomeric form, the O—H stretching band appears around 3500 cm<sup>-1</sup>.

In addition to the above, carboxylic acid shows coupled C—O and O—H stretching bands at 1420 and 1300 cm<sup>-1</sup>.

The presence of carboxyl group can be confirmed by conversion to the acid salt (in situ in CHCl<sub>3</sub> solution by the addition of Et<sub>3</sub>N). In the spectra of the resulting carboxylate anion, the IR absorptions due to carboxyl group disappear and those of -COO<sup>-</sup> anion appear. These are discussed below:

## **Carboxylate Anion**

The carboxylate anion is represented as:

$$-c$$
  $\leftarrow$   $-c$ 

The two equivalent C·····O bonds of COO<sup>-</sup> exhibit a band at  $1650-1550 \text{ cm}^{-1}(s)$  and a band at  $1400 \text{ cm}^{-1}(w)$ .

#### **Esters**

In addition to carbonyl absorption in the region of 1735-1750 cm<sup>-1</sup> (*see* carboxyl group), the esters have a strong C—O stretching band at 1280-1000 cm<sup>-1</sup>.

#### **Amines**

Primary and secondary amines in dilute solution give sharp peaks in the region 3300-3500 cm<sup>-1</sup> due to free N—H stretching vibrations. Primary amines give two sharp peaks, secondary amines give only one. Tertiary amines do not absorb in this region, since they do not have any N—H bond.

RNH <sub>2</sub>	R₂NH
1 Amine	2 Amine
Two peaks in the region	One peak in the region
3300-3500 cm <sup>-1</sup>	3300-3500 cm <sup>-1</sup>

Hydrogen bonding causes N—H stretching peaks of 1 and 2 amines to broaden.

Besides N—H stretching vibrations, the amines also exhibit C—N stretching vibrations.  $\mathring{1}$  and  $\mathring{2}$  aliphatic amines exhibit C—N stretching vibrations in the region 1250-1020 cm $^{-1}$ , the exact position of the absorption depends on the structure of the amines. Thus, aliphatic  $\mathring{1}$  and  $\mathring{2}$  amines with primary cabrons (—CH $_2$  — NH $_2$ , — CH $_2$  — NH — CH $_2$ —) exihibit bands near 1090 cm $^{-1}$  and 1140 cm $^{-1}$  due to asymmetric C—C—N and C—N—C stretching vibrations respectively. On the

other hand,  $\mathring{1}$  amines with secondary carbon atoms  $\begin{pmatrix} C \\ C \end{pmatrix}$   $CH-NH_2$  exhibit a weak

band near 1040 cm<sup>-1</sup> and a strong band near 1110 cm<sup>-1</sup>. 1 Amines containing a

tertiary carbon atoms 
$$(C_1 - C_2 - NH_2)$$
 absorb near 1030 cm<sup>-1</sup> (w) and 1200 cm<sup>-1</sup> (s).

The **aromatic amines** exhibit a characteristic C—N stretching vibrations in the region 1360–1250 cm $^{-1}$ .

Primary aromatic amine  $1340-1250 \text{ cm}^{-1}$ Secondary aromatic amine  $1350-1280 \text{ cm}^{-1}$ Tertiary aromatic amine  $1360-1310 \text{ cm}^{-1}$ 

The amino group can also be confirmed by converting into amine salts (NH<sub>3</sub>), which exhibit the following absorptions:

	1° Amine salts	2° Amine salts	3° Amine salts
N—H Stretching vibration	3200-3000 cm <sup>-1</sup>	2800-2400 cm <sup>-1</sup>	2500-2600 cm <sup>-1</sup>
N—H Bending vibration	1625-1560 cm <sup>-1</sup> 1550-1505 cm <sup>-1</sup>	1620-1560 cm <sup>-1</sup>	The bands are not of any diagnostic use

#### Amino-acids

Amino acids are known to exist as zwitter ion (except when NH<sub>2</sub> group is directly attached to aromatic nucleus).

As in the case of carboxylate anion, ionized carboxyl groups give strong asymmetric  $\mathrm{CO}_2^-$  stretching vibrations near 1650–1550 cm<sup>-1</sup> and a weaker symmetric  $\mathrm{CO}_2^-$  stretching vibrations near 1440-1360 cm<sup>-1</sup>. Besdies these,  $\mathrm{NH}_3^-$  exhibit bands in the region 3200-2600 cm<sup>-1</sup> and 2200-2000 cm<sup>-1</sup>.

#### **Amides**

In non-polar solvents, dilute solutions of primary amides exhibit two intense bands due to free N—H stretching vibrations near 1500 and 3400 cm<sup>-1</sup> corresponding to asymmetric and symmetric N—H stretching vibrations. However, in the solid state (due to hydrogen bonding), the N—H stretching bands occur near 3350 and 3180 cm<sup>-1</sup>. The separation of these two bands (in the solid state) by about 150 cm<sup>-1</sup> is characteristic of amides. Secondary amides exist mainly in *trans* configuration. These exhibit free N—H stretching vibrations in dilute solutions near 3450 cm<sup>-1</sup>. However, in more concentrated solutions or in solid state, the secondary amides form dimers (*cis* configuration) (A) or polymers (B) (*trans* configurations). In this case, the free N—H stretching band (near 3450 cm<sup>-1</sup>) is replaced by multiple bands in the region 3330 – 3100 cm<sup>-1</sup>.

In addition to N—H stretching vibration (as stated above), the amides exhibit C = O stretching and N–H in plane bending vibrations (which generally occur in the solid state as doublet near 1655 and 1630 cm<sup>-1</sup>) give absorptions near 1690-1670 cm<sup>-1</sup> (C = O, called amide I band) and 1620–1615 cm<sup>-1</sup> (N—H in plane bending, called amide II band).

In secondary amides, the C = O stretching frequency occurs in the region 1680–1640 cm<sup>-1</sup>(S). However, tertiary amines do not form hydrogen bonds and these exhibit C = O stretching frequency in the range, 1690-1650 cm<sup>-1</sup>.

#### **Ethers**

Ethers exhibit absorption in the range 1300-1000 cm<sup>-1</sup> due to C—O—C stretching. However, a band in this region in also observed in other oxygen containing compounds like alcohols, aldehydes, ketones, carboxylic acids etc. So the presence of ether is confirmed only it there is no absorption in the hydroxy region (3650–3200 cm<sup>-1</sup>) or carbonyl region (1870–1650 cm<sup>-1</sup>).

Thus, as an illustration, in case of diethyl ether there is no absorption above 3000 cm $^{-1}$  excluding the presence of groups like —OH, —NH $_2$ , —NHR, H—C  $\equiv$  C — R and C $_6$ H $_5$ . However, following bonds are observed.

2990 and 2875 cm $^{-1}$  due to  $C_{sp}^3$  – H stretching frequencies of alkyl group.

1450 cm<sup>-1</sup> due to methylene scissoring and methyl asymmetre bending vibrations.

 $1140\ cm^{-1}$  due to symmetrical C—O—C stretching vibration of ether group.

A compound,  $C_2H_6O$ , can either be dimethyl elter ( $CH_3$ —O— $CH_3$ ) or ethyl alcohol ( $CH_3CH_2OH$ ). How will you differentiate between the two on the basis of IR spectra.

**Sol.** Both compounds have  $sp^3$  -hybridised C—H bonds ( $\sim 3000 \text{ cm}^{-1}$ ). However, ether does not have an OH group, but ethyl alcohol has an OH group (absorption at  $3600-3200 \text{ cm}^{-1}$ ).

#### Nitriles and Isonitriles

Aliphatic nitriles absorb near 2260–2240 cm<sup>-1</sup>. Electron attracting atoms, such as oxygen or chlorine, attached to the carbon alpha to the  $C \equiv N$  group reduce the intensity of absorption. Conjugation such as in aromatic nitriles, reduces the frequency of absorption to 2240–2222 cm<sup>-1</sup> and enhance the intensity.

The isonitriles, however, absorb strongly in the region 2185–2121 cm<sup>-1</sup>.

#### **Nitro Compounds**

There are two identical NO bonds in nitro group. These cause strong absorption in the region 1565–1515 cm<sup>-1</sup> due to asymmetric stretching and in the region 1385–1335 cm<sup>-1</sup> due to symmetrical stretching.

In aliphatic nitro compounds, the bands are observed around 1550 and 1380 cm $^{-1}$ , whereas in aromatic nitro compounds the bands occur around 1530 and 1350 cm $^{-1}$ . The variation in the bands depend on the substituent on alpha carbon to NO<sub>2</sub> group in case of aliphatic compounds and on the o-/p- position with respect to NO<sub>2</sub> group in aromatic compounds. The presence of electronegative substituents on alpha carbon results in increase in asymmetric NO<sub>2</sub> $^-$  stretching frequency and decrease in the symmetric frequency. As an example, chloropicin absorbs at 1610 and 1307 cm $^-$ 1 respectively.

The presence of electron releasing group (e.g.,  $NH_2$ ) at o- or p- w.r.t.  $NO_2$  group in aromatic compound results in lowering the  $NO_2^-$  stretching frequency due to resonance occurring in the o- and p-isomers resulting in weakening of N—O bond (p-nitroaniline absorbs at 1475 and 1310 cm<sup>-1</sup>).

#### 23.8 INTERPRETATION OF THE INFRARED SPECTRA

The position of various absorption bands in the infrared chart is noted and compared with those given in Table 23.1. The diagnostic infrared absorption bands of some important compounds are given in Table 23.2.

Table 23.3 summaries the absorption bands (mostly stretching) of some common functional groups. The correlation of absorption bands of some important bands is given in Table 23.4.

Table 23.1 Characteristic Infrared frequencies for common compounds

Type of Compound	Characteristic	Frequencies (cm <sup>-1</sup> )
Alkanes	1850–3000 (s)	C—H stretch
	1480-1440 (s)	CH <sub>2</sub> and CH <sub>3</sub> band
	1370-1380 (s)	
	2900–2800 (w)	CH stretching

(Contd...)

Type of Compound		Characteristic	Frequencies (cm <sup>-1</sup> )
Alkenes	RCH=CH <sub>2</sub>	3080–3140 (m)	=C-H stretch
	vinyl	1640–1645 (m)	C=C stretch
	$R_2C = CH_2$	3080–3140 (m)	=C-H stretch
	vinylidene	1645–1650 (m)	C=C stretch
	cis RCH—CHR	1655–1660 (m)	C=C stretch
		3020-3010 (m)	=C—H stretch
		675–720 (m)	C—H out of plane bending
	trans RCH=CHR	1670–1675 (m)	C—H stretch
		3020-3010 (m)	=C-H stretch
		970–980 (s)	C—H out of plane bending
	R <sub>2</sub> C—CHR (trisubstituted)	1660–1670 (w)	C=C stretch
	R <sub>2</sub> C=CR <sub>2</sub> (tetrasubstituted)	1660–1670 (w)	C=C stretch
	$C = C - C = CH_2$	1660–1580 (s)	C=C stretch
	(conjugated)		
Alkynes	RC≡CH	3310-3300 (s)	C—H stretch
		2100-2140 (m)	C≡C stretch
		600-700 (s)	C—H bend
	RC≡CR	2190–2260 (w)	C≡C stretch
Benzene and	R	3080–3030	C—H stretch
its derivatives	monosubstituted	(1–5 bands)	C=C stretch
		1625–1440	
		(4 bands)	
		770–690	
		(2 bands)	
		1225–960	C—H bend
		(4 bands)	
	R o-disubstituted	770–735 (s)	
	R	(1 band)	

Type of Compound		Characteristic Frequencies (cn	
	R m-disubstituted	1170–1000 (3 bands) 900–680 (3 bands)	C—H bend
	R p-disubstituted	1225–1000 (3 bands) 860–800 (s) (1 band)	C—H bend
Alcohols	R—OH	3600–3200 (s)	O—H stretch
	(primary)	1350–1250 (s)	O—H bend
		1080–1010	C—O stretch
		3600-3200 (s)	O—H stretch
	СНОН	1350-1250 (s)	O—H bend
	(secondary)	1120-1030 (s)	C—O stretch
		3600-3200 (s)	O—H stretch
	_сон	1410-1300 (s)	O—H bend
	(tertiary)	1170-1100 (s)	C—O stretch
Phenols	R = 0	3500–2400 (s)	O—H stretch
	hydrogen bonded intermolecular	3400–3200 (s)	O—H stretch
	к п к Intermolecular (Chelate)		
Ethers	—CH <sub>2</sub> —O—CH <sub>2</sub> —	3570–3450 (s)	O—H stretch
	saturated acyclic	1150–1060 (s)	C—O stretch
	Ar—O—Ar/R aryl and arylakyl	1270–1250 (s)	C—O stretch
	-CH-CH <sub>2</sub>	980–970 (s)	C—O stretch
Ketones	c=o aliphatic	1720–1700 (s)	C=O stretch
	(saturated acyclic)  Ar—CO—R  aromatic	1700–1680 (s)	C=O stretch
	Ar—CO—Ar aromatic diaryl	1670–1660 (s)	C=O stretch
			(Contd.)

Type of Compound		Characteristic	Frequencies (cm <sup>-1</sup> )	
	c=o (cyclic)	1775–1705 (s)	C=O stretch	
	$R$   $-C=C-C=O$ $\alpha$ , $\beta$ -unsaturated (acyclic)	1680–1665 (s)	C=O stretch	
	C   C   C   C   C   C   C   C   C   C	1725–1665 (s)	C—O stretch	
	—CO—CO— α-diketone	1730–1710 (s)	C=O stretch	
	-CO-CH <sub>2</sub> -CO-	1640–1535 (s)	C=O stretch	
	β-diketone (enol)	2800–2600	O=H stretch	
	$\begin{array}{lll}\text{CO}\text{CH}_2\text{CH}_2\text{CO}\\ \text{$\gamma$-diketone} \end{array}$	1725–1705	C—O stretch	
	c=c=o ketenes	approx. 2150 (s)	C=O stretch	
Aldehydes	R—CH=O	1740-1720 (s)	C=O stretch	
	aliphatic saturated	2870–2710	C—H stretch	
		975-780 (w)	C—H bend	
	R—CH=CH—CH=O $\alpha$ , $\beta$ -unsatured	1705–1680 (s)	C=O stretch	
	ArCHO aromatic	1715–1695 (s)	C=O stretch	
Carboxylic	R—COOH	1725-1700 (s)	C=O stretch	
acids	aliphatic saturated	3550–3500 (m)	O—H stretch	
	R—CH—CH—COOH α, β-unsaturated	1715–1690 (s)	C=O stretch	
	ArCOOH aryl	1700–1680 (s)	C=O stretch	
		1680–1650(s) (hydrogen bonded	C—O stretch	
	X–HC <sub>2</sub> COOH α-halogeno acids	1740–1715 (s)	C=O stretch	
Anhydrides	R—CO	1850-1800 (s)	C=O stretch	
	R-CO O	1790-1740 (s)	C=O stretch	
	acyclic	1175–1045 (s)	C—O stretch	
	(ArCO) <sub>2</sub> O aromatic	1770–1720 (s)	C—O stretch	
			(Contd	

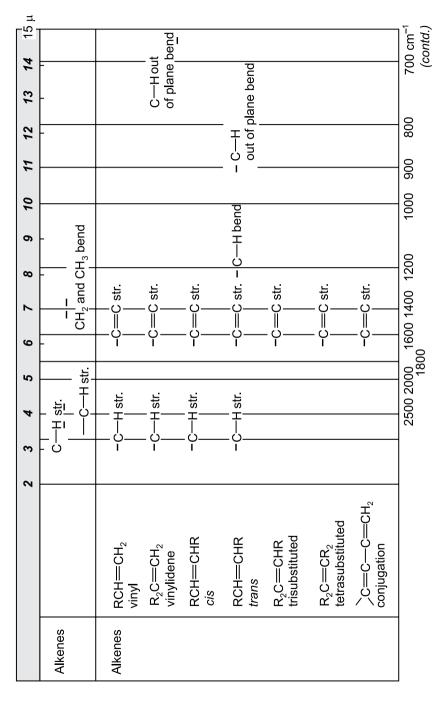
Type of Compound		Characteristic	Frequencies (cm <sup>-1</sup> )
	CH₂CO ੑ	1870–1820 (s)	C=O stretch
	CH <sub>2</sub> CO	1800–1750 (s)	C=O stretch
	cyclic saturated 5-membered	1310–1210 (s)	C—O stretch
	CO	1850–1800 (s)	C—O stretch
	cyclic (α, β-unsaturated)	1830–1780 (s)	C=O stretch
Esters	R-COOR'	1750–1735 (s)	C=O stretch
	saturated acyclic	1280–1050 (2 bands)	C—O stretch
	—C—C—COOR $\alpha$ , $\beta$ -unsaturated	1730–1715 (s)	C—O stretch
	—CO—COOR α-keto esters	1755–1740 (s)	C—O stretch
	—CO—CH <sub>2</sub> —COOR β-keto ester (enolic)	1655–1635 (s)	C=O stretch
	—CO—C—C—COOR (γ-keto ester)	1750–1735 (s)	C=O stretch
Lactones	C—C—C— O——CO saturated γ-lactone	1780–1760 (s)	C—O stretch
		1750–1735 (s)	C=O stretch
	$\begin{array}{c c} &   &   \\ -C-C=C- \\ &   &   \\ OCO \\ \alpha, \beta\text{-unsaturated} \\ \gamma\text{-lactone} \end{array}$	1760–1740 (s)	C—O stretch
	-C-C=C-C-C- $ C-C-C-C-C-C-C-C-C-C-C-C-C-C-C-C-C-C-C-$	1805–1785 (s)	C—O stretch
Amines	Ar—NH <sub>2</sub> or RNH <sub>2</sub> (primary)	3500–3200 (m) (2 bands)	N—H stretch
			(Contd.)

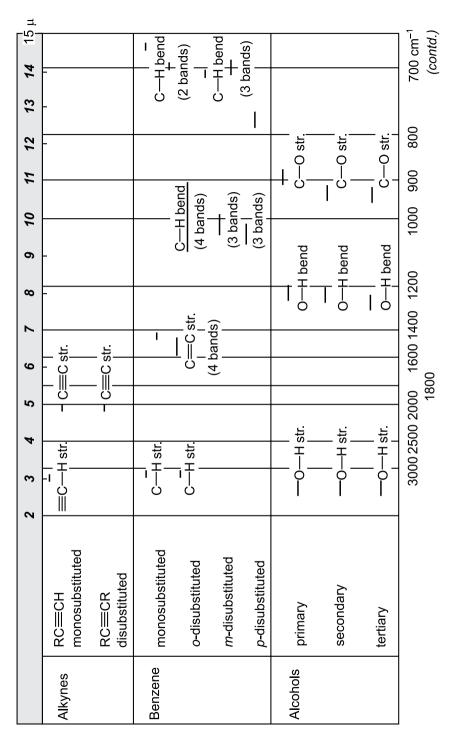
Type of Compound		Characteristic	Frequencies (cm <sup>-1</sup> )	
		1340–1200 (s)	C—N stretch (aromatic)	
		1650-1590 (m)	N—H bend	
	NH	3500-3200 (m)	N—H stretch	
	secondary	1360-1310 (s)	C—Nstretch(aromatic)	
		1580–1490 (w)	N—H bend	
	NH tertiary	1360–1310 (s)	C—Nstretch(aromatic)	
Imine	=NH	3400-3000 (m)	N—H stretch	
Amides	—CONH <sub>2</sub> primary free	3500 (m) <sup>7</sup> 3400 (m)	N—H stretch	
		1690 (s)	C=O stretch	
		(dilute solution) 1620–1590	N—H bend	
	—CONH <sub>2</sub> hydrogen bonded	3350 (m) <sup>1</sup> / <sub>2</sub> 3180 (m)	N—H stretch	
	—CO—NH—	3430 (m)	N—H stretch	
	secondary free	1700–1670 (s)	C=O stretch	
		1550–1510	N—H bend	
	hydrogen bonded secondary amide	3320-3140 (m)	N—H stretch	
	-co-n	1670–1630 (s)	C=O stretch	
	tert. amide			
Imides	acyclic	1710 (s)	C=O stretch	
	cyclic (5-membered)	1770 (s),	C=O stretch	
		1700 (s)		
	cyclic (6-membered)	1710 (s),	C=O stretch	
		1700 (s)		
Ureas	acyclic	1660 (s)	C=O stretch	
Lactams	β-lactam	1760–1730 (s)	C=O stretch	
	γ-lactam	approx. 1700 (s)	C=O stretch	
	$\delta$ -lactam	approx. 1680 (s)	C=O stretch	
Nitro	R—NO <sub>2</sub> aliphatic and aromatic	1570–1550 (s) 1380–1370 (s)	N=O stretch	

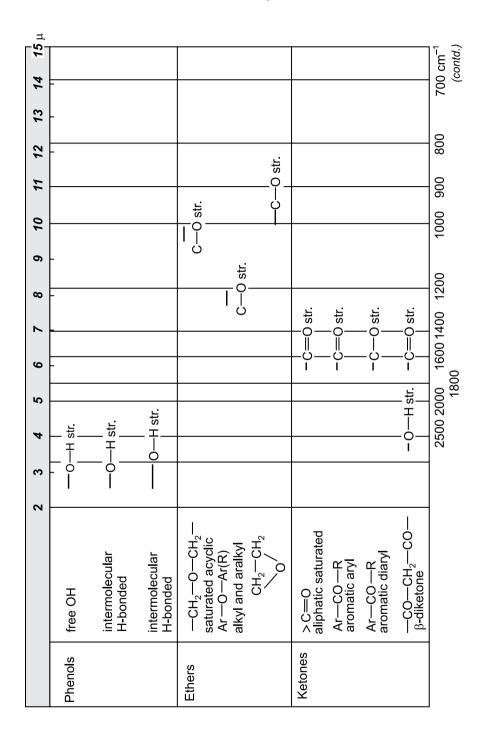
Type of Compound		Characteristic Frequencies (cm	
Oximes —C—N—OH		3650–3500	O—H stretch
		1690–1640	C=N stretch
Azo	—N==N—	1640	N=N stretch
Nitriles	R—CN aliphatic, saturated	2250 (m)	C=N stretch
	—C—C—CN aliphatic, α, β-unsaturated	2225 (m)	C≡N stretch
	ArCN aromatic	2240-2220 (m)	C≡N stretch
Isocyanides	C—NC	2220–2070	C≡N stretch
Isocyanates	C—NCO	2275–2240	C=N stretch
Nitrates	C—O—NO <sub>2</sub>	1655–1610 (s)	N=O stretch
		1300–1250 (s)	
Nitrites	C—O—NO	1680–1610 (s)	
		1300-1250 (s)	N=O stretch
Nitroso	C—NO	1600-1500 (s)	N=O stretch
Alkyl halides	R—X	1400-1000 (s)	C—F stretch
		800–600 (s)	C—CI stretch
		600-500 (s)	C—Br stretch
		500 (s)	C—I stretch
Thiols	C—SH	2600–2550 (w)	S—H stretch
Thioketones	_c=s	1200–1050 (s)	C=S stretch

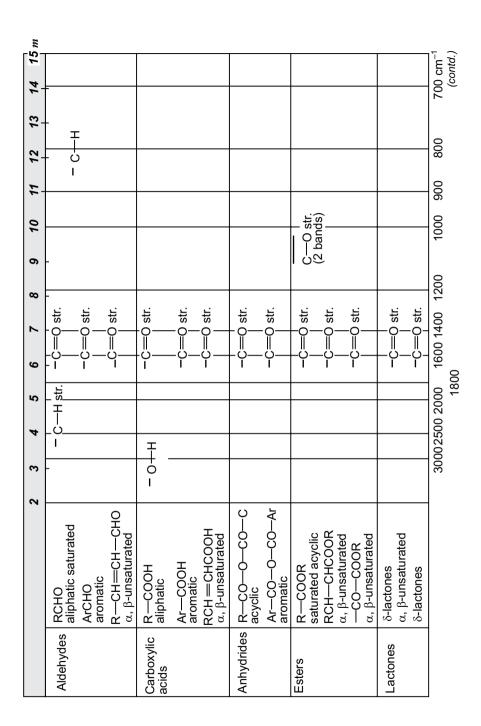
S = strong; m = medium; w = weak.

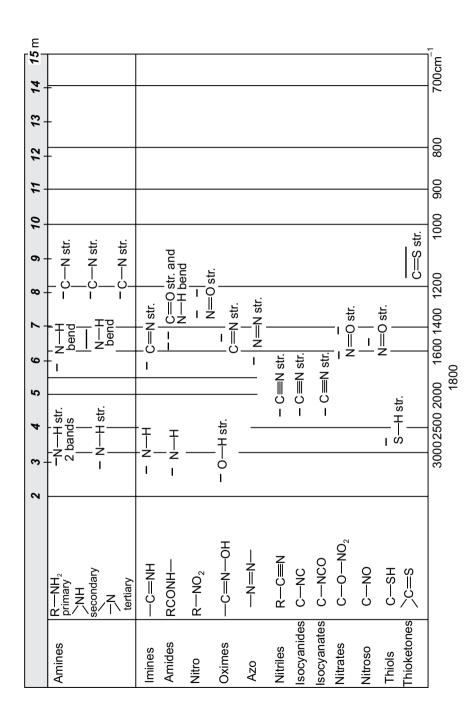
Table 24.2 Diagnostic Infrared frequencies for some compounds







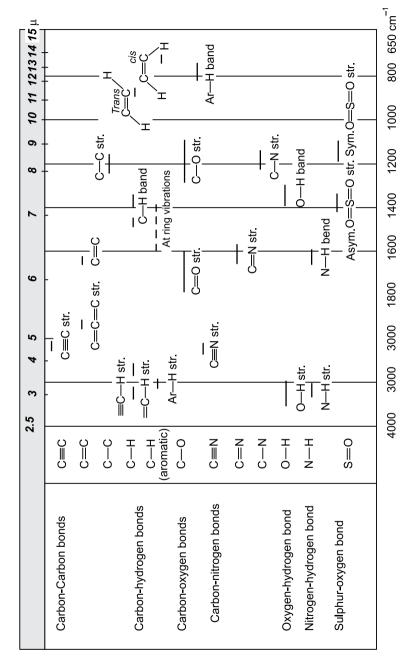




**Table 23.3** Infrared Absorption Peaks (mostly stretching) of some common functional groupings

	stic frequencies (cm <sup>-1</sup> )	s F	Function Groupings			
1050–1400 1150–1360 1440–1475 1340–1500 1450–1600 1620–1680 1630–1690 1690–1750	$\begin{array}{c} \text{C}\text{O} \\ \text{SO}_2 \\ \text{C}\text{H} \\ \text{NO}_2 \\ \text{C}\text{C} \\ \text{C}\text{C} \\ \text{C}\text{C} \\ \text{C}\text{O} \\ \text{C}\text{O} \\ \text{C}\text{O} \\ \text{OH} \\ \end{array}$	(in ethers, alcohols and esters) (in sulphonic acid derivatives) (2 bands seen) (in alkenes) bending mode (2 bands) (bands in aromatic ring) (usually several peaks) (not for symmetrical alkenes) (in amides, O—C—N) (2 bands) (in aldehydes, ketones and esters) (in carboxylic acids)				
1770–1820	C=O   CI	(in acid chlorides)				
2100–2200 2210–2260 2500 (one p	C≕C C≕N eak) S—H	(not for symmetrical alkynes) (nitriles)				
2700–2820 2500–3000 3000–3100	C—H O—H C—H	(of aldehydes) (in COOH) (C is part of aromatic ring)				
3300 3020–3080 2800–3000	C—H C—H C—H	(C is acetylenic) (C is in ethylenic) (C is in alkanes)				
3300–3500 3200–3600 2100	N—H O—H O—D	(in amines and amides) (in H bonded ROH and ArOH)				
	Bending f	requencies of I	nydrocarbons	s (cm <sup>-1</sup> )		
Alkenes	CH <sub>3</sub>	$=$ CH $_2$	-CH(CH <sub>3</sub>	) <sub>2</sub> –C(	CH <sub>3</sub> ) <sub>3</sub>	
	1420–1470, 1375	1430–1470	doublet of intensity a 1375, 187 and also	at 137 75 139	blet at 0 (strong) 5 (moderate)	
Alkenes (out of plane)	RCH=CH <sub>2</sub> 910-920 990-1100	R <sub>2</sub> C=CH <sub>2</sub> 880-900	R CH—C cis 675–7 trans 965	30		
Aromatic C—H (out of plane	<del>)</del>	Monosubstit 690–710 730–770	uted <i>ortho</i> 700–770	Disubstitute meta 690–710 750–810	ed <i>para</i> 810–840	





#### 23.9 APPLICATIONS OF INFRARED SPECTROSCOPY

Following are given some important applications of IR spectroscopy:

- (i) **Identification of an Organic Compound:** The identity of an organic compound is decided on the basis of spectra in the fingerprint region (from 1400 cm<sup>-1</sup> to 900 cm<sup>-1</sup>) which is characteristic of a compound. The compounds containing the same functioned groups may have similar absorptions above 1500 cm<sup>-1</sup> (functional group region) but their spectras differ in the fingerprint region. On the other hand, identical compounds will have superimpossible IR spectra.
- (ii) **Progress of a Reaction:** Most of the reactions, as we know, involve change in the functional groups of the reacting moieties. By periodic examination of a small portion of reaction mixture, is sufficient to indicate the progress of the chemical reaction. For example, if an alcohol is oxidised to an aldehyde or a ketone, can be followed by appearance of the carbonyl band and disappearance of the OH band. In fact, the reaction can be carried out in special cells which are placed in the path of the IR beam of the instrument.
- (iii) **Detection of Functional Groups:** Careful examination of the functional group region (1400 900 cm<sup>-1</sup>) gives an idea of the presence of a functional group in an organic compound (*see* section 23.7).
- (iv) **Determination of Structure:** IR spectroscopy is helpful to establish the structure of a compound. For example, amino acids are known to exist as Zwitter ions. The IR spectra does not give bands due to COOH or NH<sub>2</sub> groups. However, absorption at 1650–1550 cm<sup>-1</sup> (due to asymmetric CO<sub>2</sub><sup>-</sup> stretching vibration) and bands near 3200–2600 cm<sup>-1</sup> and 2200–2000 cm<sup>-1</sup> are observed due to NH<sub>3</sub><sup>+</sup>.
  - In a similar way, the IR spectra of sulphanalic acid gives absorptions due to  $NH_3^-$  and  $SO_3^-$ .
- (v) **Study of Complex Molecules:** IR spectroscopy is helpful to establish the structure of complex molecules. A typical example is the establishment of  $\beta$ -lactam structure of penicillin and differentiate between the  $\beta$ -lactam structure and oxazolone structure.

The IR spectra of a number of model 5 (4H) oxazolones of the type II exhibited absortion band at  $1825~\text{cm}^{-1}$  (C = O) and  $1675~\text{cm}^{-1}$  (C  $\equiv$  N stretching). None of these bands was observed in the IR spectra of penicillins. This eliminated what was earlier thought to be a structure (II) of penicillin.

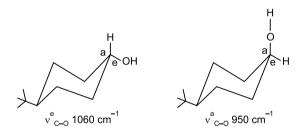
In simple  $\beta$ -lactams there is no absorption band near 1770 cm<sup>-1</sup>. However,  $\beta$ -lactam fused to a thiazolidine ring shows a band at 1770 cm<sup>-1</sup>. Thus, pencillins had the  $\beta$ -lactam structure.

#### (vi) Keto-enol Tautomerism

On the basis of IR spectra of ethyl acetoacetate, it was found to be a mixture of keto and enolic forms. The IR spectra showed the presence of keto group [absorption at 1733, 1710 cm $^{-1}$  (doublet)], hydroxy group [absorption at 3300 cm $^{-1}$  (w, broad)] and C = C (absorption at 1645 cm $^{-1}$ ). On the basis of the IR spectra it is possible to find the ratio of keto and enolic forms.

In a similar way keto and enolic forms were found to be present in  $\beta$ -diketones.

(vii) Conformation Studies: On the basis of IR spectra it is possible to differentiate between axial and equatorial positions. It is found that trans *t*-tert butyl cyclohexanone shows absorption due to C—O stretching at 1060 cm<sup>-1</sup> and the *cis* form at 950 cm<sup>-1</sup>. The higher frequency of C—X bond in equatorial position is attributed to less steric interaction of C—X bond with adjacent hydrogens.



(viii) **Geometrical Isomerism**: In alkenes, if there is no hetero atom substitutent on the  $C_{sp^2}$  carbon, the IR bands in general take place at separate positions in cis-(970 cm<sup>-1</sup>) and trans (730–650 cm<sup>-1</sup>) alkenes. In case the  $C_{sp^2}$  carbon has a hetero atom as a substituent, the stretching vibration of the = C—X bond is higher by 20–40 cm<sup>-1</sup> in trans-isomers compared to that in cis-isomers.

(ix) **Rotational Isomerism:** In alkanes, *e.g.*, 1,2-dichloroethane, the Gauche (skew) form absorbs as 1235 cm<sup>-1</sup> and the staggered (trans) form absorbs at 1291 cm<sup>-1</sup>. The staggered form predominates at lower temperature and the gauche form becomes more prominent as the temperature is raised.

In case of  $\alpha$ -halocarbonyl compounds, *e.g.*, chloroacetone the two carbon atoms are  $sp^2$  and  $sp^3$  hybridised respectively. In such cases, the eclipsed form the IR band is observed at about 1745 cm<sup>-1</sup> and in the staggered form the absorption takes place at 1725 cm<sup>-1</sup>.

The low frequency of absorption in case of eclipsed conformation is attributed to replusion of negative field of chlorine by the non-bonding electrons of oxygen atom.

- (x) **Determination of Purity of a Sample:** In case reference spectrum of a pure compound is available, it is possible to determine if the compound under investigation is impure. Compared to the spectra of a pure compound, the spectra of a impure compound is generally blurred and contains extra bonds. As an example, the presence of a ketone as impurity in an alcohol gives C=O absorption peak in the IR spectra of impure alcohol.
- (xi) Application of 1R spectroscopy to Inorganic Compounds (Salts).

# (a) Identification of inorganic ions

It was only in 1952 that Miler and Walkin's carried out investigation about the identification of inorganic ions (*see* Table ahead):

**Table 23.5** IR absorption of inorganic ions

Carbonates (CO <sub>3</sub> <sup>2-</sup> )	1450–1410 cm <sup>-1</sup> (vs); 880–860 cm <sup>-1</sup> (m)
Nitrate (NO <sub>3</sub> <sup>-</sup> )	1380–1350 cm <sup>-1</sup> (vs); 840–815 cm <sup>-1</sup> (m)
Nitrite (NO <sub>2</sub> <sup>-</sup> )	1250–1230 cm <sup>-1</sup> (vs); 840–800 cm <sup>-1</sup> (w)
Sulphate (SO <sub>4</sub> <sup>-2</sup> )	1130–1080 cm <sup>-1</sup> (vs); 680–610 cm <sup>-1</sup> (m)
Phosphate PO <sub>4</sub> <sup>3-</sup> , HPO <sub>4</sub> <sup>2-</sup> , H <sub>2</sub> PO <sub>4</sub> <sup>-1</sup> Silicates	1100–1000 cm <sup>-1</sup> (s) 1100–900 cm <sup>-1</sup> (s)
Cyanides, thiocyanates, cyanates	2200–2000 cm <sup>-1</sup> (s)
Ammonium (NH <sub>4</sub> <sup>+</sup> )	3300 –3030 cm <sup>-1</sup> (vs); 1430–1320 cm <sup>-1</sup> (s).

#### Notes:

- 1. Most of the carbonates show IR absorptions as mentioned in the table. However, in case of PbCO<sub>3</sub>, the second band is observed at 840 cm<sup>-1</sup>.
- 2. In case of bicarbonates no difinite correlation have been made.
- 3. The IR spectra of basic carbonates is very complex. A number of bands occur in the range 1110-700 cm<sup>-1</sup>.

# (b) Study of inorganic complexes

It is rather difficult to perform the spectral analysis of complexes even if their structures are known. This is mainly because of the presence of a number of atoms in the complex, there are a large number of modes of vibrations, thus making the spectra complex. It is however, possible to study their geometrical isomerism. For example, bipyridyl cobalt (III) chloride mixed complex (emprical formula [Co(Bipy)<sub>2</sub>(Cl<sub>2</sub>)Cl] exist in geometrical isomers. Using IR spectra, it was found that violet compound is the trans form whereas the pink compound is the *cis* form.

#### 23.10 IR SPECTRAS OF SOME TYPICAL COMPOUNDS

In this section IR spectras of some typical compounds are given along with the assignment of various peak. The IR spectras of following compounds are given below:

Hydrocarbons	1-Octene (Fig. 23.4) Propyl benzene (Fig. 23.5)
Hydroxy compounds	Benzyl alcohol (Fig. 23.6) Phenol (Fig. 23.7)
Ethers	Dipropyl ether (Fig. 23.8) Phenetole (Fig. 23.9)
Carbonyl compounds	Diethyl ketone (Fig. 23.10) Benzaldehyde (Fig. 23.11)

Carboxylic acids and their
derivatives

Ethyl acetate (Fig. 23.13)
Benzamide (Fig. 23.14)

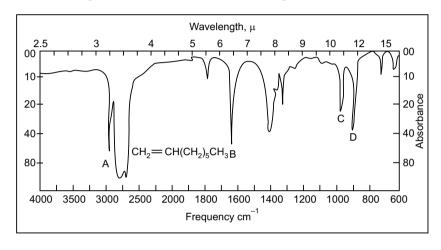
Cyanides

Benzonitrile (Fig. 23.15)

Nitro compounds

Amino compounds

Aniline (Fig. 23.17)



**Fig. 23.4** IR spectrum 1-octene of  $(CH_2 = CH(CH_2)_5CH_3)$  A. 3100 cm<sup>-1</sup> (olefinic C—H stretch), B. 1630 cm<sup>-1</sup> (C = C stretch), C. and D. 980, 900 cm<sup>-1</sup> (= C—H out of plane bend)

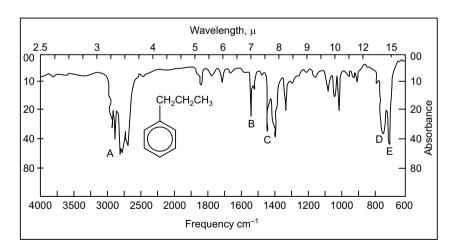
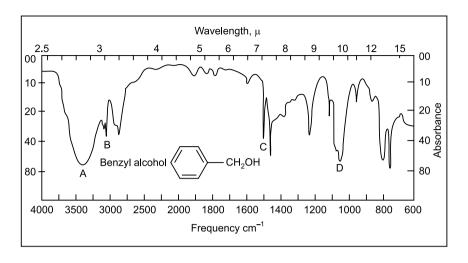


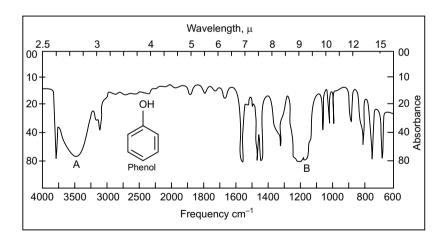
Fig. 23.5 IR spectrum of n-propyl benzene

A. 3050 cm<sup>-1</sup> (olefinic C—H stretch) , B. 1600 and C. 1500 cm<sup>-1</sup> (C = C stretch)

D. 750 and E. 700 cm<sup>-1</sup> (=C—H out of plane bend)



 $\label{eq:Fig. 23.6} \begin{array}{l} \text{Fig. 23.6 IR spectrum of benzyl alcohol} \\ \text{A. 3300 cm}^{-1} \text{ (O--H stretch), B. 3100 C. 1500 cm}^{-1} \text{ (C--C ring stretch)} \\ \text{D. 1017 cm}^{-1} \text{ (C--O stretch)} \end{array}$ 



 $\label{eq:Fig. 23.7} \mbox{ IR spectrum of phenol}$  A. 3350 and 3600 cm  $^{\!-1}$  (O—H stretch), B. 1200 cm  $^{\!-1}$  (C—O stretch)

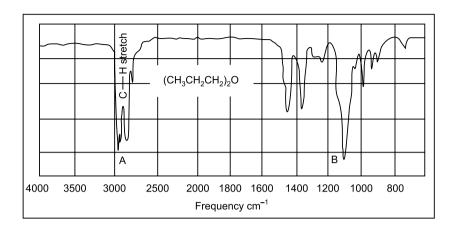


Fig. 23.8 IR spectrum of di-n-propyl ether A. 2950 cm<sup>-1</sup> (C—H stretch), B. 1110 cm<sup>-1</sup> (C—O stretch)

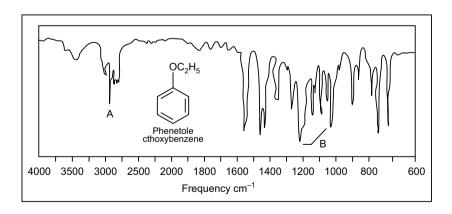


Fig. 23.9 IR spectrum of phenetole A. 2950 cm<sup>-1</sup> (C—H stretch), B. 1230 and 1050 cm<sup>-1</sup> (C—O stretch)

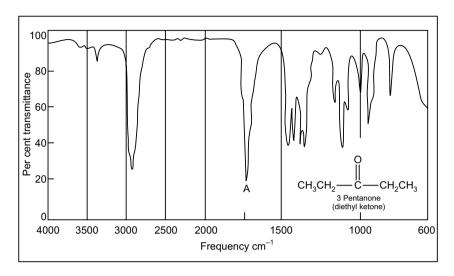
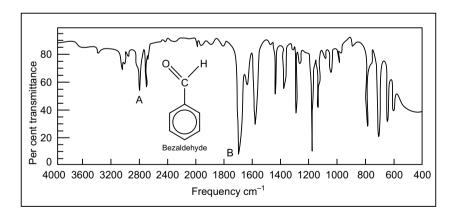


Fig. 23.10 IR spectrum of 3-pentanone (diethyl ketone) A. 1715 cm<sup>-1</sup> (C = O stretch)



 $\label{eq:Fig. 23.11} \ \ IR \ spectrum \ of benzaldehyde$  A. 2800 and 2780 cm  $^{-1}$  (C — H stretch of aldehyde), B. 1700 cm  $^{-1}$  (C = O stretch)

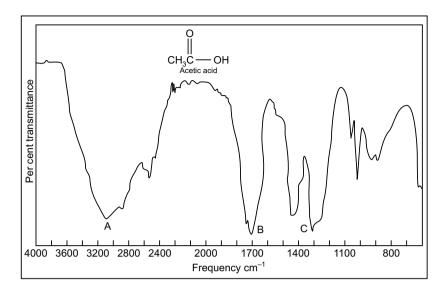


Fig. 23.12 IR spectrum of acetic acid A. 3100 cm  $^{-1}$  (O—H stretch), B. 1700 cm  $^{-1}$  (C = O stretch) , C. 1290 cm  $^{-1}$  (C—O stretch)

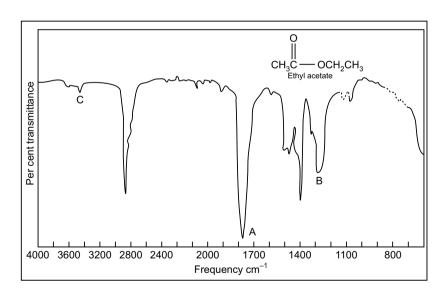
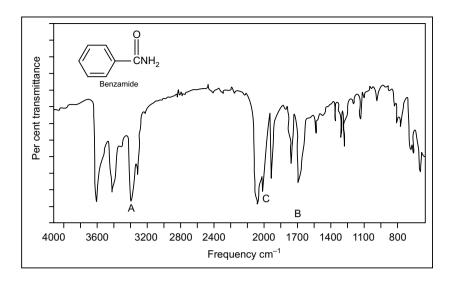


Fig. 23.13 IR Spectrum of ethyl acetate A. 1740 cm<sup>-1</sup> (C=0 stretch), B. 1250 cm<sup>-1</sup> (C=0 stretch), C. 3478 cm<sup>-1</sup> (overtone of C=O stretch)



 $\label{eq:Fig. 23.14} \begin{array}{l} \text{Fig. 23.14} \hspace{0.1cm} \text{IR Spectrum of Benzamide} \\ \text{A. 3370 cm}^{-1} \hspace{0.1cm} \text{(N--H stretch), B. 1660 cm}^{-1} \hspace{0.1cm} \text{(C == O stretch),} \\ \text{C. 1631 cm}^{-1} \hspace{0.1cm} \text{(N--H stretch)} \end{array}$ 

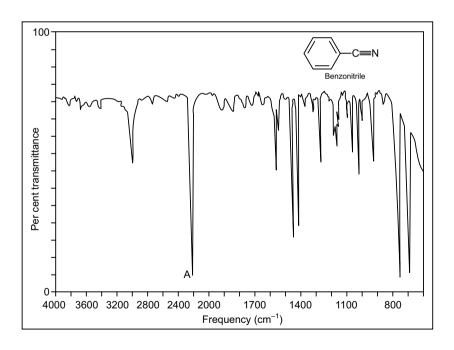


Fig. 23.15 IR spectrum of benzonitrile A. 2210 cm<sup>-1</sup> ( $C \equiv N$  stretch)

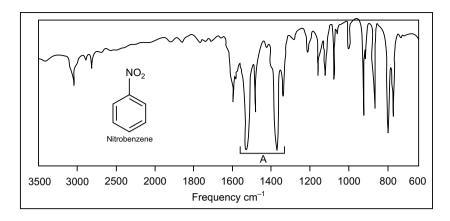
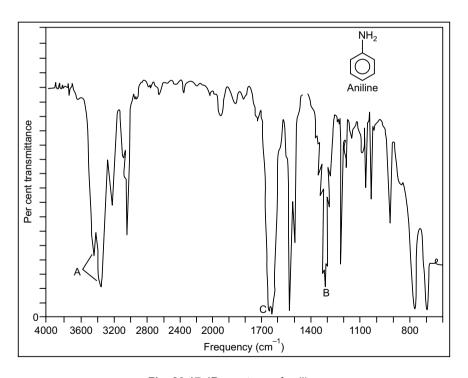


Fig. 23.16 IR spectrum of nitrobenzene A.  $1520 \text{ cm}^{-1}$  and  $1345 \text{ cm}^{-1}$  (N = 0 stretch)



**Fig. 23.17** IR spectrum of aniline

A. 3440 and 3350 cm<sup>-1</sup> (N—H stretch), B. 1280 cm<sup>-1</sup> (C—N stretch),

C. 1620 cm<sup>-1</sup> (N—H bend)

#### 23.11 NON-DISPERSIVE INFRARED SPECTROSCOPY

This technique is commonly used to monitor gas streams particularly in air pollution studies. In non-dispersive IR spectroscopy, the IR radiation from a source is not dispersed according to wavelength by a prism as in a standard IR spectrometer. It depends mainly on selective filtering for specificity. The radiation from an IR source is chopped by a rotating device so that it alternately passes through a sample cell and a reference cell (Fig. 23.18). Both the beams are incident on a detector which is filled with CO gas. The detector is divided into two compartments by a flexible diaphragm. The relative amounts of IR radiations absorbed by CO in the two sections of the detector are determined by the CO level in the sample. The difference in the amount of IR radiations absorbed in the two compartments give rise to temperature difference so that the diaphragm even to a slight extent can be measured with relative accuracy of 5 per cent.

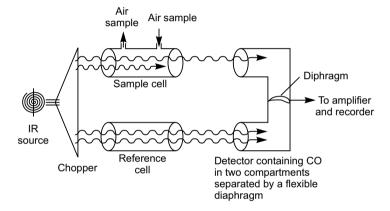


Fig. 23.18 Non-dispersive IR analyser for monitoring CO



#### I. Multiple Choice Questions

- 1. Which of the following statements are correct?
  - (a) The infrared region in the electromagnetic spectrum is between the visible and microwave region.
  - (b) The region 12500–4000 cm<sup>-1</sup> is referred to as near IR region.
  - (c) The region  $666-50 \text{ cm}^{-1}$  is referred to as for IR region.
  - (d) All are correct.
- 2. The fingerprint region in the IR spectrum is
  - (a)  $900-1400 \text{ cm}^{-1}$

(b) 12500-400 cm<sup>-1</sup>

 $(c) 666-50 \text{ cm}^{-1}$ 

(d)  $666-4000 \text{ cm}^{-1}$ 

3.	A compound shows IR absorption in tof	the range 2100–2260. It is due to the presence
	$(a)$ C $\equiv$ N	$(b) C \equiv C$
	(c) C = C	(d) C = O
4.	A compound showing IR absorption at 2760 and 2850 cm <sup>-1</sup> can be an	in the range 1680–1800 cm <sup>-1</sup> and two bands
	(a) Aldehyde	(b) Ketone
	(c) Alkene	(d) None of the above
5.	The presence of a broad absorption pethe presence of	ak in the region 3200 and 3600 cm <sup>-1</sup> indicates
	(a) OH group	(b) NH group
	$(c) C \equiv N$	(d) None of the above
6.	The carbon-hydrogen stretching band in the range	d of the type C $(sp^3)$ –H shows IR absorption
	(a) $3300 \text{ cm}^{-1}$	$(b) 3000-3100 \text{ cm}^{-1}$
	$(c) 2800-3000 \text{ cm}^{-1}$	(d) In any of the above
7.	The C-C bonds of benzene rings sho	ws a set of characteristic peaks in the region
	(a) $1620-1680 \text{ cm}^{-1}$	(b) $2100-2260 \text{ cm}^{-1}$
	$(c) 1450-1600 \text{ cm}^{-1}$	(d) Both $(a)$ and $(b)$
8.	Trans-alkenes normally give absorpt	ion peak in the region
	(a) $675-730 \text{ cm}^{-1}$	(b) 960–975 cm <sup>-1</sup>
	$(c) 1620-1680 \text{ cm}^{-1}$	( <i>d</i> ) None of the above
9.	A compound exhibiting $C = O$ absolute to	orption in the range 1710–1780 cm <sup>-1</sup> can be
	(a) Aldehyde	(b) Ketone
	(c) Ester	(d) Carboxylic acid
10.	An amine showing two peaks in the	region $3300 - 3500 \text{ cm}^{-1}$ can be
	(a) primary amine	(b) Secondary amine
	(c) Tertiary amine	( <i>d</i> ) None of the above
11.	Amides exhibit bands in the region	
	(a) $1630-1690 \text{ cm}^{-1}$	(b) $1710-1610 \text{ cm}^{-1}$
	(c) $1670-1642 \text{ cm}^{-1}$	(d) Any of the above
12.	Aromatic nitro compounds absorb in	_
	(a) $1550 \text{ cm}^{-1}$	(b) $1372 \text{ cm}^{-1}$
	$(c) 1360-1290 \text{ cm}^{-1}$	(d) $1550-1500 \text{ cm}^{-1}$
13.	Aromatic nitriles absorb in the IR re	
	(a) 2240–2222 cm <sup>-1</sup>	(b) 2260–2240 cm <sup>-1</sup>
	$(c) 1360-1290 \text{ cm}^{-1}$	(d) $1550-1500 \text{ cm}^{-1}$

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14.	The O—H stretch range	frequency in case	e of	intermolecular	hydrog	en bonding is in the
	(a) 3400–3200 cm	$^{-1}$ (s)	(	<i>b</i> ) 3500–3450 c	m <sup>-1</sup> (s)	1
	(c) 1350–1250 cm	$^{-1}$ (s)	(	d) 3600–3200 c	m <sup>-1</sup> (s)	1
15.	$\alpha$ , $\beta$ -Unsaturated a	aldehydes shows	IR a	bsorption in th	e range	;
	(a) 1740–1720 cm	-1	(	<i>b</i> ) 1725–1700 c	$\mathrm{m}^{-1}$	
	(c) 1715–1690 cm	-1	(	d) 1705–1680 c	$\mathrm{m}^{-1}$	
16.	A compound exhib	oiting N—H stret	tch b	and in the region	on 1360	$0-1310 \text{ cm}^{-1} \text{ can be}$
	(a) Primary amine		(	b) Secondary a	mine	
	(c) Aromatic tertia	ry amine	(	d) None of the	above	
17.	A compound exhib		e reg	ion 1570–1550	cm <sup>-1</sup> a	nd 1380–1370 cm <sup>-1</sup>
	(a) Nitro group		(	b) Nitrate group	)	
	(c) Nitrite group		(	d) Nitroso grou	p	
18.	Identical compoun	nd have				
	(a) Similar IR spec	etras				
	(b) Identical absor	ptions in the fund	ction	nal group region	n	
	(c) Superimpossib	le IR spectras				
	(d) None of the ab	ove				
19.	The progress of the can be followed by		ing	oxidation of se	condar	y alcohol to ketone
	(a) Appearance of	carbonyl band	(	b) Disappearan	ce of th	ne OH bond
	(c) Both (a) or (b)		(	d) None of the	above	
20.	IR spectroscopy is	helpful for				
	(a) Identification of	of an organic com	npou	nd		
	(b) Following prog	gress of a reaction	n			
	(c) Both (a) and (b	p)				
	(d) None of the ab	ove				
		ANS	ΝE	RS ‡		
1.	(d) 2.	(a)	3.	(b)	4.	(a)
5.	(a),(b) 6.	(c)	7.	(c)	8.	( <i>b</i> )
9.	(d) <b>10.</b>	(a)	11.	(a),(b),(c)	12.	(c), (d)
13.	(a) 14.	( <i>b</i> )	15.	( <i>d</i> )	16.	(c)
17.	(a) 18.	(c)	19.	(c)	20.	(c)

# II. Short and Long Answer Questions

- 1. Describe the electromagnetic spectrum indicating the usual IR region, near IR region, far IR region and fingerprint region.
- 2. Describe the various mode of vibrations in a molecule joined by covalent bond.
- **3.** How do you calibrate an IR spectrometer?
- **4.** How will you find that a carbonyl group belongs to an aldehyde, ketone, ester, carboxylic acid or an amide?
- 5. On the basis of IR spectra, how will you differentiate between primary, secondary and tertiary amine?
- **6.** How will you find that a nitro group is present in nitro alkanes or aromatic compound.
- 7. On the basis of IR spectra, how will you differentiate between a *cis* and *trans*-alkenes?
- **8.** How will you differentiate between a monosubstituted, *o*-disubstituted, *m*-disubstituted and *p*-disubstituted benzene?
- **9.** Using IR spectroscopy, how will you differentiate between (*i*) an aldehyde and a ketone (*ii*) primary, secondary and tertiary amine?
- **10.** On the basis of IR spectra, how will you differentiate between (a) cyanide and isocyanide and (b) an alcohol and phenol?
- 11. What do you look for in the IR spectra of 1-octene, isopropyl bromide and *n*-butyl benzene, which will be helpful to establish their structure?
- 12. An organic compound (A), C<sub>7</sub>H<sub>6</sub>O on reduction gave a compound (B), C<sub>7</sub>H<sub>8</sub>O. The IR absorption peaks of A and B are:

 $A: 1700, 2780 \text{ and } 2800 \text{ cm}^{-1}$ 

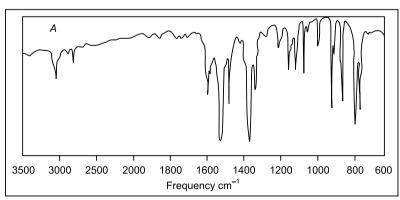
 $B: 1017, 3100, 1500 \text{ and } 3300 \text{ cm}^{-1}.$ 

Suggest structures of the compounds A and B.

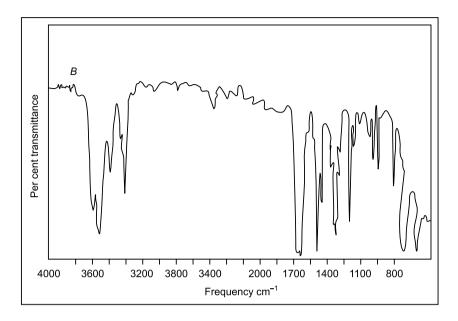
[**Ans.** *A* is Benzaldehyde *B* is Benzyl alcohol]

**13.** A compound *A* (mol. wt. 123) on treatment with tin and hydrochloric acid gave a compound *B* (mol. wt. 93). The IR spectra of both *A* and *B* are given below. Suggest structures of the compounds A and B.

[**Ans.** *A* is Nitrobenzene *B* is Aniline]

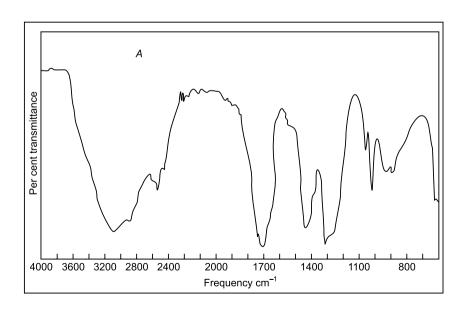


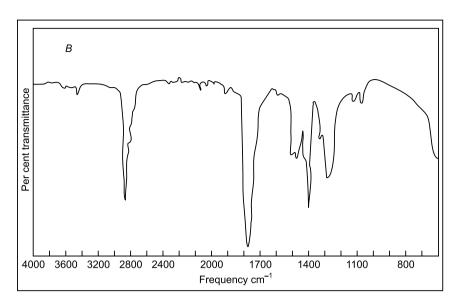




**14.** An organic compound A ( $C_2H_4O_2$ ) on heating with ethyl alcohol and conc.  $H_2SO_4$  gave a product B ( $C_4H_8O_2$ ). The IR spectra of both the products are given below. Suggest structures of A and B.

[Ans. A is Acetic acid B is Ethyl acetate]

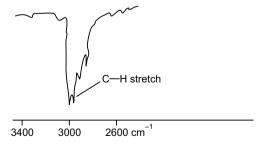




**15.** An organic compound A showed IR absorption at 2210 cm<sup>-1</sup>. On heating with dilute  $H_2SO_4$ , the peak at 2210 cm<sup>-1</sup> disappeared and peaks at 1290, 1700 and 3100 cm<sup>-1</sup> appeared. What type of conversion is effected in the above case.

[Ans. A cyanide is converted into carboxylic acid]

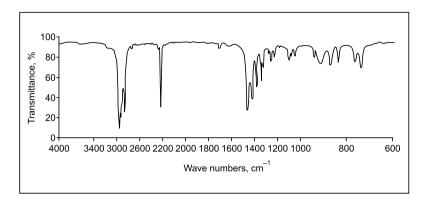
- **16.** Hydrogen molecule does not exhibit any absorption in the IR region. Explain why? [**Hint:** Hydrogen molecule does not display a hydrogen-hydrogen stretching vibration because the dipole moment does not change as the H—H bond is elongated or compressed.]
- 17. In the case of 4-octyne (CH<sub>3</sub>CH<sub>2</sub>CH<sub>2</sub>—C ≡ C—CH<sub>2</sub>CH<sub>2</sub>CH<sub>3</sub>), no absorption is observed due to C ≡ C stretching mode. Explain why?
   [Hint: The dipole moment for C ≡ C bond in internal alkynes is zero. There must be a net dipole change when the bond interacts with IR radiation.
- 18. What especial IR absorption do you expect in the IR spectra of 4-octyne  $(CH_1CH_2CH_2C) = C-CH_2CH_2CH_2C$ .



[Hint: Besides the absorption in the fingerprint region, there will be no absorption for the  $C \equiv C$  stretching vibration in the region 2300–2100 cm<sup>-1</sup>. The spectra

will show the typical C—H stretching vibrations in the region  $2800-3000 \text{ cm}^{-1}$  (*see* spectra on page 235). This is because the H atoms in 4-octyne are attached to  $sp^3$ -hybridged carbon atoms.

**19.** The IR spectra of a compound is given below. What functional group, in your opinion, is present in this compound?



[Hint: The functional group present most likely is  $C \equiv N$ . This is because there is a strong absorption band at about 2220 cm<sup>-1</sup>. The presence of terminal alkyne is ruled out since no absorption band is observed at 3300 cm<sup>-1</sup>.

**20.** Besides C=O absorption in the region (1680 – 1800 cm<sup>-1</sup>), what other features you look for in the IR spectra of acid chloride, aldehyde, amide (1°, 2° or 3°), carboxylic acid, ester and a ketone.

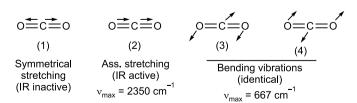
[Hint: The special features in the IR spectra of the above compounds are given below.]

Type of Compound	IR Absorption for C—O Group	Other Features
Acid chloride	1800	Single C = O stretching band
Aldehyde	1725	C—H stretching vibration at about 2720 cm <sup>-1</sup> . Can also be confirmed by $^1H$ NMR ( $\delta$ = 10.0).
Amide (1° or 2°)	1680	N—H stretching vibration at about 3300 cm <sup>-1</sup> .
Amide (3°)	1650	Single C $=$ O stretching band.
Carboxylic acid	1710	O—H stretching vibration at 3500–1500 cm <sup>-1</sup> . Can also be confirmed by $^1$ H NMR ( $\delta$ = 12.0).
Ester	1735	Strong $C = O$ stretching vibration at about 1200 cm <sup>-1</sup> .
Ketone	1715	No other special feature.

- 21. On the basis of IR spectroscopy, how will you differentiate between:
  - (i) Phenol and cyclohexane
  - (ii) cis and trans 2-butene
  - (iii) Acetaldehyde and acetone
  - (iv) cis-stilbene and trans-stilbene
  - (v) o- and p- hydroxybenzoic acid
  - (vi) Propionaldehyde and acetone
  - (vii) p-methoxyacetophenone and p-nitro acetophenone.
- 22. Explain non-dispersive infrared spectroscopy. Give its applications.
- 23. How does 1R spectra of carboxyl group and carboxylate ion differ?
- **24.** Amino acids do not exhibit the usual NH<sub>2</sub> absorption and COOH absorption. Explain why?

#### III. Solved Problems

- **Q.1.** When a molecule is subjected to IR radiation what is expected?
- **Ans.** Due to absorption of IR radiation by a molecule there is transition between vibrational and rotational energy levels which results in the formation of IR spectrum.
- **Q.2.** What is the normal IR region?
- **Ans.** The normal IR region is in the range 4000 cm<sup>-1</sup> to 667 cm<sup>-1</sup>. This region is particularly interested to organic chemists, since most of the absorptions of organic compounds are in this region.
- **Q.3.** Why certain organic compounds are IR inactive?
- **Ans.** In case there is no change in the dipole character of a compound on subjecting it to IR radiation, the compound does not show any IR absorption and is said to be IR inactive. Example includes, ethene, 2-butene.
- **Q.4.** In case of 1, 2-dichloroethylene, the *cis* form is IR active with respect to  $v_{c=o}$  where as trans form is not. Explain.
- **Ans.** In case of *cis* isomes, dipole moment changes occur. However in case of *trans* isomer no such changes occur since *trans* isomer is symmetrical.
- **Q.5.** In case of CO<sub>2</sub>, how many fundamental vibrational frequencies are expected in its IR spectrum.
- **Ans.** Being linear,  $CO_2$  will theoretically have  $(3n-5 \text{ or } 3 \times 3 5 = 4)$  four fundamental vibrational modes as shown below:



Thus,  $\mathrm{CO}_2$  will exhibit two fundamental vibrations at 2350 cm $^{-1}$  and 667 cm $^{-1}$ .

Q.6. How many vibrational degrees of freedom are expected in the following:

Ans. For linear molecules and non-linear molecules, the vibrational degrees of freedom are 3n-5 and 3n-6 respectively.

Thus, the above compounds will have 1, 3, 9, 30 and six vibrational degrees of freedom respectively.

- **Q.7.** Which will have higher absorption C—H or C—D stretching frequencies.
- **Ans.** C—H  $v_{\text{max}}$  2900 cm<sup>-1</sup> C—D  $v_{\text{max}}$  2060 cm<sup>-1</sup>
- **Q.8.** Arrange the following bands in their decreasing vibrational frequencies. Give reasons

Ans. As the atom bonded to C increases in mass, the frequency absorption decreases

cm
$$^{-1}$$
 C—H C—C C—O C—Cl C—Br C—I  $\nu_{\rm max}$  2460 1200 1100 800 550 500

- **Q.9.** Arrange C—C, C = C and C  $\equiv$  C in the increasing order of frequency.
- **Ans.** The frequency of vibration is directly proportional to the force constant of the band.

C—C 
$$C = C$$
  $C \equiv C$   
1200 cm<sup>-1</sup> 1650 cm<sup>-1</sup> 2150 cm<sup>-1</sup>

- **Q.10.** On the basis of hybridization, arrange the following C—H bonds in order of decrease in frequencies.
- Ans. The strength of the bonds is in the order

$$sp > sp^2 > sp^3$$

Thus, the frequencies decrease in shown:

$$\equiv$$
 C-H  $=$  C - H  $-$  C - H  $sp$   $sp^2$   $sp^3$   $3300 \text{ cm}^{-1}$   $3100 \text{ cm}^{-1}$   $2960 \text{ cm}^{-1}$ 

- **Q.11.** On the basis of IR spectra how will you differentiate between 1-hexene and 1-hexyne.
- $\begin{array}{lll} \textbf{Ans.} & \text{CH}_3\text{CH}_2\text{CH}_2\text{CH}_2\text{CH} = \text{CH}_2 \\ & 1\text{-Hexene} \end{array} & 3090 \text{ cm}^{-1} \text{ (C}_{sp}\text{2}\text{—H absorption)} \\ & 1640 \text{ cm}^{-1} \text{ (C} = \text{C streching absorption)} \\ & \text{CH}_3\text{CH}_2\text{CH}_2\text{C}\text{=CH} \\ & 1\text{-Hexyne} \end{array} & \text{Strong band at } 330 \text{ cm}^{-1} \text{ due to C}_{sp}\text{—H stretching} \\ & \text{absorption and a weak band at } 2120 \text{ cm}^{-1} \text{ due to} \\ & \text{C}\text{=C stretching absorption.} \\ \end{array}$
- **Q.12.** On the basis of IR spectra, how will you distinguish between a carbonyl, hydroxyl and ether group?
- **Ans.** Carbonyl group–strong absorption between  $1800-1650~\rm cm^{-1}$  Hydroxyl group–strong absorption between  $3650-3590~\rm cm^{-1}$  and strong absorption between  $1200-1000~\rm cm^{-1}$ .

Ether group-strong absorption between 1250-1000 cm<sup>-1</sup>.

- Q.13. Normally an unconjugated aliphatic aldehyde exhits C=O and C=C stretching at 1720 1740 cm<sup>-1</sup> and about 1650 cm<sup>-1</sup> resp. However, croton aldehyde exhibit absorptions at 1700 cm<sup>-1</sup> and 1630 cm<sup>-1</sup>. Explain why?
- Ans. Cotanaldehyde is resonance stabilised.

$$CH_3-CH-CH-C-O \longleftrightarrow CH_3CH-CH-C-O \longleftrightarrow H$$

Due to resonance both the frequencies of the carbonyl group (more single C—O bond character) and C = C absorption (more single C—C bond character).





# Chapter 24

# **Ultraviolet Spectroscopy**

#### 24.1 INTRODUCTION

The ultraviolet spectroscopy is comparatively less informative than the infrared or NMR spectroscopy. However, it is a valuable tool for the identification of organic compounds. The main features of ultraviolet spectroscopy is to get information about the extent of multiple bond and conjugation, aromatic and heteroaromatic systems.

The ultraviolet spectrum is not used to show the presence of individual functional groups as in the case with infrared spectroscopy. It is used to show the relationship between functional groups, mainly conjugation between two or more carbon-carbon double (or triple) bonds; between carbon-carbon and carbon-oxygen double bonds; between double bonds and an aromatic ring. The presence of an aromatic ring can also be inferred. It can also reveal the number and location of substituents attached to the conjugated system.

As an example, characteristic groups can be recognised in molecules of varying complexities. Thus, the spectrum of cholest-4-en-3 one closely resembles the spectra of mesityl oxide; the absorption results from conjugated enone portion of the componds.

$$\begin{array}{c} \text{O} \\ \text{CH}_3 - \text{C} - \text{C} = \text{C} \stackrel{\text{CH}_3}{\longrightarrow} \\ \text{Cholest-4en-3-one} \end{array}$$

The UV region of primary interest to an organic chemist is from 200 to 380 nm. This is known as near or common ultraviolet region. Most of the unsaturated

organic compounds absorb in this region. The saturated compounds absorb below 200 nm (far UV). The region extending from 380 to 800 nm is known as the visible range. All coloured compounds absorb in this region. The ultraviolet portion of the electromagnetic spectrum is indicated in Fig. 24.1.

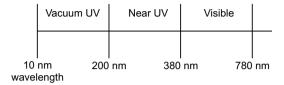


Fig. 24.1 Ultraviolet portion of the electromagnetic spectrum

In a standard UV spectrophotometer, a beam on light is split into two halves. One half of the beam (the sample beam) is directed through a transparent cell containing a solution of the compound being analysed, and the other half (the reference beam) is directed through an identical cell that contains only the solvent. Solvents are chosen to be transparent in the region of the spectrum being used for analysis. The instrument is so designed that it makes a comparison of the intensities of the two beams as it scans over the desired region of wavelengths. If the compound absorbs light at a particular wavelength, the intensity of the sample beam  $(I_S)$  will be less than that of the reference beam  $(I_R)$ . The instrument indicates this by producing a graph, a plot of wavelength of the entire region *versus* the absorbance (A) of light at each wavelength. Such a graph is called an **absorption spectrum**.

A typical UV absorption spectrum of 2,5-dimethyl-2,4-hexadiene is given in Fig. 24.2. It shows a broad absorption band in the region between 210 and 260 nm. The absorption is maximum at 242.5 nm. It is this wavelength that is usually reported as  $\lambda_{\rm max}$ .

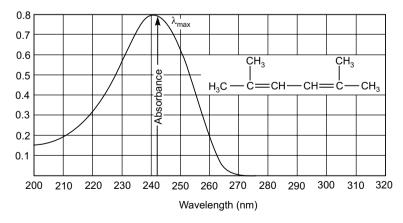


Fig. 24.2 The UV absorption spectrum of 2,5-dimethyl-2,4-hexadiene

The intensity of absorption is expressed as transmittance (T), which is expressed as

$$T = I/I_0$$

where  $I_0$  is intensity of radiant energy striking the sample.

*I* is the intensity of the radiation emerging from the sample.

However, it is convenient to express the intensity of absorption using Lambert-Beer Law, which is represented as

$$A = \log_{10} \left( I_0 / I \right) = Kcb$$

where A is absorbance.

*K* is a constant characteristic of the solute

c is the concentration of the solute

b is the path length through the sample.

In case, c is expressed as mole/litre and b in centimeters, the above expression becomes

$$A = \varepsilon cb$$

 $\varepsilon$  is the molar absorptivity.

However, if c is in g/litre, the equation becomes

$$A = abc$$

where a is the absorptivity.

The intensity of an absorption band in the ultraviolet spectrum is expressed as

$$E_{1\,\mathrm{cm}}^{1\%} = \frac{A}{cb}$$

where c is the concentration in g/100 mL.

b is the path length through the sample in centimeters.

#### 24.2 TERMS USED IN UV SPECTROSCOPY

Following terms are commonly used in UV spectroscopy:

- Chromophore: A chromophore is a covalently unsaturated group that is responsible for the absorption of radiation. Examples, include C=C, C = O and NO<sub>2</sub>.
- 2. Auxochrome: It is a saturated group (e.g., OH, NH<sub>2</sub>, SH, :Br, etc.) with non-bunded electrons. When attached to a chromophore, it may alter the intensity of wavelength of absorption.
- 3. Bathochromic Shift: When the  $\lambda_{max}$  of a compound shifts to a longer wavelength, the compound is said to have undergone a bathochromic shift. This may be due to conjugation or solvent effect. It is also known as a red shift.

- **4. Hypsochromic Shift:** When  $\lambda_{\text{max}}$  of a compound shift to a shorter wavelength, the compound is said to have undergone a hypsochromic shift. It may be due to substitution or solvent effect acid is also known as blue shift.
- 5. Hyperchromic Effect: It is the increase in absorption intensity.
- **6. Hypsochromic Effect:** It is the decrease in absorption intensity.
- 7. **B-Bands (Benzenoid Bands):** These are characteristic of aromatic and heteroaromatic molecules.
- **8.** E-Bands: These bands are characteristic of aromatic structures.

# 24.3 ELECTRONIC TRANSITIONS

The total energy of a molecule is the sum of its electronic energy, vibrational energy and rotational energy. The magnitude of these energies decrease in the order:  $E_{\rm elect}$ ,  $E_{\rm vib}$  and  $E_{\rm rot}$ . In a molecule, the energy absorbed in the UV region produces changes in the electronic energy from transition of valence electrons in the molecule. These transitions consist of excitation of an electron from an occupied molecular orbital (usually bonding  $\pi$  orbital) to the next higher energy orbital (an antibonding  $\pi^*$  or  $\sigma^*$  orbital). The anitbonding orbital is designated by an asterisk. Thus, promotion at an electron from  $\pi$ -bonding orbital to an antibonding ( $\pi^*$ ) orbital is indicated as  $\pi \to \pi^*$ .

The relationship between the energy absorbed in an electronic transition and the frequency (v), wavelength  $(\lambda)$ , and wave number  $(\bar{v})$  of radiation producing the transition is given by the expression.

$$\Delta E = hv = \frac{hc}{\lambda} = h\overline{v}c$$

Where  $\Delta E$  is the energy absorbed in an electronic transition from a low energy state (ground state) to a high energy state (excited state), h is the Plank's constant and c is the velocity of light. The relative energies of important orbitals are shown in Fig. 24.3

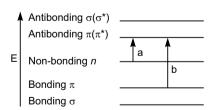


Fig. 24.3 Designation of electronic energy levels. The antibonding  $\pi$  and  $\sigma$  are represented as  $\pi^*$  and  $\sigma^*$ , respectively

The electronic transitions of concern to an organic chemist are  $n \rightarrow \pi^*$  and  $\pi \rightarrow \pi^*$  (represented as a and b, respectively, in Fig. 24.3). In  $n \rightarrow \pi^*$  transition, *one* 

of the electrons of an unshared pair goes to an unstable (antibonding)  $\pi^*$  orbital. For example,

$$C=\ddot{O}: \longrightarrow C \doteq \dot{O}: \quad n \longrightarrow \pi^*$$

The  $n \rightarrow \pi^*$  transition is also referred to as the K-band. In the  $\pi \rightarrow \pi^*$  transition, an electron goes from a stable (bonding)  $\pi$  orbital to an unstable (antibonding)  $\pi^*$  orbital. For example,

$$C = \ddot{O}: \longrightarrow C : \ddot{O}: \quad \pi \longrightarrow \pi^*$$

The  $\pi \to \pi^*$  transition is also referred to as the K-band.

The  $n\to\pi^*$  transition (Fig. 24.3) requires less energy compared to the  $\pi\to\pi^*$  transition. The  $\pi\to\pi^*$  transition can occur in a simple alkene, like ethylene, but the absorption occurs in the far ultraviolet. The conjugation of double bonds lowers the energy required for the electronic transition and the absorption moves to longer wavelength. In case there are enough double bonds in conjugation, absorption will move into the visible region and the compound will be coloured. For example,  $\beta$ -carotene is a yellow pigment found in carrots and green leaves and is a precursor of vitamin A; it contains 11 carbon-carbon double bonds in conjugation and owes its colour to absorption at the violet end of the visible spectrum ( $\lambda_{max}$  451 nm).

The electronic transitions of some simple molecules is shown in Table 24.1. The designations of the bands are those used by A. Buraway (Ber, 1930, **63**,3155; J. Chem. Soc., 1939, 1177). The significance of R-bands and E-bands has already been explained. B-Bands (benzenoid bands) are characteristic of aromatic and heteroaromatic molecules. E-bands, like the B-bands are characteristic of aromatic structures.

The  $E_1$ - and  $E_2$ -bands of benzene are observed near 180 nm and 200 nm respectively.

Example	Electronic Transition	λ <sub>max</sub> (nm)	ε <sub>max</sub>	Band
Ethane	σ→σ*	135	_	_
Water	n→σ*	167	7,000	_
Methanol	n→σ*	183	500	_
1-Hexanethiol	n→σ*	224	126	_
<i>n</i> -Butyl iodide	n→σ*	257	486	_
Ethylene	$\pi \rightarrow \pi^*$	165	10,000	_
Acetylene	$\pi \rightarrow \pi^*$	173	6,000	_
Acetone	$\pi \rightarrow \pi^*$	about 150	-	_

Table 24.1 Electronic Transitions of some simple molecules

(Contd...)

Example	Electronic Transition	λ <sub>max</sub> (nm)	ε <sub>max</sub>	Band
	<i>n</i> →σ*	188	1,860	_
	<i>n</i> →π*	279	15	R
1,3-Butadiene	$\pi \rightarrow \pi^*$	217	21,000	К
1,3,5-Hexatriene	$\pi \rightarrow \pi^*$	258	35,000	К
Acrolein	$\pi \rightarrow \pi^*$	210	11,500	К
	<i>n</i> →π*	315	14	R
Benzene	Aromatic π→π*	about 180	60,000	E <sub>1</sub>
	Aromatic π→π*	about 200	8,000	E <sub>2</sub>
	Aromatic π→π*	255	215	В
Styrene	Aromatic π→π*	244	12,000	К
	Aromatic π→π*	282	450	В
Toluene	Aromatic π→π*	208	2,460	E <sub>2</sub>
	Aromatic π→π*	262	174	В
Acetophenone	Aromatic π→π*	240	13,000	К
	Aromatic π→π*	278	1,110	В
	<i>n</i> →π*	319	50	R
Phenol	Aromatic π→π*	210	6,200	E <sub>2</sub>
	Aromatic $\pi \rightarrow \pi^*$	270	1,450	В

#### 24.4 ULTRAVIOLET SPECTROMETER

A UV spectrometer consists of five features. These include a radiation source, sample and reference cells, monochromator, photodetector and amplifier and recorder. A schematic diagram of a UV spectrometer is given in Fig. 24.4.

- (*i*) Radiation Source: A number of radiation sources can be used. These include tungsten-filament lamp [for scanning the visible region (350 nm to about 800 nm)], hydrogen or deuterium discharge tube [for scanning near UV region (185 nm to 390 nm)].
- (ii) Sample and Reference Cells: Two identical cells made of silica or quartz are used. The monochromatic beam, which is split into two parallel beams using a mirror system is passed through one cell containing the solution of the substance to be examined and the other beam is passed through the cell containing pure solvent. Both the cells are placed in a special holder so that two equal beams of UV light are passed through both the cells.

The solvent to be used should be of 'spectrograde'. The common solvents used are hexane, ethanol and cyclohexane, which are transparent down to about 210 nm.

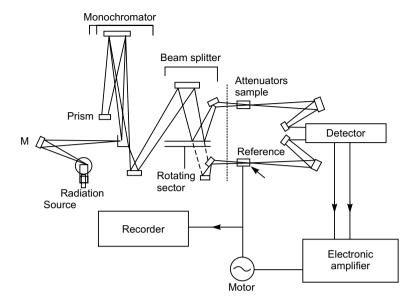


Fig. 24.4 Schematic diagram of a UV spectrometer

A number of solvents are available for use in the UV region. The solvent should be freed of aromatic and olifinic impurities. This is achieved by precolation through activated silica gel and should be transparent down to about 210 nm. A solvent should be selected so that it does not react with the sample. For example, the UV spectra of aldehydes should not be determined in alcohol. The uv cut-off wave lengths ( $\lambda_{max}$  nm) for some common solvents are given below.

Solvent	Cut-off wavelength (λ <sub>max</sub> nm)
Acetonitrile	210
<i>n</i> -Butanol	210
Cyclohexane	210
Ethanol (95%)	210
Methanol	215
Dioxane	225
Chloroform	245
Water	210
Acetone	330

Usually 0.1 to 100 mg of the sample is required for running UV spectrum. The percentage transmittance of the solution should be in the range 20 to 65 per cent.

- 240
- (iii) Monochromator: Silica prisms of different types or different gratings are used as monochromator. It is capable of selecting a narrow band of wavelength.
- (iv) The Detector, Amplifier and Recorder: Photomultiplier or photoelectric tubes are normally used as detector. The dector receives an intense beam from the reference cell and a less intense beam from the sample cell. The generated alternating current flows from the generator to the amplifier and finally to a recorder, which plots an absorption curve relating to wavelength (abscissa) and absorption (ordinate).

# 24.5 CHARACTERISTIC ABSORPTION OF ORGANIC COMPOUNDS

It has already been stated that the ability of an organic compound to absorb UV radiation depends on its electronic structure. Following are given characteristic absoprtion of different types of organic molecules:

# Saturated Hydrocarbons

Saturated hydrocarbons contain only  $\sigma$  electrons. As the energy required to bring about  $\sigma \to \sigma^*$  transition is of the order 185 Kcal per mole is available only in the far (vacuum) ultraviolet region, saturated hydrocarbons are transparent in the near ultraviolet region and so can be used as solvents.

# Compounds Containing $\pi$ -Electrons (Chromophores)

Compounds containing  $\pi$ -electrons may also contain non-bonding electron pairs. Groups containing both  $\pi$ - and *n*-electrons can undergo three transitions:  $n \rightarrow \sigma^*$ ,  $\pi \rightarrow \pi^*$  and  $n \rightarrow \pi^*$ . The absorption characteristics of a number of compounds containing single isolated chromophonic groups are given in Table 24.2.

Group	Structure	Example	Solvent	Absorption band	Transition
				1 E	

 Table 24.2
 Ultraviolet Absorption Bands for some important functional groups

Group	Structure	Example	Solvent		orption and	Transition
				I <sub>max</sub> (nm)	<sup>€</sup> max	
Alkenes	>c=c<	Ethylene	Vapour	165	15,000	$\pi \rightarrow \pi^*$
				193	10,000	$\pi \rightarrow \pi^*$
Alkynes	—C≡C—	Acetylene	Vapour	173	6,000	$\pi \rightarrow \pi^*$
Carbonyl	H   C=O aldehyde	Acetaldehyde	Heptane	290	16	$n \rightarrow \pi^*$

Group	Structure	Example	Solvent	Absoi ba	•	Transition
				I <sub>max</sub> (nm)	E <sub>max</sub>	
	C=0 ketone	Acetone	n-Hexane	180	900	$\pi \rightarrow \pi^*$
				279	15	n → π*
Carboxyl	—СООН	Acetic acid	Water	204	60	n→π*
Amide	-CONH <sub>2</sub>	Acetamide	n-Hexane	< 208	_	n→π*
Esters	—COOR	Ethyl acetate	Petroleum ether	207	69	<i>n</i> →π*
Nitro	$-NO_2$	Nitromethane	Alcohol	271	18.6	n→π*
Nitrate	-ONO <sub>2</sub>	Ethyl nitrate	Dioxane	270	12	n→π*
Nitrite	—ONO	Amyl nitrite	Petroleum ether	218.5	1,120	$\pi \rightarrow \pi^*$
Nitrile	—C≣N	Acetonitrile	Vapour	167	_	$\pi \rightarrow \pi^*$
Azo	—N=N—	Azomethane	Dioxane	347	4.5	<i>n</i> →π*

# **Ethylenic Chromophore**

In case of isolated ethylenic chromophore, there is intense absorptions in the far UV region due to  $\pi \rightarrow \pi^*$  transition. In the vapour phase, ethylene absorbs at 165 nm ( $\epsilon_{max}$  15,000); in this case another band at about 200 nm also occurs due to the elevation of two  $\pi$ -electrons to  $\pi^*$  orbitals (Table 24.2).

Allenes, having the unit C=C=C show a strong absorption band at about 170 nm.

Acyclic conjugated dienes show a intense  $\pi \rightarrow \pi^*$  transition bands (K-bands) in the region 215-230 nm. Thus, 1,3-butadiene shows absorption at 217 nm ( $\epsilon_{max}$  21,000). An empirical rule for predicting, the bathochromic effect of alkyl substitution in 1,3-butadiene has been formulated by Woodward. According to this rule, each

Compound	λ <sub>max</sub> (nm)
Base value for 1,3-butadiene	217
Increment for	
Alkyl substituent or ring residue	+5
Exocyclic double bond	+5
or For double bond exocyclic to two rings	+10

Table 24.3 Woodward rule for absorption for conjugated dienes

alkyl group or ring residue, attached to the parent diene (1,3-butadiene) shifts the absorption 5 nm towards long wavelength region; the presence of exocyclic double bond causes additional bathochromic shift of 5 nm; the shift is 10 nm in case the double bond is exocyclic to two rings (Table 24.3).

On the basis of the above rule,  $\lambda_{max}$  for 2,3-dimethyl-1,3-butadiene is calculated as  $217 + (2 \times 5) = 227$  nm. The observed  $\lambda_{max}$  is 226.

Absorption data for conjugated dienes is given in Table 24.4.

**Table 24.4** Absorption data for  $\pi \rightarrow \pi^*$  transition (K-band) in conjugated dienes

Compound	λ <sub>max</sub> (nm)	ε <sub>max</sub>	Solvent
Butadiene	217	21,000	Hexane
2,3-Dimethyl-1,3-butadiene	226	21,400	Cyclohexane
1,3,5-Hexatriene	253	50,000	Isooctane
	263	52,500	
	274	50,000	
1,3-Cyclohexadiene	256	8,000	Hexane
1,3-Cyclopentadiene	239	3,400	Hexane

In case of alkenes, *cis*- and *trans*-isomers are possible, the *trans*-isomers absorbs at longer wavelength.

For homoannular and heteroannular dienes, the  $\lambda_{max}$  can be calculated by the Fieser's rules (Table 24.5).

Table 24.5 Fieser's rules for homo and heteroannular dienes

Compound	λ <sub>max</sub> (nm)
Parent heteroannular diene	214 (Base value)
Parent homoannular diene	253 (Base value)
Add for	
Double bond extending conjugation	30
Ring residue or alkyl substituent	5
Exocyclic double bond	5
OAc	0
OAlk	6
SAIk	30
Cl, Br	5
N(Alk) <sub>2</sub>	60
Solvent correction	0

On the basis of the above rules, the  $\lambda_{max}$  for cholesta-3, 5-diene and cholesta-2,4-diene is calculated as follows:

Base value for heteroannular diene 3 ring residues 
$$(1, 2, 3)$$
 5 × 3

Exocyclic double bond (one) Total  $234$  Cholesta-3, 5-diene Observed  $\lambda_{max}$  = 235 mm ( $\epsilon_{max}$  19,000).

Base value for heteroannular diene 3 ring residues  $(1, 2, 3)$  3 × 5

Exocyclic double bond (one)  $1 \times 5$  Cholesta-2, 4-diene Observed  $\lambda_{max}$  = 275 nm ( $\epsilon$  10,000).

Fieser's rule (Table 24.5) work well for conjugated systems of four double bonds or less. In case, a conjugated polyene contains more than four double bonds, the Fieser-Kuhn rules are used. According to this rule, both  $\lambda_{max}$  and  $\epsilon_{max}$  are related to the number of conjugated double bonds, as well as other structural units by using the following equation:

$$\lambda_{\text{max}} = 114 + 5 \text{ M} + n (48.0 - 1.7 n) - 16.5 R_{\text{endo}} - 10 R_{\text{exo}}$$

$$\varepsilon_{\text{max}} = (1.74 \times 10^4) n$$

Where

n = number of conjugated double bonds.

M = number of alkyl or alkyl-like substituents on the conjugated system.

 $R_{\text{endo}}$  = number of rings with endocyclic double bonds in the conjugated system.

 $R_{\rm exo}$  = number of rings with exocyclic double bonds.

Using the above rule,  $\lambda_{max}$  for lycopene can be calculated as follows:

$$\lambda_{\text{max}}^{\text{calc.}} = 114 + 5(8) + 11[48.0 - 1.7(11)] - 0 - 0$$

$$= 476 \text{ nm}$$

$$\lambda_{\text{max}}^{\text{obs}} = 474 \text{ nm (hexane)}$$

$$\varepsilon_{\text{max}}^{\text{calc.}} = 1.74 \times 10^4 (11) = 19.1 \times 10^4$$

$$\varepsilon_{\text{max}}^{\text{obs}} = 18.6 \times 10^4 (\text{hexane})$$

# **Acetylenic Chromophore**

Acetylene shows a weak band at 173 nm due to transition. Conjugated polynes exhibit two principal band in the near UV. Typical absorption data for conjucated polynes is given in Table 24.6.

Conjugated Polynes	λ <sub>max</sub> (nm)	<sup>€</sup> max	λ <sub>max</sub> (nm)	<sup>€</sup> max
2,4-Hexadiyne	_	_	227	360
2,4,6—Octatriyne	207	135,000	268	200
2,4,6,8—Decatetrayne	234	281,000	306	180

Table 24.6 Absorption for conjugated polynes

# **Carbonyl Chromophore**

The carbonyl group contains a pair each of  $\sigma$ -electrons and  $\pi$ -electrons and two pairs of nonbonding (n or p) electrons.

**Saturated aldehydes and ketones** give three absorption bands, two of which are in the far UV region. A  $\pi \rightarrow \pi^*$  transition absorbs strongly at about 150 nm and an  $n \rightarrow \sigma^*$  transition absorbs about 190 nm. The third band (*R*-band) is weak ( $\varepsilon_{\text{max}}$  < 30) and appears in the near UV region at 270-300 nm; this band results from the forbidden transition of a loosely held *n*-electron to the  $\pi^*$  orbital. The *R*-band undergoes a blue shift as the polarity of the solvents increased. The absorption data of saturated ketones and aldehydes is given in Table 24.7.

terene [					
Compound	λ <sub>max</sub> (nm)	ε <sub>max</sub>	Solvent		
Acetone	279	13	Isooctane		
	264.5	_	Water		
Methyl ethyl ketone	279	16	Isooctane		
Diisobutyl ketone	288	24	Isooctane		
Hexamethyl acetone	295	20	Alcohol		
Cyclopentanone	299	20	Hexane		
	1	1			

**Table 24.7** Absorption data for saturated aldehydes and ketone  $[n \rightarrow \pi^* \text{ transition (R-band)}]$ 

Compound	(nm) λ <sub>max</sub>	<sup>€</sup> max	Solvent
Cyclohexanone	285	14	Hexane
Acetaldehyde	290	17	Isooctane
Propionaldehyde	292	21	Isooctane
Isobutyraldehyde	290	16	Hexane

In case of aliphatic ketones, the introduction of more highly branched alkyl group results in bathochromic effect (*see* Table 24.7). In case of cyclic ketones (like cyclohexanone), having halogen substitution in the  $\alpha$ -position has a marked effect on the absorption (Table 24.8).

**Table 24.8** Shift in absorption maxima for  $\alpha$ -substituted cyclohexanones

	X	eq	ax
0	—CI	-7	+22
www. X	—Br	-5	+28
	—ОН	-12	+17
	-O <sub>2</sub> CCH <sub>3</sub>	<b>-</b> 5	+10

As seen (Table 24.8), the presence of halogen atoms in saturated cyclic ketones has marked effect on the absorption (Table 24.8).  $\lambda_{max}$  of the parent compound is reduced by 5-10 nm when the substituent is equatorial. In case the substituent is axial, a bathochromic shift of 10-30 nm is noticed. These findings are useful in the structure elucidation of halogenated steroids and terpenes.

It is found that the attachment of groups containing lone electron pairs to carbonyl groups has a profound effect on the  $n\rightarrow\pi^*$  transition. The *R*-band is shifted to shorter wavelength. Absorption values for the  $n\rightarrow\pi^*$  transitions of a number of simple carbonyl campounds is given in Table 24.9.

**Table 24.9**  $n \rightarrow \pi^*$  Transition (R-bands) of simple carbonyl containing compounds

Compound	λ <sub>max</sub> (nm)	€ <sub>max</sub>	Solvent
Acetaldehyde	293	11.8	Hexane
Acetic acid	204	41	Ethanol
Ethyl acetate	207	69	Pet. ether
Acetamide	220(s)	_	Water
Acetyl chloride	235	53	Hexane
Acetic anhydride	225	47	Isooctane
Acetone	279	15	Hexane

#### **Diketones**

 $\alpha$ -Diketones show a weak *R*-band at about 275 nm and a weak band near 450 nm due to interaction between the carbonyl groups. Following are given some of the  $\alpha$ -diketones with their  $\lambda_{max}$  value:

The UV spectra of  $\beta$ -diketones is dependent on the degree of enolization. A typical example of  $\beta$ -diketone is acetyl acetone. The enolic species exist to the extent of about 15% in aqueous solution and about 90% in the vapour phase or in non-polar solvents.

H<sub>3</sub>C—C—CH<sub>2</sub>—C—CH<sub>3</sub>
Acetyl acetone Keto

$$\lambda \frac{H_2O}{max} 274 \text{ mm}, \quad \varepsilon_{max} 2050$$

$$\lambda \frac{\text{iscooctane}}{max} 272 \text{ mm}, \quad \varepsilon_{max} 12,000$$

Cyclic  $\beta$ -diketones like 1,3-cyclohexanedione, exist almost exclusively in enolic form even in polar solvents. It shows strong absorption in the region 230-260 nm due to  $\pi \rightarrow \pi^*$  transition.

O HO enol

$$\lambda \operatorname{EtOH}_{\text{max}} 253 \operatorname{mm} (\varepsilon_{\text{max}} 22,000)$$

$$\lambda \operatorname{NaOH}_{\text{max}} 270-300 \operatorname{nm}$$

# α,β-unsaturated Ketones and Aldehydes

Compounds containing a carbonyl group in conjugation with an ethylene groups are called **enones.** Their spectra is characterised by an intense absorption band (*K*-band) in the region 215-250 nm ( $\varepsilon_{\text{max}}$  10,000–20,000) and a weak *R*-band at

310-330 nm, which is poorly defined. The absorption of a number of conjugated aldehydes and ketones is given in Table 24.10.

Compound	K-b	and	R-band		Solvent
	λ <sub>max</sub> (nm)	log <sup>E</sup> max	λ <sub>max</sub> (nm)	log ε <sub>max</sub>	Solven
Methyl vinyl ketone	212.5	3.85	320	1.32	Ethanol
Methyl isopropenyl	218	3.90	315	1.4	Ethanol
Ketone					
Acrolein	210	4.06	315	1.41	Water
Crotonaldehyde	220	4.17	322	1.45	Ethanol
Crotonaldehyde	214	4.20	329	1.39	Isooctane
			341	1.38	
			352(s)	1.25	

Table 24.10 Absorption for conjugated aldehydes and ketones

The intensity of *K*-band is considerably reduced due to steric hinderance which prevents coplanarity as is observed in the example given below:

$$\lambda_{\text{max}} \text{ 243 nm } (\epsilon_{\text{max}} \text{ 1400})$$

1-Acetyl-2-methyl cyclohexene

Effect of substitution on the position of  $\pi \rightarrow \pi^*$  transition (*K*-band), in case of  $\alpha,\beta$ -unsaturated ketones has been generalised by Woodward (Table 24.11).

$$R = C = C = C$$

Table 24.11 Woodward Rule for  $\lambda_{\text{max}}$  in case of  $\alpha,\beta\text{-unsaturated}$  ketones

	λ <sub>max</sub> (nm)	
Unsubstituted		215
$\alpha$ or $\beta$	No exocyclic C = C	225
αβ or ββ	No exocyclic C = C	235
αβ or ββ	One exocyclic C = C	240
αββ	No exocyclic $C = C$	247
αββ	One exocyclic $C = C$	252

The spectra  $\alpha,\beta$ -unsaturated aldehydes (Table 24.12) are similar to those of  $\alpha,\beta$ -unsaturated ketones.

<b>Table 24.12</b>	Maxima (	of unsaturated	aldehydes
--------------------	----------	----------------	-----------

Substitution	$\lambda_{max}(nm)^1$
Unsubstituted	208
$\alpha$ or $\beta$	220
αβ or ββ	230
αββ	242

1 exocyclic double bond + 5.

Similar rules apply to the cyclopentenone system

Parent system 214 nm (Value calculated from data in Table 25.13)  $\alpha$  or  $\beta$  substitution 224 nm  $\alpha$ ,  $\beta$  substitution 236 nm

Extensive rules of enone absorption have been given by Fieser (Table 24.13).

**Table 24.13** Woodward empirical rules for enone and dienone absorption  $(\alpha, \beta$ -unsaturated carbonyls of ketones)

(Enone) (Dienone)	λ <sub>max</sub> (nm)
Acyclic α,β-unsaturated ketone	215
α,β-unsaturated aldehydes	210
α,β-unsaturated acids and esters	195
Six-membered cyclic α,β-unsaturated ketones	215
Five-membered cyclic α,β-unsaturated ketones	202
Add for	
Double bond extending conjugation	30
Alkyl groups or ring residue $\alpha$	10
β	12
γ, δ	18
—OAc $\alpha$ , $\beta$ and $\delta$	6
—OH α	35
β	30
γ	50

β 	none) a     =C—C=O	( <b>Dienone)</b> δ    γ    β    α	λ <sub>max</sub> (nm)
—OCH <sub>3</sub>	α		35
	β		30
	γ		17
	δ		31
—CI	α		15
	β		12
—Br	α		25
	β		30
-NR <sub>2</sub>	β		95
Exocyclic do	uble bond		5
Homodiene d	component		39
Solvent corre	ection (See Table 2	24.14)	variable

Table 24.14 Solvent Correction

Solvent	Correction (nm)
Ethanol	0
Methanol	0
Dioxane	+5
Chloroform	+1
Ether	+7
Water	-8
Hexane	+11
Cyclohexane	+11

Using the above rules (Table 24.13),  $\lambda_{max}$  of some of the compounds can be calculated as shown below:

Diosphenol Enol of 1,2-cyclopentanedione calc. 
$$\lambda_{\text{max}}^{\text{EIOH}}$$
 215 (base) calc.  $\lambda_{\text{max}}^{\text{EIOH}}$  202 (base) 12 ( $\beta$ -subst.) 35 ( $\alpha$ -subst.) 35 ( $\alpha$ -subst.) 35 ( $\alpha$ -subst.) obs.  $\lambda_{\text{max}}^{\text{EIOH}}$  270 obs.  $\lambda_{\text{max}}^{\text{EIOH}}$  = 247

### **Carboxylic Acids**

**Saturated carboxylic acids** give a weak band at about 200 nm resulting from forbidden  $n-\pi^*$  transition. This band is of little diagnostic value.

 $\alpha,\beta$ -unsaturated carboxylic acids show a strong K-band characteristic of conjugated system. Acrylic acid, the first member of the series exhibits absorption near 200 nm ( $\epsilon_{max}$  10,000). As in this case of  $\alpha,\beta$ -unsaturated ketones, in case of  $\alpha,\beta$ -unsaturated carboxylic acids also there is a bathochromic shift of the K-band due to alkyl substitution in the basic structure (of acrylic acid).  $\lambda_{max}$  for unsaturated carboxylic acids can be calculated by using the data given in Table 24.15. The absorption maxima of a number of  $\alpha,\beta$ -unsaturated carboxylic acids is given in Table 24.15 and Table 24.16.

Table 24.15  $\lambda_{\text{max}}$  values of unsaturated carboxylic acids and esters

Substitution pattern	EtOH λ <sub>max</sub> (± 5 nm)
$\alpha$ or β-monosubstituted	208
$\alpha$ , $\beta$ or $\beta$ , $\beta$ -disubstituted	217
α, β, β-trisubstituted	225
exocyclic $\alpha$ , $\beta$ -double bond	+5
endocyclic α, β-double bond	+5
in 5 or 7 membered ring	
Data from Table 24.13 can be used here.	

**Table 24.16** Absorption of  $\alpha,\beta$ -unsaturated acids

Compound	λ EtOH <sub>(nm)</sub> max	<sup>€</sup> max
CH <sub>2</sub> = CH—COOH	200	10,000
CH <sub>3</sub> CH = CH COOH (trans)	205	14,000
(cis)	205.5	13,500

(Contd...)

Compound	λ EtOH <sub>(nm)</sub> max	<sup>€</sup> max	
CH <sub>3</sub>	210	_	
CH <sub>3</sub> —(CH=CH) <sub>2</sub> —COOH	254	25,000	
CH <sub>3</sub> —(CH=CH) <sub>3</sub> —COOH	294	37,000	
CH <sub>3</sub> —(CH=CH) <sub>4</sub> —COOH	332	49,000	

Sodium salts of carboxylic acids and esters exhibit absorptions at the same wavelengths as the parent acid. Unsaturated conjugated lactones give absorptions similar to unsaturated esters. Lactones give absorption is the region 200-240 nm. However, extended conjugation exhibit a bathochrome shift of  $\pi \rightarrow \pi^*$  transition (K band). The  $\lambda_{max}$  of such compound can be predicted by using the data given in Table 24.13.

### Oximes and Imines

Conjugated oximes and imines absorb in the region 220-230 nm ( $\varepsilon_{\text{max}} > 10,000$ ) due to  $\pi \rightarrow \pi^*$  transition (K-band). In presence of acids, imines produce positive charge on N, thereby shifting the absorption in the region 270-290 nm. Simple imines show a weak  $n \rightarrow \pi^*$  Thus,

$$\begin{array}{ll} \lambda_{max}^{cyclohexane} &=\; 246\; nm\; (\epsilon=140)\; and \; \lambda_{max}^{EtOH} = 233\; nm\; (\epsilon=200)\\ & \quad in\; the\; case\; of\; CH_{2}CH_{2}N = C(CH_{3})_{2}. \end{array}$$

#### **Nitriles**

The  $\alpha$ ,  $\beta$ -unsaturated nitriles absorb just inside the UV region near 213 nm ( $\epsilon_{max}$  10,000).

## **Azo Group**

In the azo group ( $-\dot{N}=\dot{N}$ ) the ethylenic linkage with two  $\sigma$  bonds is replaced by two lone pair of electrons. In this case, the  $\pi\to\pi^*$  transition occurs in the far UV region. Thus, *trans*-azobenzene absorbs at 320 nm ( $\epsilon_{max}$  21,000) [compare absorption for *trans*-stillbene,  $\lambda_{max}$  295 nm ( $\epsilon_{max}$  28,000)].

### Nitro, Nitroso, Nitrates and Nitrites

All these groups contain multiple nitrogen to oxygen linkages. These exhibit weak absorption in the near UV region due to  $n\rightarrow\pi^*$  transition. (Table 24.16A)

Compound	n→π∗ Transition (R-band)				
	λ <sub>max</sub> (nm)	<sup>€</sup> max	Solvent		
Nitromethane	275	15	Heptane		
2-Methyl-2-nitropropane	280.5	23	Heptane		
1-Nitro-1-propene	229 ( <i>n</i> →π*)	9400	Ethanol		
	235 ( <i>n</i> →π*)	9800	Ethanol		
Nitrobutane	300 665	100 20	Ether		
Octylnitrate	270	15	Pentane		
	(inflexion)				
Cyclohexyl nitrate	270 (inflexion)	22	Pentane		
<i>n</i> -Butylnitrite	218 313-384	1050 17-45	Ethanol		

Table 24.16(a) Absorption of compounds containing N—O linkages

### **Compounds Containing Sulphur**

Sulfoxides (saturated) show bands at 220 nm ( $\epsilon$  1500)  $n\rightarrow\pi^*$  transition. Aromatic sulfoxides give an intense *K*-band in addition to displaced *K*-band. The UV absorption of some thiocarbonyl compounds is given in Table 24.16(*b*).

Compound	n→π* transition (K-band)			
	λ <sub>max</sub> (nm)	log ε <sub>max</sub>		
Thiobenzophenone	599	2.81		
Thioacetamide	358	1.25		
Thiourea	291 (Shoulder) 1.85			

**Table 24.16(b)**  $n \rightarrow \pi^*$  transition of thiocarbonyl compounds

## **Benzene Chromophore**

Benzene shows three absorption bands at 184 nm  $(E_1)$  ( $\varepsilon_{\rm max}$  60,000); 204 nm  $(E_2)$  ( $\varepsilon_{\rm max}$  7900); and 256 nm (B) ( $\varepsilon_{\rm max}$  200); all these bands originate from  $\pi \to \pi^*$  transitions. The first band (184 nm) results from allowed transitions while the other two bands (near 200 and 260 nm) results from forbidden transitions in the highly symmetrical benzenes and are weak bands.

Alkyl group substitution on benzene ring produces a bathochromic shift of the *B* band. The absorption data for alkyl benzenes is given in Table 24.17.

Compound	λ <sub>max</sub> (nm) (B band)	<sup>€</sup> max
Benzene	256	200
Toluene	261	300
<i>m</i> -Xylene	262.5	300
1,3,5-Trimethyl benzene	266	305
Hexamethyl benzene	272	300

Table 24.17 Absorption data for alkyl benzenes

Substitution on benzene ring by auxochromic groups (OH, NH<sub>2</sub>, etc.) shifts *E*- and *B*-bands to longer wavelength due to n- $\pi$  conjugation (Table 24.8).

Table 24.18 Effect of auxochromic substitution on the spectrum of benzene

Compound	E <sub>2</sub> -band		B-band		Solvent
	λ <sub>max</sub> (nm)	E <sub>max</sub>	λ <sub>max</sub> (nm)	<sup>€</sup> max	
Benzene	204	7,900	256	200	Hexane
Chlorobenzene	210	7,600	265	240	Ethanol
Thiophenol	236	10,000	269	700	Hexane
Anisole	217	6,400	269	1,480	2% Methanol
Phenol	210.5	6,200	270	1,450	Water
Phenolate anion	235	9,400	287	2,600	Aq. Alkali
o-Catechol	214	6,300	276	2,300	Water (pH 3)
o-Catecholate anion	236.5	6,800	292	3,500	Water (pH 11)
Aniline	230	8,600	280	1,430	Water
Anilinium cation	203	7,500	254	160	Aq. acid
Diphenyl ether	255	11,000	272	2,000	Cyclohexane
			278	1,800	_

As seen (Table 24.18), conversion of phenol into phenolate anion results in a bathochromic shift of  $E_2$ – and B-bands with increase in  $\varepsilon_{\rm max}$ . This is attributed to the availability of nonbonding electrons (in the anion) for interaction with the  $\pi$  electron system of the ring. Similarly, conversion of aniline into anilium cation, the nonbonding electrons of aniline are not available for interaction with  $\pi$  electrons of the ring; in this case the spectrum is more or less identical with that of benzene.

The presence of a bulky substituent in the ortho position (as in the case of N,N-dimethyl aniline causes a hypsochromic shift in the  $E_2$  band and is accompanied by reduction in  $\varepsilon_{\max}$ . Thus,

N,N-Dimethylaniline	$\lambda_{max}$ 251 nm	$\epsilon_{max}$	15,500
2-Methyl-N,N-dimethyl	$\lambda_{max}$ 248 nm	$\epsilon_{ m max}$	6360
anilene			

In case, an unsaturated group (chromophore) is directly attached to the benzene ring, there is bathochromic shift of the *B* band accompanied by appearance of a *K*-band ( $\varepsilon_{\text{max}} > 10,000$ ) in the region 200-250 nm.

Table 24.19 gives the absorption data of certain structures such as acetophenone and benzaldehyde; in these cases the displaced *B*- and *R*-bands can be recognised.

**Table 24.19** Absorption characteristics of benzenes substituted with chromophores

Compound	$\pi{ o}\pi^*$ Transition K-band		B-band		n→π* Transition R-band		Solvent
Compound	λ <sub>max</sub> (nm)	ε <sub>max</sub>	λ <sub>max</sub> (nm)	ε <sub>max</sub>	λ <sub>max</sub> (nm)	ε <sub>max</sub>	
Benzene	_	_	255	215	_	_	Alcohol
Styrene	244	12,000	282	450	_	-	Alcohol
Phenylacetylene	236	12,500	278	650	_	_	Hexane
Benzaldehyde	244	15,000	280	1,500	328	20	Alcohol
Acetophenone	240	13,000	278	1,100	319	50	Alcohol
Nitrobenzene	252	10,000	280	1,000	330	125	Hexane
Benzoic acid	230	10,000	270	800	_	_	Water
Benzonitrile	224	13,000	271	1,000	_	_	Water
Diphenyl sulphoxide	232	14,000	262	2,400	_	_	Alcohol
Phenyl methyl sulphone	217	6,700	264	977	_	_	_
Benzophenone	252	20,000	_	_	325	180	Alcohol
Biphenyl	246	20,000	Submerged	_	_	_	Alcohol
Stilbene (cis)	283	12,300*	Submerged		_	-	Alcohol
Stilbene (trans)	295 <sup>@</sup>	25,000*	Submerged		_	-	Alcohol
1-Phenyl-1,3-butadiene							
cis-	268	18,500	_	_	_	_	Isooctane
trans-	280	27,000	_	_	_	_	Isooctane
1,3-Pentadiene							
cis-	223	22,600	_	_	_	_	Alcohol
trans-	223.5	23,000	_	_	_	_	Alcohol

<sup>@</sup> Intense band of fine structure.

The calculation of the principal band  $(\pi \rightarrow \pi^*)$  transition of substituted benzene derivatives is given in Table 24.20.

<sup>\*</sup> Intense band also occur in the region 200-230 nm.

<b>Table 24.20</b>	Calculation of the Principal Band ( $\pi \rightarrow \pi^*$ transition) of substituted
	benzene derivatives (Woodward Empirical Rules)

Substituted benzene derivatives Ar-COG		λ EtOH max
Parent chromophore Ar=C <sub>6</sub> H <sub>5</sub>		
G = Alkyl or ring' residue (e.g., ArCOR)		246
G = H (e.g., Ar CHO)		250
G = OH, O Alk (e.g., ArCO2H, ArCO2R)		230
Increment for each substituent or Ar		
Alkyl or ring residue	o, m	+3
	p	+ 10
—OH, OCH <sub>3</sub> , —OAlk	o, m	+7
	p	+25
CI	o, m	+0
	p	+10
Br	o, m	+2
	p	+15
NH <sub>2</sub>	o, m	+13
	p	+58
NHCOCH <sub>3</sub>	o, m	+20
	p	+45
NHCH <sub>3</sub>	р	+73
N(CH <sub>3</sub> ) <sub>2</sub>	o, m	+20
	р	+85

Using the data given in Table 24.20,  $\lambda_{max}$  can be calculated for substituted benzene derivative. Thus, as an example  $\lambda_{max}$  for 6-methoxytetralone can be calculated as follows:

Calc. 
$$\lambda_{\text{max}}^{\text{EtoH}} = 246 \text{ (Parent chromophore)}$$

$$+ 3 \text{ ($o$-ring residue)}$$

$$\text{Total} = \frac{+25 \text{ ($p$-OM}_e$)}{274}$$
Obs.  $\lambda_{\text{max}}^{\text{EtoH}} = 276$ 
6-Methoxytetralone

In case auxochromic groups are on the same ring as the chromophore, both groups influence the absorption. The influence is more pronounced when electron donating group and electron attracting groups are *para* to one another (Table 24.21).

Compound	$\pi { ightarrow} \pi^*$ Transition K-band $\lambda {{EtOH}\over {max}}$		B-b	pand
	λ <sub>max</sub> ε <sub>max</sub>		λ <sub>max</sub>	<sup>€</sup> max
Nitrophenols				
0-	279	6,600	351	3,200
m-	274	6,000	333	1,960
p-	318	10,000	Submerged	Sumerged
Nitroanilines				
0-	283	5,400	412	4,500
m-	280	4,800	358	1,450
p-	381	13,500	Submerged	Submerged

Table 24.21 Absorption data for disubstituted benzenes

### **Biphenyls**

In biphenyls, the two aromatic rings are in conjugation. In this case, the resonance energy is maximum when the two rings are coplaner and basically zero when the two rings are at  $90^{\circ}$  to one another. This is seen by the absorption data of biphenyl and 2,2'-dimethylbiphenyl whose absorption is similar to that of o-xylene.

In the case of diphenylmethane, the conjugation is destroyed and the absorption changes in comparisation to disphenyl

$$\lambda_{\text{max}} = 262 \text{ nm}$$
 
$$\epsilon_{\text{max}} = 5000 \text{ nm}$$
 
$$\lambda_{\text{max}} = 5000 \text{ nm}$$
 
$$\lambda_{\text{max}} = 252 \text{ nm}$$
 
$$\epsilon_{\text{max}} = 252 \text{ nm}$$
 
$$\epsilon_{\text{max}} = 19,000$$

In some substituted dimethyl methanes, *e.g.*, 4-nitro-4' methoxydiphenylmethane, there is effective overlap of  $\pi$  orbitals of the two rings. This results in homoconjugation. The  $\lambda_{\max}$  of 4-nitro-4' - methoxydiphenyl methane is not the sum of  $\epsilon_{\max}$  of *p*-nitrotoluene and p-methoxytoluene as shown ahead.

In case of substituted benzenes, the prediction of the  $\lambda_{max}$  of the primary band can be made using the data in Table 24.22.

**Table 24.22** Calculation of primary band  $(\pi \rightarrow \pi^* \text{ transition})$  ( $E_2$ -band) of substituted benzene (Base value 203.5 nm)

Substituent	Shift	Substituent	Shift
—CH <sub>3</sub>	3.0	-NH <sub>2</sub>	26.5
—CN	20.5	—NHCOCH₃	38.5
—СНО	46.0	$-NO_2$	65.0
—COCH <sub>3</sub>	42.0	—OH	7.0
—CO <sub>2</sub> H	24.5	<b>—</b> 0 <sup>-</sup>	31.5
—Br	6.5	—OCH₃	13.5
—CI	6.0	,	

Source: C.N.R. Rao, J. Sci. Res. (India), 1958, 17B, 56; Current Sci, 1957, 276.

On the basis of the above data (Table 24.22) the  $\lambda_{\text{max}}$  for toluene, phenol, aniline will be (203.5 + 3.0 = 206.5 nm), (203.5 + 7.0 = 210.5 nm) and (203.5 + 26.5 = 230 nm), same as the observed value given in Table 24.18.

The absorption is also affected by steric effects. A typical example is stilbene. In *cis*-stilbene, steric interference destroys coplanarity as is shown by lower frequency of the 283 nm band compared to the corresponding band (295 nm) in the *trans* isomer.

In case, the two phenyl rings are separated by more C=C groups [Ph-(C=C)<sub>n</sub>-Ph] the absorption moves to longer wave length. Thus, in case n=7, the resulting molecule absorbs in the region 440–465 nm ( $\varepsilon_{max}$  135,000).

In the case of polynuclear aromatic compounds *e.g.*, naphthalene, anthracene), as the number of condensed rings increase, the  $\lambda_{max}$  moves to longer wavelength (Table 24.23).

Compound	bound $E_1$ -band $E_2$ -band		B-band		
	$\lambda_{max}$ ( $\epsilon_{max}$ )	λ <sub>max</sub> (ε <sub>max</sub> )	$\lambda_{max}$ ( $\epsilon_{max}$ )		
Benzene	184 (60,000)	204 (7900)	256 (200)		
Naphthalene	221 (133,000)	286 (9300)	312 (289)		
Anthracene	256 (180,000)	375 (9000)	Submerged		

Table 24.23 Absorption of some polynuclear aromatic compounds

### **Heterocyclic Compounds**

Only the unsaturated heterocyclic compounds exhibit absorption in the near UV region (saturated 5- and 6-membered heterocyclic compounds do not exhibit absorption at wavelength longer than 200 nm).

## **Five-membered Rings**

The  $\lambda_{max}$  of five-membered ring heterocycles can be compared to that of cyclopentadiene, the *cis* and the *trans* forms exhibit absorption at about 200 and 238 nm respectively. The absorption of some five-membered heteroaromatics is given in Table 24.24 in comparison to that of cyclopentadiene.

	Band I		Bna	nd II	Solvent
Compound	λ <sub>max</sub>	ε <sub>max</sub>	λ <sub>max</sub>	ε <sub>max</sub>	
(Cyclopentadiene)	200	10,000	238.5	3,400	Hexane
Furans	200	10,000	252		Cyclohexane
Pyrrole	209	6,730	240	300	Hexane
Thiophene	231	7,100	269.5	1.5	Hexane
Pyrazole	214	3,160	_	_	Ethanol

Table 24.24 UV absorption data of some five-membered heteroaromatics

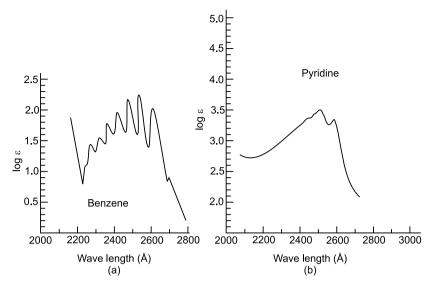
Substitution of the five-membered unsaturated heterocyclics with auxochromic or chromophoric substitution causes a bathochromic shift with increase in the intensity of the bands of the parent compound (Table 24.25).

**Table 24.25** UV absorption data of substituted five membered heteroaromatics

Parent compound	Substituent	Ва	and I	В	and II
compound		<sup>Ն</sup> max <b>(nm)</b>	<sup>€</sup> max	λ <sub>max</sub> (nm)	<sup>€</sup> max
Furan		200	10,000	252	1
Furan	2—CHO	227	2,200	272	13,000
Furan	O    2—C—CH <sub>3</sub>	225	2,300	270	12,900
Furan	2—NO <sub>2</sub>	225	3,400	315	8,100
Furan	2—Br, 5—NO <sub>2</sub>	_	_	315	9,600
Pyrrole		183	_	209	6,730
Pyrrole	2—CHO	252	5,000	290	16,600
Pyrrole	O    2—C—CH <sub>3</sub>	250	4,400	287	16,000
Pyrrole	2—COOH	228	4,500	258	12,600
Pyrrole	O    1—C—CH <sub>3</sub>	234	10,800	288	760
Thiophene		231	7,100	_	_
Thiophene	2—CHO	265	10,500	279	6,500
Thiophene	O    2—C—CH <sub>3</sub>	252	10,500	273	7,200
Thiophene	2—COOH	249	11,500	269	8,200
Thiophene	2—NO <sub>2</sub>	266–272	6,300	294–248	6,000
Thiophene	2—Br	230	9,100	_	_

## Six-Membered Rings

The simplest six-membered heterocycle is pyridine. Its spectrum is similar to that of benzene. In case of pyridine, the B-band is more intense and has less distinct fine structure than that of benzene. A comparison of the UV spectra of benzene and pyridine is given in Fig. 24.5.



**Fig. 24.5** UV spectra of Benzene (a) (Solvent cyclohexane) and Pyridine (b) (Solvent 95% ethane)

The UV absorption maxima of 2-methyl pyridine (Picoline) is given in Table 24.26.

Solvent	λ <sub>max</sub> (nm)	<sup>€</sup> max
Hexane	260	2000
Chloroform	263	4500
Ethanol	260	4000
Water	260	4000
Ethanol-HCl (1:1)	262	5200

Table 24.26 UV absorption maxima of 2-methyl Pyridine

The UV absorption data of pyridine derivatives is given in Table 24.27.

Table 24.27 UV absorption data of substituted pyridine

Substitutions	λ <sup>(PH &gt; 7)</sup> λmax	<sup>€</sup> max
Pridine	257	2,750
	270	450
2-CH <sub>3</sub>	262	3,500
3-CH <sub>3</sub>	263	3,110
4-CH <sub>3</sub>	255	2,100

(Contd....)

Substitutions	λ <sup>(PH &gt; 7)</sup> max	<sup>€</sup> max
2-F	257	3,350
2-Cl	263	3,650
2-Br	265	3,750
2-1	272	400
2-OH	230	10,000
	295	6,300
4-OH	239	14,100
3-OH	260	2,200

The UV spectra of heteroaromatic is found to be related to their isocyclic analogs (Table 24.28).

**Table 24.28** UV absorption maxima of some hetroaromatics containing N and their isocyclic analogs

Compound	E <sub>1</sub> -band		E <sub>2</sub> -band		B-band		Solution
	<sup>Ն</sup> max (nm)	E <sub>max</sub>	λ <sub>max</sub> (nm)	E <sub>max</sub>	λ <sub>max</sub> (nm)	<sup>€</sup> max	Solution
Benzene	184	60,000	204	7,900	256	200	_
Naphthalene	221	100,000	286	9,300	312	280	_
Quinoline	228	40,000	270	3,162	315	2,500	(1)
Isoquinoline	218	63,000	265	4,170	313	1,800	(1)
Anthracene	256	180,000	375	9,000	_	_	_
Acridine	250	200,000	358	10,000	_	_	(2)

(1) Cyclohexane; (2) Ethanol.

### 24.6 INTERPRETATION OF UV SPECTRA

The UV spectrum of the unknown organic compound is taken and the absorption bands compared with those given in Section 24.5.

### 24.7 APPLICATIONS OF UV SPECTROSCOPY

UV spectroscopy alone does not often give a great deal of information, though some generalization can be made, which will be more meaningful when combined with IR spectral data. However, in the absence of IR spectral data following applications are of use:

(i) Absence of certain groups in organic compounds. If an organic compound exhibits no  $\lambda_{max}$  above 200 nm, it cannot have a conjugated

chromophore (e.g., C=C—C=); an aldehyde or keto group or a benzene ring. However, the compound may possess an isolated double or triple bond, a carboxyl group, cyano group or a hydroxy group. UV absorption bands for some important functional group are given in Table 24.2.

(ii) Determination of the concentration of a compound in solution. The concentration of a compound in solution can be determined by using Lambert-Beers law

$$A = \varepsilon \times c \times l$$

where A is observed absorbance at a particular wavelength  $(\lambda)$ , c is the molar concentration of the sample and l is the length in centimeter of the path of the light beam through the sample cell. The procedure consists in measuring the absorbance (A) by using a UV-vis spectrophotometer of various concentration (c) of the solution. A graph is finally plotted between A versus c. The absorbance by the spectrophotometer and its concentration (c) then determined by extrapolation from the graph.

(iii) Determination of the per cent content of vitamin  $A_1(I)$  and vitamin  $A_2(II)$  in natural fats and oils by measurement of the intensities of peaks of 325 nm (five conjugated double bonds) and at 351 nm (six conjugated double bonds), respectively. Comparison of the intensities with those of solution of the two vitamins (I and II) of known concentration enables the concentrations to be calculated.

Vitamin A<sub>1</sub>, (I); 
$$\lambda_{max} = 325 \text{ nm}$$

Vitamin A<sub>2</sub>, (II);  $\lambda_{max} = 351 \text{ nm}$ 

(iv) Differentiation of *cis* and *trans* isomers—when an alkene is capable of exhibiting geometrical isomerism, the *trans* isomer displays longer wavelength absorption and higher intensity than the corresponding *cis* isomer. Thus, in case of stilbene the  $\lambda_{max}$  and  $\varepsilon_{max}$  for *cis* and *trans* form are as given below:

(v) **Keto-Enol Tautomerism:** β-keto esters and β-diketones are known to exist as tautomeric mixtures. Their UV spectra is expected to exhibit absorptions characteristic of both keto and enol forms. Thus, in case of ethylacetoacetate. The keto form exhibits a low intensity band around 275 nm (ε about 20) due to an isolated keto-carbonyl group. The enol exhibits a high intensity band around 245 nm (ε about 18000) due to conjugated double bond.

In a similar way, acetyl acetone ( $\beta$ -diketone) enolizes (84%) in ethanol and absorbs strongly at about 273 nm ( $\epsilon$  10500). This high value of  $\lambda_{max}$  is probably due to chelation in the enol form

The absorption maxima and the percentage of the enol form of ethyl acetoacetate and acetyl actone in different solvents are given below.

Ethyl acetoacetate			Ace	etyl aceto	ne
λ <sub>max</sub> (nm)	ε <sub>max</sub>	% Enol	λ <sub>max</sub> (nm)	ε <sub>max</sub>	% Enol
246 244	1900 8100	12 51	273 269	10500 12100	84 96
	λ <sub>max</sub> (nm)	$\frac{\lambda_{max}}{(nm)}$ $\frac{\varepsilon_{max}}{1900}$	$\frac{\lambda_{max}}{(nm)}$ $\frac{\varepsilon_{max}}{1900}$ $\frac{\%}{12}$	$\lambda_{max}$ (nm) $\varepsilon_{max}$ % Enol $\lambda_{max}$ (nm) 246 1900 12 273	$\lambda_{max}$ $(nm)$ $\varepsilon_{max}$ $\%$ $Enol$ $\lambda_{max}$ $\varepsilon_{max}$ $(nm)$ $\varepsilon_{max}$

Cyclic  $\beta$ -diketones like 1,3-cyclohexanedione, exist almost exclusively in the enolic form even in polar solvents. It shows strong absorption in the region 230-260 due to  $\pi \rightarrow \pi^*$  transition

O HO enol
$$\lambda \underset{\text{max}}{\text{EtOH}} 253 \text{ mm} (\varepsilon_{\text{max}} 22,000)$$

$$\lambda \underset{\text{max}}{\text{NaOH}} 270-300 \text{ nm}$$

- 264
- (vi) Conformational Analysis: The UV spectroscopy can be used for conformational analysis. As an example cyclohexanol having a α-substituted halogen (or similar electron acceptor group) in axial position leads to a bathochromic shift.
- (vii) Determination of Configuration: UV spectroscopy is helpful for the determination of configuration. As an example, in conjugated systems like biphenyls, and 2,2'-dimethyl biphenyl exhibit  $\lambda_{max}$  252 nm ( $\epsilon_{max}$  19000) and 224 nm ( $\epsilon_{max}$  700) respectively. The hypsochromic shift and decrease in extinction coefficient in the case of 2,2'-dimethylbiphenyl is attributed to steric inhibition of resonance indicating non-planar configuration.
- (viii) Study of Charge-transfer Complexes: Charge-transfer complexes are formed when an electron rich molecule (e.g. 2,4-6-trinitrophenol, tetrachloro p-benzoquinone, tetracyano ethylene, iodine, aluminium chloride) is mixed with an electron acceptor molecule (like anthracene, mesitylene, aniline, anisole). The charge-transfer complexes are responsible for new absorption bands in the UV spectra.

A typical example is benzene-iodine charge transfer complex. In this case the  $\pi$ -cloud of benzene interacts with the outer electrons of iodine molecule. The UV absorption data of these are:

$$\lambda_{\rm max}$$
 255 nm  $\lambda_{\rm max}^{\rm hexane}$  500 nm Charge transfer complex  $\lambda_{\rm max}$  290 nm ( $\epsilon$  150000) (additional bond)

Similarly in  $\lambda_{max}$  for aniline-tetracyano ethylene complex gives an additional  $\lambda_{max}$  at 610 nm.



1. Which of the following statements are correct about UV spectrum:

## I. Multiple Choice Questions

	(a) The presence of individual functional	groups as in the case of IR spectroscopy
	(b) Presence of aromatic ring	
	(c) Conjugated system	
	(d) All are correct	
2.	All coloured compounds absorb in the re-	gion:
	(a) Vacuum UV (200 nm to 10 nm)	
	(b) Near UV (200 nm to 380 nm)	
	(c) Visible UV (380 nm to 780 nm)	
	(d) In any of the above region	
3.	Which of the following is a chromophore	::
	(a) C=O	$(b)  \mathrm{NH}_2$
	(c) Br	$(d) NO_2$
4.	Which of the following is a auxochrome:	
	(a) OH	$(b)  \mathrm{NH_2}$
	(c) C==C	$(d) NO_2$
5.	When $\lambda_{\text{max}}$ of a compound shifts to a long have undergone:	er wavelength, the compound is said to
	(a) Bathochromic shift	(b) Hypsochromic shift
	(c) Either (a) or (b)	(d) None of the above
6.	For an organic chemist, important electron	nic transition is/are:
	(a) $n \rightarrow \pi^*$	$(b) \sigma \rightarrow \sigma^*$
	$(c) \pi \rightarrow \pi^*$	$(d) \pi \rightarrow \sigma^*$
7.	The $\pi \rightarrow \pi^*$ transition is also known as:	
	(a) R-band	(b) K-band
	(c) Any of the above	(d) None of the above
8.	Which of the bands are characteristic of a	aromatic structures:
	(a) B-band	(b) E <sub>1</sub> -Band
	$(c)$ $E_2$ -band	(d) None of the above
9.	Groups containing both $\pi$ and $n$ -electron transitions:	as can undergo which of the following
	(a) $n \rightarrow \sigma^*$	$(b) \pi \rightarrow \pi^*$
	(c) $n \rightarrow \pi^*$	(d) None of the above

10.	Which of the following compounds show maximum $\lambda_{max}$			
	(a)	Ethylene	(b) Allene	
	(c)	Butadiene	(d) Ethane	
11.	$\lambda_{max}$ for butadiene is 217 nm and that for 2,3-dimethyl-1,3-butadiene is 226 nm In this case the shift in $\lambda_{max}$ is;			
	(a)	Bathochromic shift	(b) Hypsochromic shift	
	(c)	Can be $(a)$ or $(b)$	(d) None of the above	
12.	Out of cholesta-3,5-diene (I) and cholesta-2,4-diene (II), which one will exhibit maximum $\lambda_{\text{max}}$			
	(a)	I		
	(b)	П		
	(c) Can be I and II depending on the solvent			
	(d) Both I and II will have equal $\lambda_{max}$			
13.	Which of the following will show maximum absorption in the ultraviolet region			
	(a)	2,4-Hexadiyne	(b) 2,4,6-Octatriyne	
	(c) 2	2,4,6,7-Decatetrayne	(d) All will show equal $\lambda_{max}$	
14.	Which type of transitions are normally exhibited by carbonyl chromophore?			
	(a)	$\pi{ ightarrow}\pi^*$	(b) $n \rightarrow \pi^*$	
	(c) ?	$ au{ o}\sigma^*$	$(d) \sigma \rightarrow \sigma^*$	
15.	The introduction of axial halogen in the $\alpha$ -position of cyclohexanone results in			
	(a)	Hypochromic shift	(b) Bathochromic shift	
		Can be $(a)$ or $(b)$	(d) There is no change in $\lambda_{max}$	
16.	Which of the following compounds will exhibit maximum $\lambda_{max}$ ( $n-\pi^*$ transition)			
	(a) .	Acetaldehyde	(b) Acetic acid	
	(c) A	Acetamide	(d) Acetylchloride	
17.	Which of the following statements are correct:			
	(a) The UV spectra of $\beta$ -diketones (e.g., acetyl acetone) is dependent on degree of enolization			
	(b) Cyclic $\beta$ -diketones (e.g., 1,3-cyclohexanedione) exists almost exclusively in the enolic form even in polar solvents.			
	(c)	(c) The UV spectra of $\alpha$ , $\beta$ -unsaturated ketones and aldehydes shown a characteristic intense absorption band ( <i>K</i> -band) in the region 215-250 nm.		
	( <i>d</i> )	(d) All are correct		
18.		Which of the following substituted unsaturated carboxylic acid will exhibit maximum $\lambda_{\text{max}}$ value:		
	(a) (	α or β-monosubstituted	(b) $\alpha$ , $\beta$ or $\beta$ , $\beta$ -disubstituted	
	(c)	$\alpha$ , $\beta$ , $\beta$ -trisubstituted	(d) All will give same $\lambda_{\text{max}}$ value	

19. Benzene shows three absorption band 184 nm  $(E_1)$   $(\varepsilon_{\text{max}}$  60,000), 204 nm  $(E_2)$   $(\varepsilon_{\text{max}}$  7900) and 256 (B)  $(\varepsilon_{\text{max}}$  200). Which of these is/are weak bands:

(a) 204 nm ( $\varepsilon_{\text{max}}$  7900)

(b) 184 nm ( $\varepsilon_{\text{max}}$  60,000)

(c) 256 nm ( $\varepsilon_{max}$  200)

(d) All are of equal strength

#### **ANSWERS** 2. (c)**3.** (a), (d) **4.** (a), (b) **1.** (b), (c) **8.** (a), (b), (c)**5.** (a) **6.** (a), (c)7. (a) **9.** (a), (b), (c) **10.** (*c*) **11.** (a) **12.** (a) **13.** (*c*) **14.** (a), (b) **15.** (b) **16.** (a) **17.** (*d*) **18.** (*c*) **19.** (a), (c)

### **II. Short Answer Questions**

- 1. According to Fieser-Kuhn rule, calculate  $\lambda_{max}$  for  $\beta$  carotene.
- Using Woodward rule for absorption for conjugated dienes, (Table 24.3), calculate the λ<sub>max</sub> for 2,3-dimethyl 1,3-butadiene.
- **3.** Saturated hydrocarbons can be used as solvents in UV spectroscopy. Explain why? [**Hint:** Saturated hydrocarbons are transparent in the near UV region.]
- **4.** On the basis of Woodward Empirical rules for enone and dienone absorption (Table 24.13), calculate the  $\lambda_{max}$  for 1-methyl cyclohexene.

[Ans. 
$$\lambda_{\text{max}} = 237$$
]

- 5. Why saturated carboxylic acid do not exhibit any UV absorption? [Hint: These give a weak band at about 200 nm resulting from forbidden  $n\rightarrow\pi^*$  transition; this band is of little diagnostic value.]
- 6. Using the data given in Table 24.13, calculate  $\lambda_{max}$  for  $CH_2 = C COOH$

[Ans. 
$$\lambda_{\text{max}} = 210 \text{ nm}$$
]

7. Using the data given in Table 24.20, calculate  $\lambda_{\text{max}}$  for 3-carbethoxy-4-methyl-5-chloro-8-hydroxytetralone and 3,4-dimethoxy-4b, 5, 6, 7, 8, 8a, 9, 10-octahydrophenonthren-10-one.

[Ans. 256 and 278 nm respectively]

8. Calculate the  $\lambda_{\text{max}}$  for toluene, phenol and aniline from the primary band  $(\pi \rightarrow \pi^*$  transition) using the data given in Table 24.22.

[Ans. 206.5, 210.5 and 230 nm respectively]

- **9.** Discuss the applications of UV spectroscopy.
- **10.** Comment on the following:
  - (i) UV absorption of biphenyl and 2,2'-dimethyl biphenyl
  - (ii) UV absorption of diphenyl methane, 4-nitro-4'-methoxy diphenyl methane, p-nitrotoluene and p-methoxytoluene.

- 11. The UV spectrum of mesityl oxide and choles-4-ene-3-one is similar. Comment. [Hint: The UV spectrum of the two compounds is similar; the absorption results from the conjugated enol portion of the two compounds.]
- 12. A diene has  $\lambda_{max}$  175 nm. What information you can obtain from this?

### III. Solved Problems

- 1. What do you expect when a organic molecule absorbs UV radiation?
- **Ans.** Absorption of UV radiation is responsible for electronic transitions, promotion of electrons from ground state (low energy) to excited state (higher energy)
  - 2. What is a chromophore?
- **Ans.** It is a covalently unsaturated groups that is responsible for the absorption of UV radiation. Examples include C=C, C=O, NO<sub>2</sub>.
  - 3. Identity the auxochromic groups present in phenol, aniline and bromine.

Ans. -OH, NH, and Br.

4. How is the intensity of an absorption band in the UV spectrum expressed.

**Ans.** 
$$E_{1 \text{ cm}}^{1\%} = \frac{A}{ch}$$

Where c is the concentration in g/100 mL and b is the path length through the sample in centimeter.

**5.** Which of the transition  $n \rightarrow \pi^*$  or  $\pi \rightarrow \pi^*$  requires more energy.

**Ans.**  $\pi \rightarrow \pi^*$  transition.

**6.** What type of electronic transions are there in ethane, ethylene, butadiene, Acrolein and benzene

Ans.	Ethane	$\sigma \rightarrow \sigma^*$
	Ethylene	$\pi{\to}\pi^*$
	Butadiene	$\pi{\to}\pi^*$
	Aerolein	$\pi{\to}\pi^*$
		$n{ ightarrow}\pi^*$
	Benzene	$\pi{\to}\pi^*$

- 7. What is an UV spectrum?
- **Ans.** It is a plot of wavelength of light absorbed versus the absorption intensity and is recorded conveniently by plotting molar absorptivity (E) against wavelength (nm).
  - **8.** Which one *cis* or *trans* isomer of stilbene absorbs at shorter wavelength.
- Ans. Cis-isomer. In cis-isomer, the steric strain prevents complete coplanarity necessary for conjurgation and so it absorbs at shorter wavelength ( $\lambda_{max}$  274 nm, compared to *trans* isomer,  $\lambda_{max}$  294 nm).
  - 9. The  $\lambda_{max}$  for aniline is 230 nm in neutral medium. However,  $\lambda_{max}$  for aniline in acidic medium is 203 nm. Explain.
- **Ans.** In aniline the pair of electrons on N is in conjugation with the aromatic ring so in this case  $\lambda_{\text{max}}$  is at 230 nm. However, in acidic medium, the lone pair of electrons on N are not in conjugation with the aromatic ring. So  $\lambda_{max}$  in this case is at shorter wavelength (203 nm).

- 10. Biphenyl exhibits  $\lambda_{max}$  at 232 nm ( $\epsilon$  19000) but the corresponding 2,2' dimethyl derivative exhibits  $\lambda_{max}$  at 270 ( $\epsilon$  800). Explain.
- **Ans.** In biphenyl, the two aromatic rings, are in conjugation. However, in case of 2,2'-dimethyl biphenyl there is twisted conformation. In fact this compound can be regarded as two molecules of *o*-xylene as shown by absorption similar to *o*-xylene, due to loss of conjugation in the twisted conformation.



# Chapter 25

## Nuclear Magnetic Resonance (NMR) Spectroscopy

The nuclear magnetic resonance (NMR) spectroscopy is of three types: Proton nuclear magnetic resonance (<sup>1</sup>HNMR) spectroscopy, carbon-13 nuclear magnetic resonance (<sup>13</sup>CNMR) spectroscopy and electron spin resonance (ESR) spectroscopy.

## 25.1 PROTON NUCLEAR MAGNETIC RESONANCE (1HNMR OR PMR) SPECTROSCOPY

### 25.1.1 Introduction

The proton magnetic resonance (PMR) spectroscopy is the most important technique used for the characterisation of organic compounds. It gives information about different kinds of protons in the molecule. In other words, it tells about different kinds of environment of the hydrogen atoms in the molecule. PMR also gives information about the number of protons of each type and the ratio of different types of protons in the molecule.

It is well-known that all nuclei carry a positive charge. In some nuclei this charge 'spins' on the nuclear axis, and this circulation of nuclear charge generates a magnetic dipole along the axis. Thus, the nucleus behaves like a tiny bar magnet. The angular momentum of the spinning charge is described in terms of spin number (I) which can have values of 0,  $\frac{1}{2}$ , 1,  $\frac{3}{2}$ , etc., (I = 0 denotes no spin). The magnitude of the generated dipole is expressed in terms of nuclear magnetic moment ( $\mu$ ).

In a nucleus, each proton and neutron has its own spin, and I is the resultant of these spins. In case, the sum of protons and neutrons is even, I is zero or integral

(0, 1, 2,...). On the other hand, if the sum of protons and neutrons is odd, I is half-integral  $\left(\frac{1}{2}, \frac{3}{2}, \frac{5}{2}, ...\right)$ . However, if both protons and neutrons are even numbered, I is zero. Both <sup>12</sup>C and <sup>16</sup>O belong to this category and give no NMR signal.

Nuclei with spin number I of  $\frac{1}{2}$  (examples include <sup>1</sup>H, <sup>19</sup>F, <sup>13</sup>C and <sup>31</sup>P) have a uniform spherical charge distribution. However, nuclei with spin number I of 1 or higher have a non-spherical charge distribution. Examples of nuclei with spin number I of 1 are <sup>14</sup>N and <sup>2</sup>H and examples of nuclei with spin number I of  $\frac{3}{2}$  are <sup>11</sup>B, <sup>35</sup>Cl, <sup>79</sup>Br and <sup>81</sup>Br.

In PMR, we are concerned with  $^1H$  or proton. The spinning nucleus of a hydrogen atom ( $^1H$  or proton) is the simplest and is commonly encountered in organic ocmpounds. The hydrogen nucleus has a magnetic moment,  $\mu = 2.79268$  and its spin number (I) is  $\pm \frac{1}{2}$ . Hence, in an applied external magnetic field, its magnetic moment may have two possible orientations. The orientation in which the magnetic moment is aligned with the applied magnetic field is more stable (lower energy) than in which the magnetic moment is aligned agaist the field (higher energy). The energy required for flipping the proton from its lower energy alignment to the higher energy alignment depends upon the difference in energy ( $\Delta E$ ) between the two states and is equal to hv ( $\Delta E = hv$ ). Just how much energy is needed to flip the proton over depends on the strength of the external field; the stronger the field, the greater the tendency to remain lined up with it, and higher the frequency of the radiation needed to do the spin flip;  $v \propto H_0$ 

i.e., 
$$v = \frac{H_0 v}{2\pi}$$

where, v is the frequency in Hz,

 ${\cal H}_0$  is the strength of the magnetic field in gauss, and

v is a nuclear constant, the magnetogyric ratio, 26750 for the proton.

In a field of 14,092 gauss, for example, the energy required to flip a proton corresponds to electromagnetic radiation of frequency 60 MHz (60 megahertz or 60 million cycles per second). For higher frequencies of operation, proportionately higher field has to be used.

In principle, the substance could be placed in a magnetic field of constant strength, and then the spectrum can be obtained in the same way as an infrared or an ultraviolet spectrum by passing radiation of steadily changing frequency through the substance and observing the frequency at which radiation is absorbed. In practice, however, it has been found to be more convenient to keep the radiation frequency constant and to vary the strength of the magnetic field. At some value of

the field strength, the energy required to flip the proton matches the energy of the radiation, absorption occurs and a signal is obtained. Such a spectrum is called a nuclear magnetic resonance (NMR) spectrum (Fig. 25.1).

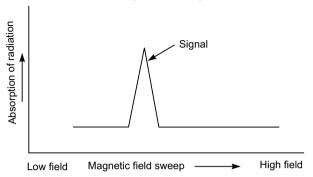


Fig. 25.1 The PMR spectrum

## 25.1.2 The NMR Spectrometer

It consists of the following components. A powerful magnet with a homogeneous field that can be continuously and precisely varied over a relative narrow range; this is accomplished by a sweep generator. A radio frequency transmitter and a radio-frequency receiver are the two other components of the spectrometer. Besides these, the instrument has a recorder, a calibrator and an integrator. A sample holder positions, the sample between the magnetic poles; the holder also spins the sample by an air driven turbine to average the magnetic field over the sample. The solution of the sample is normally prepared in a solvent containing no protons. The most common solvents are deuterated chloroform (CDCl<sub>3</sub>) and carbon tetrachloride (CCl<sub>4</sub>). A small amount of (1%) tetramethylsilane (TMS) is added as an internal standard.

A diagramatic representation of a NMR spectrotmeter is shown in Fig. 25.2.

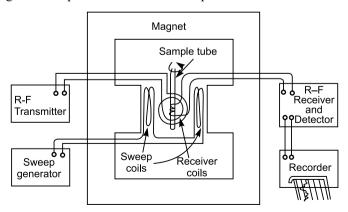


Fig. 25.2 Diagrammatic representation of a NMR spectrometer

The essential features of a NMR spectrometer are given below:

- (i) The Magnet: A permanent or electromagnet is used in a NMR spectrometer. It should be capable of generating a stable, strong and homogeneous magnetic field (up to 25000 gauss).
  - The homogeneous magnetic field is due to a small air gap between the poles (3-5 inches) and spinning of the sample.
- (ii) Sweep Generator: It is helpful to the magnetic field can be continuously and pricely varied over a narrow range.
- (iii) Sample Holder: It holds the sample in between the two poles of the magnet. The holder permits the sample tube spinning by air driven turbine. This is necessary to average the magnetic field over the sample; This avoids the formation of spinning side bands.
- (iv) Radio Frequency Generator: It generates radio frequency radiation. The coil of the generator is wound perpendicular to the applied magnetic field and transmits radio waves of fixed frequency like 60, 100, 200, 300 MHz to a coil that encircles the sample tube. The radio frequency generator uses a power supply device in order to vary the level of RF power and adjust to get the best results.
- (v) Radio Frequency Detector: It detects the frequency at which radiation is absorbed. The coil of the RF detector is prependicular to both the magnetic field and the coil of the radio frequency generator.
- (vi) Recorder and Integrator: The amplified signal from the radio frequency detector is sent to the recorder, which records the spectrum as a plot of resonance signal on the y-axis verses strength of the resonance signal (x-axis).

Most of the NMR spectrometers are equipped with automatic integrators in order to measure the area under each peak.

## 25.1.2.1 Commonly Used NMR Solvents

The NMR solvents should be such that there are no protons. Following are given some of the commonly used solvents.

- (i) Carbon Tetrachloride (CCl<sub>4</sub>): Being cheap and aprotic, it is the best solvent provided the compound is soluble in it. However, appropriate care should be taken in handling this solvent since it is known to be carcinogenic.
- (ii) **Deutro Chloroform (CDCl<sub>3</sub>):** It is the most useful solvent. A small signal due to residual CHCl<sub>3</sub> at δ7.27 does not cause any problem. However, being volatile, the compound can be recovered whenever

necessary. In case the compound is sparingly soluble in  $CDCl_3$ , few drops of DMSO- $d_6$  may be added. DMSO- $d_6$  also causes the residual  $CHCl_3$  signal to shift out field ( $\delta 8.38$ ). It is normally mixed with 1% TMS (internal standard).

- (iii) Deutrated Dimethyl (Sulphoxide (DMSO-d<sub>6</sub>): It is a good solvent for dissolving heterocyclic compounds. However, being a mild oxidising agent, it can react with some compounds. Being non-volatile it is rather difficult to recover the compound if desired. Adequate precaution should be taken while using DMSO-d<sub>6</sub> as it gets absorbed through the skin along with the sample. Thus, it can be a source of hazard. In case of spillage the hands must be washed immediately with plenty of water.
- (iv) **Deutrero Methanol (MeOH-d<sub>4</sub>):** It is a polar solvent and can be used for running the NMR spectra of polar compounds and salts. Since it has high affinity for water it is not possible to keep it dry. It gives a residual peak around δ4.8 and δ3.3-3.2 due to CD<sub>2</sub>HOD. A disadvantage is that it is susceptable to exchange ionizable protons (e.g., OH, NH<sub>2</sub> or CONH<sub>2</sub>) in the sample. Also protons alpha to the CO group may be exchanged due to enol formation.
- (v) Deutero Water ( $D_2O$ ): Though it is a good polar solvent, its use is limited since it readily exchanges all acidic protons exhibiting a strong HOD signal at  $\delta 4.9$ .
- (vi) **Deuterobenzene** ( $C_6 D_6$ ): It should be used with caution since it is a suspected carcogen. It shows a residual CD<sub>5</sub>H signal at  $\delta$ 7.27.
- (vii) Trifluoroacetic Acid (CF<sub>3</sub>COOH): It is useful for running NMR spectra of heterocyclic compounds and amines. It should be used with caution since it causes blisters on contact with skin. It shows a strong broad-COOH signal at  $\delta 11.0$ , whould does not interfere with other signals.

## 25.1.2.2 Preparation of Solution

Normally CDCl<sub>3</sub> is used as the solvent. Approximately 20 mg. of a pure organic compound is dissolved in 0.5 mL of the solvent and then transferred in a clean NMR tube.

The concentration of the compound should be such that an adequate signal to noise ratio be obtained. In fact, the concentration of spin active nucleus (*e.g.*, <sup>1</sup>H in case of Proton spectroscopy) and not the overall concentration of the compound. The percentage of H is a compound is calculated as follows.

% Hydrogen = 
$$\frac{\text{No. of hydrogens}}{\text{Molecular mass}} \times 100$$

Thus, in case of benzoic acid, the per cent hydrogen is  $=\frac{6}{121} \times 100 = 4.95$ .

### Internal Standard

As already mentioned, tetramethyl silane  $[(CH_3)_4 Si]$  is used as an internal standard. This is due to the following:

- TMS gives a sharp line from twelve magnitically equivalent protons.
- Its resonance position is far away from the absorption due to protons in the molecule. It gives a single absorption peack at higher field than almost all organic protons. So by arbitraily assigning TMS = 0, it is possible to have a scale in which most proton resonance will be on the left of TMS signal.
- It is chemically inert (B.P. 27°C).
- Since it is volatile, it is possible to remove it from the sample in case the sample has to be recovered.

In case  $D_2O$  is used as the solvent, TMS is used as an 'external reference' by taking it in a sealed capillary immersed in the solution. Some times 2,2-dimethyl

2-silapentane-5-sulfonate (DSS)  $\left[ (CH_3)_3 SiCH_2 CH_2 CH_2 SO_3^- Na^+ \right]$  is also used as internal standard if  $D_2O$  is used as is solvent. The reference peak in case of DSS will be within 0.01 to 0.03 ppm of the TMS peak.

These days the NMR instruments use superconducting magnets that have much higher magnetic field strength. The magnet coil in the superconducting magnet conducts electricity with virtually zero resistance. The magnetic coil in a superconducting magnet has to be kept very, very cold (as cold as liquid helium, which boils at 4.3 K *i.e.*, 4.3 degrees absolute zero). If the temperature of the coil warms, resistance in the wire builds up, heat is generated and the liquid helium boils away and the magnetic field is quenched. In order to keep the superconducting coil cool, the entire magnet is housed in a Dewar chamber, which contains liquid helium. The NMR instruments employing superconducting magnets are known as **Fourier Transform (FT) NMR spectrometers**. They also use computers that allow signal averaging followed by a mathematical calculation known as Fourier transform. These instruments achieve very high resolutions and much greater sensitivity than the usual NMR instruments.

## 25.1.3 Interpretation of the <sup>1</sup>HNMR Spectra

If the situation is as simple as described above (Fig. 25.1), all the protons in an organic compound would absorb at the same field strength, the results is that the spectrum consists of a single signal that would not tell us any thing about

the structure of the molecule. But the frequency at which a proton absorbs depends on the magnetic field which that proton feels, and this effective field strength is not exactly the same as the applied field strength. The effective field strength at each proton depends on the environment of that proton, particularly presence of other, nearly protons. Each proton or each set of equivalent protons will have a slightly different environment from any other set of protons and so will require a slightly different applied field strength to produce the same effective field strength. It is this particular field strength at which absorption takes place.

Thus, at a given radio frequency, all protons absorb at the same effective field strength, but they absorb at different applied field strength. In fact, it is this applied field strength that is measured, and against which the absorption is plotted. Thus, a spectrum shows many absorption peaks, whose relative positions reflect the environment of protons. This can give detailed information about the molecular structure.

For interpretation of an NMR spectrum we should find the following:

- (i) Number of signals, which tell us how many different kind of protons are present in a molecule.
- (ii) The position of the signals. These tell us about the electronic environment of each kind of proton.
- (iii) The intensities of the signals. These tell us how many protons of each kind are present.
- (iv) The splitting of a signal into several peaks. This tells us about the environment of a proton with respect to other nearby protons.

The interpretation of <sup>1</sup>HNMR spectra has also been dealt in Final Interpretation of <sup>1</sup>HNMR Spectra (*see* section 25.1.6).

## Number of Signals

In an organic molecule, protons with the same environment absorb at the same (applied) field strength. Such protons are said to be equivalent protons. However, protons with different environment absorb at different (applied) field strength (such protons are said to be non-equivalent). Thus, the number of signals in the NMR spectrum tell us as to how many sets of equivalent protons or how many 'kinds' of proton a molecule contains.

In order to decide whether protons are magnetically equivalent or not, replace each proton separately by another atom. Protons which on replacement form the same compound or its equivalent are magnetically equivalent. As an example, replacement of any of the three protons in a methyl groups give the same product as shown below for propane, so all the three protons of this methyl group are magnetically equivalent.

In case, the replacement procedure generates diastereomers, then the protons are not magnetically equivalent. Such types of molecules are subsequently discussed.

Protons attached to symmetry-related atoms are also magnetically equivalent. For identifying symmetry-equivalent atoms or groups, one has to look for the presence of a centre, an axis or a plane of symmetry. In case if one of these symmetry elements is present, the protons are magnetically equivalent. These are well-understood by considering the following examples:

(i) Tetrahydrofuran: Replace each of the protons of the methylene group (made bold) with "X". The two compounds obtained are enantiomers and so the protons shown is bold are magnetically equivalent.

To find, if tetrahydrofuran has a plane of symmetry. In this case, the protons with a mirror-image relationship are magnetically equivalent.

$$H_a$$
  $H_b$ 
 $H_2$   $C$   $C$ 
 $H_2$   $C$   $C$ 
 $H_a$   $H_b$ 

Plane of symmetry  $H_b$ 

As seen, tetrahydrofuran has a plane of symmetry. The protons marked H<sub>a</sub> are equivalent as they have a mirror image relationship. Similarly, the protons marked H<sub>b</sub> are equivalent since they have mirror-image relationship. Therefore, all four protons (marked H<sub>a</sub> and H<sub>b</sub>) attached to the carbon atoms adjacent to oxygen atoms are magnetically equivalent.

(ii) Toluene: In case of toluene, replacement of each of the protons of the methyl group with 'X' gives the same compound. Thus, the three protons (marked bold) are magnetically equivalent. It is also seen that a methyl group possess a three-fold axis that interrelates the positions of its protons.

The axis of symmetry (as shown above) implies that all the three protons (shown in bold face) are magnetically equivalent.

(iii) Trans-2-Butene: In this case, the alkene protons (marked bold) are magnetically equivalent. This can be ascertained not only by replacing each alkene proton by a group 'X', but can also be recognised since the molecule possesses a center of symmetry (lines connecting the same atoms groups pass through the center of the molecule.

Thus, the center of symmetry indicates that the alkene protons (made bold) are magnetically equivalent. Also, all the six methyl protons are also magnetically equivalent. Following examples illustrate the equivalent (equivalent protons are designated by the same alphabet) and non-equivalent protons and also illustrate how many different types of protons are present (depending on the number of NMR signals) (Fig. 25.3).

Fig. 25.3

Though we can tell by looking at a formula as to which protons are in different environment and should give different signal. However, we cannot always predict, particularly with stereochemically different protons. The easiest and surest way to find the equivalent and non-equivalent protons is decided in the following way. If replacement of each of the two protons by another atom (say Z) would yield the same product, then the two protons are chemically equivalent in an achiral medium.

Let us consider the example of ethyl chloride. Replacement of a methyl proton would give CH<sub>2</sub>Z—CH<sub>2</sub>Cl and replacement of a methylene proton would give CH<sub>3</sub>—CHZCl. Both these are different products. Thus, methyl protons are non-equivalent to methylene protons. However, the product CH<sub>2</sub>Z—CH<sub>2</sub>Cl is the same regardless of which one of the three methyl protons is replaced. Thus, the (average) environment of the three protons is identical and hence, only one NMR signal is expected from all the three.

In ethyl chloride, replacement of the two methylene protons would give one of a pair of enantiomers (*see* Fig. 25.4).

Fig. 25.4

Such pairs of protons are called **enantiotopic protons.** The environments of these two protons are mirror images of each other; is an achiral medium, these protons behave as if they were equivalent and one observes one NMR signal for the pair.

Consider the case of 2-bromopropene. In this case, replacement of either of the vinylic proton gives one of a pair of diasteromers (in this case, geometric isomers) (see Fig. 25.5).

Fig. 25.5

Such pairs of protons are called **disastereotopic protons.** The environments of these two protons are neither identical nor mirror images of each other; these protons are non-equivalent and one NMR signal is expected from each one.

In a similar way, in 1,2-dichloropropane, the two protons on C-1 are diastereotopic, non-equivalent and so will give separate NMR signals (Fig 25.6).

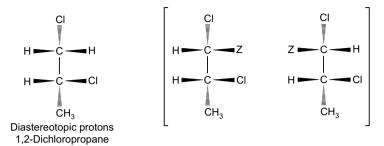


Fig. 25.6

## QUESTIONS WITH ANSWERS

**Q.1.** How many NMR signals will be obtained in the case of 2-methyl propene?

**Ans.** In 2-methyl propene, the six methyl protons will give one signal, since replacement any of the six methyl protons with chlorine gives the same compound, *i.e.*, 3-chloro-2-methyl propene,

Also, the replacement of the two vinyl hydrogen with chlorine gives the same compound, *i.e.*, 1-chloro-2-methylpropene.

Therefore, the NMR signal of 2-methylpropene will give two signals, one for two methyl groups and another for the two vinyl hydrogens.

Q.2. How many NMR signals will be obtained in the case of Ethyl bromide, CH<sub>3</sub>CH<sub>2</sub>Br?

**Ans.** Replacement of each of the two hydrogen atoms by the same group, gives compounds that are enantiomers, the two hydrogens are said to be **enantiotopic.** Enantiotopic H atoms have the same chemical shift

and given only one <sup>1</sup>HNMR singal. (Note that enantiotopic H atoms may not have the same chemical shift if the compound is dissolved in a chiral solvent. However, most <sup>1</sup>HNMR spectras taken in achiral solvents and in this case enantiotopic protons have the same chemical shift.)

Thus, the two hydrogens of the —CH<sub>2</sub>Br groups of ethyl bromide are enantiotopic and will give one NMR signal. The three equivalent protons of CH<sub>3</sub> group will give one signal. So, a total of two signals will be obtained.

However, as we will see later the <sup>1</sup>HNMR spectrum of ethyl bromide actually gives seven peaks (three in one signal and four in the other due to spin-spin splitting (*see* Fig. 25.20).

Q.3. How many NMR signals will be obtained in the case of chloroethene, CH<sub>2</sub>=CHCl?

**Ans.** If replacement of each of the two H atoms by a group Z, gives compounds that are disastereomers, the two hydrogens are said to be diastereotopic. Such diastereotopic proteins do not have the same chemical shift and give different <sup>1</sup>HNMR signals.

CI H Replacement by Z 
$$\rightarrow$$
 CI  $\rightarrow$  CI  $\rightarrow$  H  $\rightarrow$  C=C  $\rightarrow$  Chloroethene Diastereomers

So, chloroethene will give signals from three non-equivalent protons: one for the proton of Cl CH=group and the other for each of the diastereotopic protons of the =CH<sub>2</sub> group.

## Position of the Signals

Just as the number of signals in NMR spectrum indicate how many kinds of protons are present in a molecule, the positions, of the signals give an idea of the nature of the protons, *i.e.*, whether they are aromatic, aliphatic, primary, secondary, tertiary, benzylic, vinylic, acetylenic and adjacent to halogen or other hetero atoms or functional groups. These protons have different electronic environments and hence they show different absorptions at different positions in the NMR spectrum.

When a compound is placed in a magnetic field, the electrons surrounding hydrogen are caused to circulate and in circulating they generate secondary magnetic field, *i.e.*, induced magnetic field. The circulation of electrons generate a field aligned in such a way that at the proton it opposes the applied field. Thus, the field felt by the proton is diminished and the proton is said to be **shielded**. Alternatively, the circulation of electrons about nearby nuclei generates a field that can either oppose or reinforce the applied field at the proton, depending on the proton location. If the induced field opposes the applied field, the proton is shielded as before. If the induced field reinforces the applied field, then the field felt by the proton is augmented and the proton is said to be **deshielded**. A shielded proton requires a higher applied field strength and a deshielded proton requires a lower applied field strength to provide the particular effective field strength at which absorption occurs. Shielding thus shifts the absorption upfield, and the deshielding shifts the absorption downfield.

Such shifts in the position of the NMR absorptions (as described above) are called **chemical shifts**. We can say that the numeric value of a signal relative to the position of the TMS peak is called the chemical shift.

Chemical shifts are expressed in dimensionless units, and are independent of the applied frequency. The chemical shifts are obtained by dividing v by applied frequency and multiplying by  $10^6$ . As an example, a peak at 60 Hz (v60) from TMS at an applied frequency of 60 MHz will be at  $\delta 1.00 (\delta \text{ scale})$ .

$$\frac{60}{60 \times 10^6} \times 10^6 = \delta 1.00$$

The same peak at an applied frequency of 100 MHz will be at v100 will also be at  $\delta 1.00$ 

$$\frac{100}{100 \times 10^6} \times 10^6 = \delta 1.00.$$

The reference point from which the chemical shift is measured is with respect to a signal from an actual compound–usually tetramethyl silane,  $(CH_3)_4$  Si, which exhibits a single sharp peak at a high field strength because of strong shielding of the protons, due to low electronegativity of silicon. Most of the organic protons absorb downfield (left side) with respect to TMS. This separation of the spectral signals of protons due to their different chemical environments relative to TMS is called chemical shift.

The chemical shifts are expressed in a form (scale) that makes it independent of the instrumental frequency and the intrinsic strength of the magnetic field that the NMR spectrometer employs. The most commonly used scale is the  $\delta$  (delta) scale. The position of tetramethylsilane signal is taken as 0.00 ppm. Most of the chemical shifts have  $\delta$  values between 0 and 10. Another scale, *i.e.*,  $\tau$  (tau) scale is also used sometimes; in this scale the TMS signal appears at 10 ppm most of the values lie between 0 and 10. The two scales are related as  $\tau = 10 - \delta$  (Fig. 25.7).

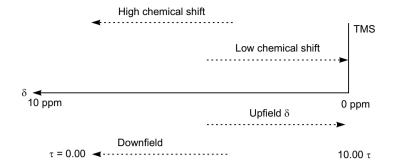


Fig. 25.7

Let us consider, the NMR spectra (Fig. 25.8) of methoxymethyl bromide,  $CH_3$  OCH<sub>2</sub>Br. Though it contains five protons but the spectrum shows only two signals. The two protons of methylene group ( $\delta$  5.7) and three protons of the methyl group ( $\delta$  3.5) are magnetically equivalent. (We know that the chemical shifts of magnetically equivalent protons are identical.)

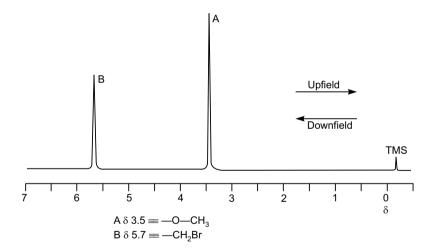


Fig. 25.8

As seen, the signals observed for methyl and methylene protons in CH<sub>3</sub>OCH<sub>2</sub>Br are downfield from TMS (appearing left of TMS signal). The signal for CH<sub>3</sub> group is upfield (to the right) of the signal for the CH<sub>2</sub> group but downfield (to the left) of the TMS signal. As already stated, the proten that give signals that are down field are deshielded implying that they experience slightly stronger magnetic field than those absorbing up field. On the other hand proton signals upfield from those other are more shielded and experience a slightly weaker magnetic field.

## 25.1.4 Chemical Shifts of Different Types of Protons

The protons on carbon differ from the protons of heteroatom. The protons on heteroatom are exchangeable and exhibit hydrogen bonding. Following is given the chemical shifts of different types of hydrogens.

### **Alcohols**

Ethyl alcohol shows hydroxylic proton at  $\delta 5.35$  (broad peak) (pure alcohol),  $\delta 2-4$  (ordinaryl ethyl alcohol in  $CCl_4$  or  $CDCl_3$ ) or  $\delta 0.5$  (vapour phase). In fact, change in solvent or temperature also shifts the hydroxylic peak; this is due to change in concentration, temperature and solvent. Hydrogen bonding moves the proton absorption to lower field; this is due to decrease in electron density around the proton. The intermolecular hydrogen bonding is decreased by dilution with nonpolar solvent and also with increase in temperature. Intramolecular hydrogen bonding is generally not affected by change of environment. Examples of intramolecular hydrogen bonding include salicylates and  $\beta$ -diketones (enolic form).

Salicylates 
$$\beta$$
-Diketone (enolic form)  $\delta$ OH = 10-16

It is found that under ordinary conditions, the hydroxylic peak of ethyl alcohol appears as a singlet. This is because under ordinary conditions, acidic impurities which are present in the solution catalyse the rapid exchange of the hydroxylic proton. In fact, the proton is not on the oxygen atom for long enough time to be affected by the three states of the methylene protons and so there is no coupling. The rate of exchange can be also be decreased by treating the solution with anhydrous sodium carbonate or alumina before obtaining the spectrum. Deuterated dimethyl sulfoxide (pure) also lowers the rate of exchange, but shifts the hydroxylic proton to lower field even in dilute solution, by H-bonding between the solute and solvent (O.L. Chapman and R.W. King, J. Am Cham. Soc., 1964, **86**, 1256; D.E. Mc Greer and M.M. Mocek, J. Chem. Ed., 1963, **40**, 358). Under these circumstances, the hydroxylic proton can couple with the protons on the α-carbon, a primary alcohol shows a triplet, a secondary alcohol a doublet and a tetrary alcohol a singlet (spectras taken in DMSO).

In all the above cases absorption at  $\delta$ .26 is due to DMSO Table below lists the chemical shift of Hydroxyl Proton in DMSO.

Compound	Chemical Shift (δ)	Multiplicity
Methyl alcohol	4.08	q
Ethyl alcohol	4.35	t
Isopropyl alcohol	4.35	d
t-Butyl alcohol	4.16	s
t-Pentyl alcohol	3.99	s
Propylene glycol, 1-OH	4.45	t
2-OH	4.38	d
Cyclohexanol	3.38	d
Benzyl alcohol	5.16	t
Phenol	9.25	s

- 1. Data taken from O.L. Chapman and R.W. King, J. Am. Chem. Soc., 1964, 86, 1256
- 2. The spectras were taken in DMSO solution (concentration 10 mole per cent or less)

In case a compound contains an exchangeable proton, the spectrum can be simplified and the absorption due to exchangeable proton removed by simply shaking the solution with excess  $D_2O$  or by taking the spectra in  $D_2O$  if the compound is soluble in  $D_2O$ . However, in these cases, a peak due to HOD occurs at  $\delta$  5-4.5 (in nonpolar solvents) and near  $\delta$  3.3 in DMSO.

### **Phenois**

The phenolic proton resembles an alcoholic proton in the NMR signal. Usually, the phenolic peak is a sharp singlet (rapid exchange, no coupling) at  $\delta$  7.5–4.0 (depending on the concentration, solvent and temperature). The presence of carbonyl group in ortho position, shifts the absorption due to phenolic proton downfield in the range ( $\delta$  12.0–10.0) due to intramolecular H bonding. Thus, o-hydroxyacetophenone, shows a peak at about 12.05. However, in case of o-chlorophenol, there is much weaker intramolecular H-bonding the phenol proton gives absorption at about  $\delta$  6.3.

### **Enols**

The enols are generalised by intramoleculer hydrogen bonding, which varies from very strong in aliphatic  $\beta$ -diketones to weak in case of cyclic  $\alpha$ -diketones. In general the enolic proton is downfield compared to alcoholic proton. As an example, the enolic proton of acetyl acetone and dibenzoyl methane absorbs at  $\delta$  15.0 and  $\delta$  16.6 respectively. The enolic proton peaks are frequently weak at room temperature; this is due to slow exchange. As the keto-enol conversion is slow, peaks of both forms can be observed and the equilibrium measured.

## **Carboxylic Acids**

The carboxylic protons absorb in the range  $\delta$  13.2 to  $\delta$  10.0, since these exist as stable H-bonded dimers in non-polar solvents even in dilute solutions. The absorption is not affected by concentration. However, polar solvents disrupt the dimer and there in shift in the absorption of proton. The carboxylic proton rapidly exchanges with protons of water and alcohols (or OH groups of hydroxy acids) to give a single peak, whose position depnds on concentration.

## **Protons on Nitrogen**

 $^{14}$ N nucleus has a spin number I = 1. It should cause a proton attached to it and a proton on an adjacent C atom to exhibit three peaks of equal intensity. However, this is not so. The absorption of proton on nitrogen depends on the exchange of the proton on nitrogen atom (which can be rapid exchange, intermediate rate or low exchange) and on the electrical quadrupole moment of the  $^{14}$ N nucleus.

If the exchange is rapid, the NH proton (s) is decoupled from the N atom and also from the protons on adjacent C atoms. In such a case, the NH peak is a sharp singlet is obtained and the adjacent CH protons are not split by NH. Most of the aliphatic amines show such a pattern.

On the other hand, at intermediate rate of exchange, the NH proton is decoupled partially resulting in a broad NH peak. In this case also the adjacent CH protons are not split by the NH proton. A typical example is that of N-methyl-*p*-nitroaniline.

However, if the NH exchange rate is low, the NH peak is still broad; this is because the electrical quadrupole moment of the N nucleus cause a moderately efficient spin relaxation and, thus an intermediate lifetime for the spin states of N nucleus. In the last case, coupling of NH protons to the adjacent proton is observed.

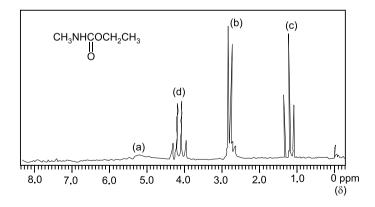


Fig. 25.9 NMR spectra of ethyl N-methyl carbamate (a)  $\delta$ 5.16 NH proton, (b)  $\delta$  2.78(d), N—CH<sub>3</sub>(J = 5 Hz], (c)  $\delta$ 1.23(t) OCH<sub>2</sub>CH<sub>3</sub> and (d) 4.14(q), OCH<sub>2</sub>CH<sub>3</sub>

Some examples include pyrroles, indoles, 1° and 2° amides and carbonates. The coupling between N and protons on adjacent carbon in almost negligible. The coupling is, however, observed in the signal due to H on carbon. The N—H proton signal is broadened due to quadrupole interaction with N. All this is clear by examining the NMR spectra of ethyl (N-methyl carbamate, CH<sub>3</sub>NHCOCH<sub>2</sub>CH<sub>3</sub> [Fig. 25.9].

Following table gives the NH proton absorption in some of the compounds.

Compound	NH proton absorption (ଚ)
Aliphatic and cyclic amine	3.0 to 0.5
Amides, pyrroles, indoles	8.5 to 5.0

In NMR spectroscopy, the use of trifluoroacetic acid as a protonating agent and also a solvent, is helpful for the classification amines as 1°, 2° or 3°. This is shown in the following table:

Classification of amines by NMR of their salts in TFA

Amine	Ammonium salt	Multiplicity of methylene unit	
Primary	C <sub>6</sub> H <sub>5</sub> CH <sub>2</sub> NH <sub>3</sub> <sup>+</sup>	Quartet	
Secondary	$\mathrm{C_6H_5CH_2NH_2R^+}$	Triplet	
Tertiary	C <sub>6</sub> H <sub>5</sub> CH <sub>2</sub> NHR <sub>2</sub> <sup>+</sup>	Doublet	

Data tables from W.R. Anderson and R.M. Silverstein. Anal. Chem. 1965, 37, 1417.

### **Protons on Sulfur**

The rate of exchange on sulfhydryl protons is usually slow. At room temperature, the SH protons couple to protons on adjacent carbons (J  $\sim$  8Hz). However, the SH protons do not exchange rapidly with hydroxyl, carboxylic or enolic protons on the same or other molecules. So separate peaks are observed. The exchange, however, is fast on shaking (2-3 min) with D<sub>2</sub>O; the sulfhydryl protons are replaced with deuterium. The absorption for aliphatic and aromatic sulfhydryl protons is in the range  $\delta$  1.6 to 1.2 and  $\delta$  3.6 to 2.8 respectively.

### **Protons on Halogens**

Due to strong electrical quadrupole moments of halogens (chlorine, bromine or iodine), the nucleus of these halogens are completely decoupled with the protons attached directly or on adjacent C atoms. However, <sup>19</sup>F having spin number 1/2 couples strongly with protons. The <sup>19</sup>F nucleus is observed at 56.4 Hz at 14,092 gauss.

## **Protons in Hydrocarbons**

The chemical shifts of protons in hydrocarbons depend on the nature of hybridization of the carbon on which the proton is attached. Thus,

It is expected on the basis of diamagnetic shielding that the chemical shifts should follow the order  $Sp > Sp^2 > Sp^3$ , depending on the electronegativity of carbon to which H is attached. It is, however found that acetylenic protons (attached to more electronegative Sp carbon) has a chemical shift intermediate to that of methyl protons (attached to least electronegative  $Sp^3$  carbon) and ethylene protons (attached to  $Sp^2$  carbon). It implies that acetylenic protons are more shielded than ethylenic protons although the former are attached to more electronegative Sp carbon. Also, it is expected that aromatic protons (in benzene) should resonate at a frequency similar to that of ethylenic protons. However, contrary to this, benzene protons resonate at lower field ( $\delta$  7–8); this is because benzene protons appear to be less shielded than ethylene protons.

In case of annulene [18], the outside protons are strongly deshielded ( $\delta$  9.28) and the protons inside are strongly shielded ( $\delta$  –2.99). In a similar way the pyrene derivative, trans –15, 16–dimethyl –15, 16–dihydro (a) is aromatic having (4n + 2) $\pi$  electrons. In this case the protons outside the rings are strongly deshielded ( $\delta$  8.14 to 8.64) and the methyl protons inside the rings are strongly shielded ( $\delta$  4.25). Thus, NMR spectroscopy helps to find whether the compound is aromatic or not.

The aromatic protons in o, m- or p-positions are deshielded if an electron withdrawing substitutent is present. The deshielding of protons is in the order o > 0p > m. This is because the electron withdrawing group, due to mesomeric effect, decreases the electron density at o-and p-positions. On the other hand, the presence of electron donating substitutent (e.g.,  $NH_2$ ) causes shielding of the aromatic protons.

The shielding in the order o > p > m due to augmentation of electron density most at o-and p-positions. The shifts in the positions of benzene protons ( $\delta$  7.27) due the presence of various substituents are given in the Table (25.1).

**Table 25.1** Shift in the position of benzene protons ( $\delta$  7.27) caused by various substitutions

Substituent	Ortho	Meta	Para
CH <sub>3</sub>	0.17	0.09	0.18
СООН	-0.8	-0.14	-0.2
COOCH <sub>3</sub>	-0.74	-0.07	-0.20
CN	-0.27	-0.11	-0.3
CONH <sub>2</sub>	0.5	0.2	0.2
—COR	0.6	0.3	0.3
NH <sub>2</sub>	0.75	0.24	0.63
1	-0.40	0.26	0.03
Br	-0.22	0.13	0.03
CI	-0.02	0.06	0.04
СНО	-0.58		-0.27
ОН	0.50	0.14	0.4
NO <sub>2</sub>	DCH <sub>3</sub> 0.43         0.4           Ph         -0.18         0.4		-0.33
OCH <sub>3</sub>			0.37
Ph			0.08
COCI			-0.3

It has already been stated that the chemical shifts are markedly affected by electron density. The presence of positive and negative charges causes deshielding and shielding respectively of the proton attached to the carbon atoms bearing the charges. This is clear by comparing the chemical shift values of cyclopentadienyl anion and cycloheptatrienyl caton. In both these species, all the protons are equivalent, a sharp singlet is observed in each case







As seen, the singlet for cyclopentadienyl anion appear at about  $\delta$  1.85 upfield from benzene protons and that of cycloheptatrienyl cation appear about  $\delta$  1.90 downfield from the benzene protons. Chemical shifts of some other cations and anions are given below:

## 25.1.5 The Splitting of Signals

The splitting of a signal into several peaks gives an idea about the environment of a proton with respect to other nearby protons. The understanding of the splitting of a signal is an important step for the structural assignment of an organic compound. Consider the NMR spectra of the three compounds, 1, 1,2-tribromoethane (CH<sub>3</sub>Br— CHBr<sub>2</sub>), 1, 1-dibromoethane (CH<sub>3</sub>—CHBr<sub>2</sub>) and ethylbromide (CH<sub>3</sub>CH<sub>2</sub>Br). Each of these contains only two kinds of protons. However, instead of two peaks, the spectra show five, six and seven peaks, respectively. This arises due to splitting of the NMR signals caused by spin-spin coupling. The signals we expect from each set of equivalent protons is appearing as a group of peaks (and not a single peak). Thus, splitting reflects the environment of the absorbing protons with respect to nearby protons. Let us consider, the case of adjacent carbon atoms carrying a pair of secondary prtons (b) and a tertiary proton (a) respectively  $\begin{pmatrix} 1 & b \\ -CH--CH_2 \end{pmatrix}$ . First, consider the absorption by one of the secondary protons. The magnetic field that a secondary proton (b) feels at a particular instant is slightly increased or decreased by the spin of the neighbouring teritary proton (a), increased if the tertiary proton happens at the instant to be aligned with the applied field, or decreased if the tertiary proton happens to be aligned against the applied field. Thus, for half the molecules, absorption by a secondary proton is shifted slightly downfield and for the other half of the molecules the absorption is shifted slighty upfield. The signal is split into two peaks-a doublet with equal peak intensities.

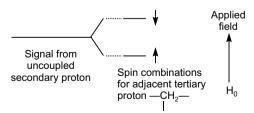


Fig. 25.10 Spin-spin coupling (coupling with one proton gives a 1:1 double)

Thus, we see that proton resonance are influenced through intervening bond by the local fields generated by the protons attached to adjacent carbon atoms. This effect is called **spin coupling**, meaning that the spins of nearby protons couple magnetically with each other. It is more appropriate to commonly use the word **spin-spin splitting** (for spin coupling) due to the appearance of the spectrum itself, in which some signals are split into more than one peaks. In magnetically equivalent protons there is no spin coupling. The influence of neighbouring protons diminishes with distance. Approximate chemical shifts ranges for various structures are given in Table 25.2.

Table 25.2 Approximate chemical shifts ranges for various structures

Structure	Chemical shift range
ROH	
ArOH	
CH <sub>3</sub> —R	
RSH	
RNH <sub>2</sub>	<del></del>
CH <sub>3</sub> —C=C	<del></del>
CH <sub>3</sub> —N<	
CH <sub>3</sub> —CO—	_
CH <sub>3</sub> —AR	_
—C≡CH	_
ArSH	_
CH <sub>3</sub> —OR	
>c=c <h< td=""><td></td></h<>	
(Non-conjugated)	_
>c=c< <sup>H</sup>	
(Conjugated)	
-NH-CO	
ArH	
(Benzenoid)	
ArH (Non-benzenoid)	<del></del>
H—CO—OR	
RCHO	_
RCOOH	
1.00011	

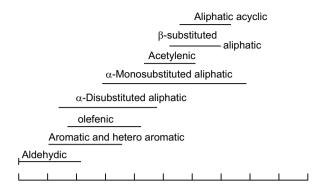


Fig. 25.11 General regions of chemical shifts of protons of various types

It has been found that a proton with a particular environment shows very nearly the same chemical shift, whatever the molecule it happens to be part of. For example, the most familiar classes of hydrogens, viz., primary R CH<sub>3</sub>, secondary R<sub>2</sub>CH<sub>2</sub> and tertiary R<sub>3</sub>CH, absorb at  $\delta$  0.9, 1.3 and 1.5, respectively (this is in the absence of other nearby substituents). However, the aromatic protons (Ar—H) because of powerful deshielding due to the circulation of  $\pi$ -electrons absorb downfield at  $\delta$  6.0—8.5.

The chemical shifts for methyl, methylene and methine protons are given in Table 25.3 and chemical shifts for the protons of some common functional groups and unsaturated systems are given in Table 25.4.

Methyl Protons	δ	Methylene Protons	δ	Methine Protons	δ
CH <sub>3</sub> —C	0.9	CH <sub>2</sub> C	1.3	—СН—С	1.5
CH <sub>3</sub> —C—C=C	1.1	_CH <sub>2</sub> _C_C=C	1.7	_CH_C_C=C	1.9
CH <sub>3</sub> —C—O	1.4	—СH <sub>2</sub> —С—О	1.9	CHCO	2.0
CH <sub>3</sub> —C=C	1.6	CH <sub>2</sub> C=_C	2.3	—CH—C—C	2.2
CH <sub>3</sub> —Ar	2.3	—CH <sub>2</sub> —Ar	2.7	—CH—Ar	3.0
CH <sub>3</sub> —CO—R	2.2	-CH <sub>2</sub> -CO-R	2.4	—CH—CO—R	2.7
CH <sub>3</sub> —CO—Ar	2.6	—CH <sub>2</sub> —CO—Ar	2.9	—CH—CO—Ar	3.5
CH <sub>3</sub> —CO—O—R	2.0	CH <sub>2</sub> COOR	2.2	CHCOOR	2.5
CH <sub>3</sub> —CO—O—Ar	2.4	—CH <sub>2</sub> —CO—O—Ar	2.7	—CH—CO—O—Ar	2.9
CH <sub>3</sub> —O—R	3.3	CH <sub>2</sub> OR	3.4	—CH—O—R	3.7
CH <sub>3</sub> —O—H	3.5	—СН <sub>2</sub> —О—Н	3.6	—CH—O—H	3.9
CH <sub>3</sub> —OAr	3.8	—CH <sub>2</sub> —OAr	4.3	—CH—OAr	4.5
CH <sub>3</sub> —O—CO—R	3.7	CH <sub>2</sub> OCOR	4.1	—CH—O—CO—R	4.8
CH <sub>3</sub> —N	2.3	—CH <sub>2</sub> —N	2.5	—CH—N	2.8

Table 25.3 Chemical shifts for methyl, methylene and methine protons

(Contd...)

$\sim$	1	
/	ч	4

Methyl Protons	δ	Methylene Protons	δ	Methine Protons	δ
CH <sub>3</sub> —NO <sub>2</sub>	4.0	-CH <sub>2</sub> -NO <sub>2</sub>	4.4	—CH—NO <sub>2</sub>	4.7
CH <sub>3</sub> —C—NO <sub>2</sub>	1.6	-CH <sub>2</sub> -C-NO <sub>2</sub>	2.2	-CH-C-NO <sub>2</sub>	2.6
CH <sub>3</sub> —C=C—CO	2.0	-CH <sub>2</sub> -C-C-CO	2.1	_CH_C=C_CO	2.4
CH <sub>3</sub> —C—CI	1.4	—CH <sub>2</sub> —C—CI	1.8	—CH—C—CI	2.0
CH <sub>3</sub> —C—Br	1.8	—CH <sub>2</sub> —C—Br	1.8	—CH—C—Br	1.9
CH <sub>3</sub> —CI	3.0	—CH <sub>2</sub> —CI	3.4	—CH—CI	4.0
CH <sub>3</sub> —Br	2.7	—CH <sub>2</sub> —Br	3.3	—CH—Br	3.9

Chemical shifts are mainly influenced by inductive effects and magnetic anisotropy effects.

### **Inductive Effects**

In case there is an electron whithdrawing group on the atom adjacent to the proton, the electron density is withdrawn from around the hydrogen atom nucleus towards a more electronegative atom; in such a case, the proton will be deshielded and its signal will appear farther downfield. This is clear from the example given below:

Presence of additional electron-withdrawing groups will result shifting the proton signal further downwards. This implies that the inductive effects are additive. Thus,

## **Magnetic Anisotropy Effects**

The term magnetic anisotropy is used to describe the effects that are caused by localized magnetic fields that are created in molecules, especially those having  $\pi$  bonds. This is understood by comparing the resonance values for the protons in 2-methylpropene and 2-methylpropane.

In the examples cited above, each substituent (H and  $CH_3$ ) on the alkene carbons (2-methylpropene) displays a resonance that is downfield from the corresponding groups in the alkane (2-methylpropane). Thus, the  $\pi$  bond of the alkene creates a magnetic field that deshields towards the protons of these attached groups.

Magnetic anisotropy effect is particularly important for aromatic compounds. Thus, the protons attached to benzene ring give absorptions farther downfield compared to those in cyclohexane.

Oxygen containing molecules containing carbonyl  $\pi$  bond also exhibit the effect of magnetic anisotropy. Thus, the aldehydic group causes a significant downfield shift of its proton resonance compared to that of either alkene or the alcohol.

The chemical shifts for the protons of some common functional groups and unsaturated systems are given in Table 25.4.

**Table 25.4** Chemical shifts for the Protons of some common Functional Groups and Unsaturated systems

Types of Protons	δ
R—C <u>H</u> O	9.4–9.7
Ar—C <u>H</u> O	9.7–10.0
R—N <u>H</u> <sub>2</sub>	5.0–8.0
Ar—N <u>H</u> <sub>2</sub>	3.4–6.0
R—0 <u>H</u>	0.5–4.0
Ar—O <u>H</u>	4.4–12
R—S <u>H</u>	1.0–2.0
Ar—S <u>H</u>	3.0–4.0
R—COO <u>H</u>	10.5–13.0

(Contd...)

Types of Protons	δ
O    R—C—OCH <sub>2</sub> R'	3.7–4.1
R—O—C— <u>H</u>	3.3–4.0
<u>H</u> C—C (isolated)	5.1–5.8
<u>H</u> C—C (conjugated)	5.8–6.6
<u>H</u> ₂C≔C (isolated)	4.5–5.0
<u>H</u> ₂C—C (conjugated)	5.3–5.8
<u>H</u> C≕C (isolated)	2.4–2.7
<u>H</u> C≕C (conjugated)	2.7–3.1
Ar— <u>H</u>	6.5–8.0

### Intensities of the Signals, Peak Areas and Proton Counting

The intensities of the signals give the ratio of protons of each type in the molecule. Most NMR spectrometers have an integrator which integrates each peak (the area of each peak is found by counting the squares on the chart and is proportional to the number of protons in that signal).

Let us look at the NMR spectras of toluene and *p*-xylene (Figs. 25.12 and 25.13).

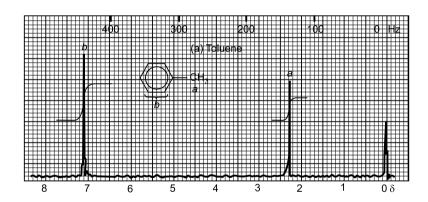


Fig. 25.12 NMR spectra of toluene

It is seen by examining the NMR spectras of toluene (Fig. 25.12) and p-xylene (Fig. 25.13) that the side chain protons (marked a) absorb at high field ( $\delta$  2.3) and the aromatic protons (marked b) absorb at lowfield ( $\delta$  7.0) and that the areas under the peaks for side chain and aromatic protons have sizes in the ratio 3:5 for toluene and 3:2 (or 6:4) for p-xylene. Thus, the area under an NMR signal is directly proportional to the number of protons giving rise to the signal.

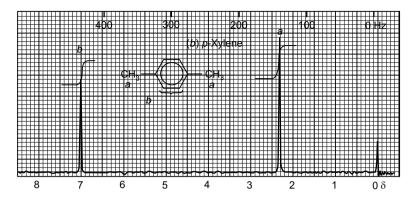


Fig. 25.13 NMR spectra of p—xylene

## The Splitting of Signals

The splitting of a signal into several peaks gives an idea about the environment of a proton with respect to other nearby protons. The understanding of the splitting of a signal is an important step for the structural assignment of an organic compound. Consider the NMR spectra of the three compounds. 1, 1,2–tribromoethane (CH<sub>2</sub>Br—CHBr<sub>2</sub>), 1, 1-dibromoethane (CH<sub>3</sub>-CHBr<sub>2</sub>) and ethylbromide (CH<sub>3</sub>CH<sub>2</sub>Br). Each of these contains only two kinds of protons. However, instead of two peaks, the spectra show five, six and seven peaks, respectively. This arises due to splitting of the NMR signals caused by spin-spin coupling. The signals we expect from each set of equivalent protons is appearing as a group of peaks (and not a single peak). Thus, splitting reflects the environment of the absorbing protons with respect to nearby protons. Let us consider, the case of adjacent carbon atoms carrying a pair

of secondary protons (b) and a tertiary proton (a) respectively 
$$\begin{pmatrix} a & b \\ -CH--CH_2 \end{pmatrix}$$
. First,

consider the absorption by one of the secondary protons. The magnetic field that a secondary proton (b) feels at a particular instant is slightly increased or decreased by the spin of the neighbouring tertiary proton (a), increased if the tertiary proton happens at the instant to be aligned with the applied field, or decreased if the tertiary proton happens to be aligned against the applied field. Thus, for half the molecules, absorption by a secondary proton is shifted slightly downfield and for the other half of the molecules the absorption is shifted slightly upfield. The signal is split into two peaks—a doublet with equal peak intensities.

Thus, we see that proton resonances are influenced through intervening bond by the local fields generated by the protons attached to adjacent carbon atoms. This effect is called **spin coupling**, meaning that the spins of nearby protons couple magnetically with each other. It is more appropriate to commonly use the word **spin-spin splitting** (for spin coupling) due to the appearance of the spectrum 298

itself, in which some signals are split into more than one peaks. In magnetically equivalent protons there is no spin coupling. The influence of neighbouring protons diminishes with distance. Only hydrogen atoms within three bond of each other (*i.e.*, H—C—C—H) have much effects. Coupling between nuclei separated by three bonds is the most common and is called **vicinial coupling**. Protons that are separated by only two bonds can also couple—this is known as **vicinal coupling**. The later type of coupling is observed for the protons of a terminal alkene with unsymmetrical substitution, R(R')C— $CH_2$ .

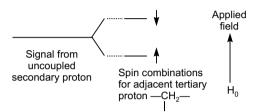


Fig. 25.14 Spin-spin coupling (coupling with one proton given a 1:1 double)

The absorption by the tertiary proton (marked *a*).  $- \overset{l}{\underset{a|}{\text{C}}} - \overset{b}{\underset{\square}{\text{CH}_2}} - \text{is affected by the}$ 

spin of the neighbouring secondary protons (marked *b*). In this case, the alignments of two secondary protons has to be considered. There are four equally probable combinations for these two protons, of which two are equivalent. Thus, at any instant, the tertiary protons feels any one of the three fields—this results in splitting of the signal (of tertiary H) into three equally spaced peaks (a triplet), with relative peak intensities 1:2:1 (Fig. 25.15).

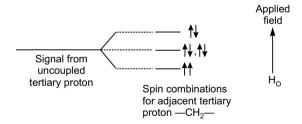


Fig. 25.15 Spin-spin coupling (coupling with two protons given a 1:2:1 triplet)

On the basis of the above, it can be said that a proton resonance involved in spin coupling will be split into n + 1 peaks, where n is the number of equivalent protons on the adjacent carbon atoms. This is known the n + 1 rule.

In case of spin-spin coupling, the value of the chemical shift is determined by finding the center of the feature. If the splitting pattern has an odd number of peaks,

the chemical shift corresponds to the central and tallest peak of the splitting pattern. In case, spin-spin splitting generates an even number of peaks, the chemical shift corresponds to a point halfway between the innermost peaks (Fig. 25.16).

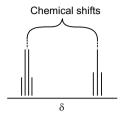


Fig. 25.16 Finding the chemical shift value in case of spin-spin coupling

The distance between peaks within a molecule is a measure of the effectiveness of spin-spin coupling, and is called the **coupling constant** (designated J), which has units of hertz (Hz) not ppm. This aspect forms the subject matter of a subsequent section (*see* page 310).

The ratios of the peak heights in a spin-spin coupling pattern will have one of the pattern given in Table 25.5.

n, No. of adjacent protons		n + 1, Feature	Intensity ratios
		1, singlet	1
	1	2, doublet	1:1
	2	3, triplet	1:2:1
3		4, quartet	1:3:3:1
4		5, quintet	1:4:6:4:1
5		6, sextet	1:5:10:10:5:1
	6	7. septet	1:6:15:20:15:6:1

Table 25.5 Intensities of peaks in the patterns caused by spin-spin splitting

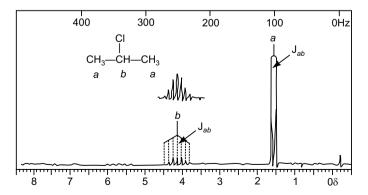


Fig. 25.17 NMR spectrum of 2-chloropropane

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Spin-spin splitting also occurs even when the protons are distributed over more than one carbon atom. As an example, in the NMR spectrum (*see* Fig. 25.17) of 2-chloropropane H<sub>3</sub>C—CH—CH<sub>3</sub>, the six protons of two methyl groups are

equivalent ( $\delta = 1.65$ ) and so the methyne proton (made in bold) is split into 6 + 1 = 7 peak (a septet) ( $\delta$  4.2).

It was stated earlier that 1,1,2-tribromoethane, 1,1-dibromoethane and ethylbromide contain two kinds of protons each. However, instead of two peaks, the spectra shows 5, 6 and 7 peaks respectively (*see* Figs. 25.18, 25.19 and 25.20).

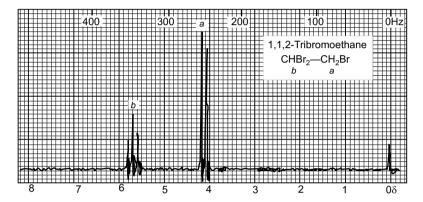


Fig. 25.18 NMR spectra of 1,1,2-tribromoethane

As seen in the above NMR spectra, the protons (marked a) absorb upfield as doublet ( $\delta$  4.2) and the proton (marked b) absorb downfield as a triplet ( $\delta$  5.7). The area of the triplet and doublet is 1:2.

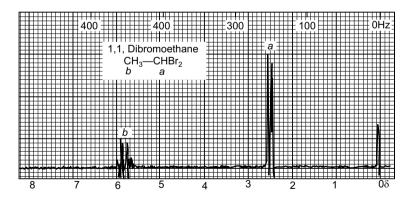


Fig. 25.19 NMR spectra of 1,1-Dibromoethane

In the case of 1,1-dibromoethane, the NMR spectra (Fig. 25.19) shows a upfield doublet ( $\delta$  2.5) of three protons due to coupling with proton (marked b)

and a downfield quartet ( $\delta$  5.8) of one proton due to coupling with three protons (marked a). The area of the quartet and triplet is 1:3.

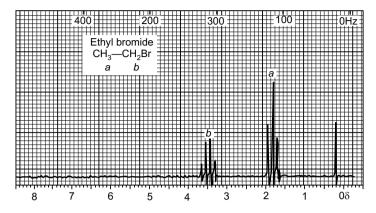


Fig. 25.20 NMR spectra of Ethylbromide

The NMR spectra of ethylbromide (Fig 25.20) shows a upfield triplet ( $\delta$  1.6) of three protons due to the coupling with two protons (marked b) and a downfield quartet ( $\delta$  3.3) of two protons due to coupling with three protons (marked a). The area of the quartet and the triplet is 2:3.

## **Coupling in Aromatic Compounds**

In case of aromatic compound like benzaldehyde or aniline, no coupling is observed between the proton attached to C=O group (in benzaldehyde) or between the two protons attached to N (in case of aniline). This is because the proton in benzaldehyde or aniline are separated from the protons attached to the benzene ring by more than three bonds. However, spin coupling occurs only between protons attached to the ring.

**Benzaldehyde:** Four bonds between H atom of CHO and H atom of benzene ring. So, no spin coupling.

**Aniline:** Four bonds between the two equivalent H atoms of NH<sub>2</sub> group and H atom of benzene ring. So, no spin coupling.

$$H_b$$

**Benzene:** The two H atoms (marked  $H_a$  and  $H_b$ ) are separated by three bonds. So, these protons engage in spin coupling.

In case of di and trisubstituted benzenes, spin coupling occurs. Thus in case of para disubstituted benzene (where the substituents x and y do not contain H),

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there is spin-spin coupling between the  $H_a$  and  $H_b$  protons. Both these protons will appear as doublets.

$$H_a$$
 $H_b$ 
 $H_a$ 
 $H_b$ 
 $H_a$  and  $H_b$  appear as doublet (1:1).

In the case of 1,2,3-trisubstituted benzene, there are three non-equivalent protons, marked  $H_a$ ,  $H_b$  and  $H_c$ . The  $H_a$  and  $H_c$  protons will appear as a doublet and  $H_b$  proton will appear as a triplet.

1,2,3-Trisubstituted benzene: The two protons 
$$H_a$$
 and  $H_c$  will appear as a doublet (1:1) each and the  $H_b$  proton will appear as a triplet (1:2:1).

In the case of 1,2,4-trisubstituted benzene, there are three non-equivalent protons, marked  $H_a$ ,  $H_b$  and  $H_c$ . The  $H_a$  and  $H_b$  protons will appear as a doublet each and the  $H_c$  proton will appear as a singlet.

1,2,4-Trisubstituted benzene: The two protons 
$$H_a$$
 and  $H_b$  will appear as a doublet (1:1) each and the  $H_c$  proton will appear as a singlet.

## **Coupling Constant**

It has already been stated that the distance between the peaks in a multiplet (arising out of spin-spin coupling) is a measure of the effectiveness of spin-spin coupling, and is called a **Coupling Constant** (designated as J), which has units of hertz (Hz). The value of the coupling constant remains the same irrespective of the applied magnetic field (*i.e.*, whatever the radiofrequency used). Thus, in this respect, the spin-spin coupling differs from chemical shift.

Most of the aromatic or aliphatic protons engaged in vicinal coupling with each other, the J values are about 7 Hz. However, in case of alkenes, alkylenes or molecules containing aldehyde group, the J values varies. Typical J values for a number of systems is given in Table 25.6.

System	Typical J <sub>ab</sub> Value (Hz)	System	Typical J <sub>ab</sub> Value (Hz)	System	Typical J <sub>ab</sub> Value (Hz)
H <sub>a</sub> C—C	2	H <sub>a</sub> C—C—	7	$H_a$ C=C $H_t$	15
H <sub>a</sub> C=C H <sub>b</sub> (geminal coupling)	2	H <sub>a</sub> C—C C—C	6	$H_a$	0–1
H <sub>a</sub> C C C (four bond coupling)	-H <sub>b</sub> 2	H <sub>a</sub> C=C H <sub>t</sub>	8	H <sub>b</sub> C—C—C—	С—Н <sub>а</sub> 0
H <sub>a</sub> (four bond coupling)	<sub>b</sub> 2	$H_a$	8		

**Table 25.6** Typical J values for some common systems (showing spin-spin coupling)

In any pair of geometric isomers, the J value is always larger between *trans* protons than between *cis* protons. Also, the size of J varies in a regular way with the electronegativity of subsitutents, so that using the J values, it is possible the assign configuration without having both isomers in hand. In fact, J value is a most important feature of an NMR spectrum.

# 25.1.6 Final Interpreting an <sup>1</sup>H NMR Spectra

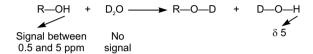
It has already been stated that for interpretation of an <sup>1</sup>HNMR spectra one should find

- the number of signals.
- the position of signals.
- the intensities of the signals and
- the splitting of the signals.

For final interpretation of an <sup>1</sup>H NMR spectra follow the following steps:

(i) First of all determine the chemical shift for each peak by referring to Tables 25.1, 25.2 and 25.3.

- 304
- (ii) Determine the integrated value for each peak.
- (iii) Find out if the molecule is a carboxylic acid, alcohol, phenol, amide or a thiol. This can be ascertained by refering to Table 25.1 in the first instance. Then, the protons attached to the above group usually undergo deuterium exchange on treatment with D<sub>2</sub>O. To find the presence of such groups, a drop of D<sub>2</sub>O is added to the NMR sample. The disappearance of a signal implies that the group responsible for that signal usually has its protons attached to a hetero atom. Alcohols (R OH), phenols (Ar OH) and carboxylic acids (R COOH) undergo exchange rapidly. An example is given below:



Amides (R CONH<sub>2</sub>) and thiols (R SH) normally need treatment with a base like NaOD in D<sub>2</sub>O for the exchange. Since DOH formed is slightly soluble in most organic solvents, its signal ( $\delta$  5) will also be observed.

- (iv) Assign multiplicity m, of each peak (2,3,4...etc., for doublet, triplet, quartet, etc.).
- ( $\nu$ ) From the value of m, calculate the number of neighbouring protons, n using the equation.

$$n = m - 1$$

As an example for a triplet the neighbouring carbon will have two protons and for a quartet, the neighbouring carbon will have three protons.

$$CH_3$$
— $CH_2$ —

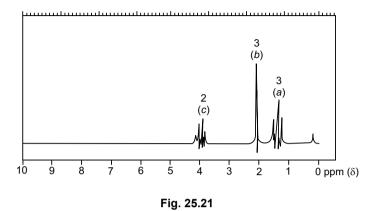
- (vi) Find the feature with the same J value that has n protons (keeping in mind the integrated intensity value). These two groups must be adjacent.
  Make use of the pattern illustrated for benzaldehyde, aniline, benzene, o-, m- and p-disubstituted benzenes given on page 309-310. This will be helpful to identify common structural fragments.
- (vii) Simple NMR spectras that have only alkyl and or benzene protons, all the multiplets must have J values around 7 Hz.

The preedure for interpretation is made clear by the following example:

1. A compound, molecular formula, C<sub>4</sub>H<sub>8</sub>O<sub>2</sub> gives the following <sup>1</sup>H NMR spectra (Fig 25.21). Suggest the structure of A.

The NMR spectra of the compound A shows three peaks (designated a, b and c). On the basis of integration, the number of protons in a, b and c are 3:3:2 since the sum of these numbers (3+3+2) is 8, the same number of H atoms are given in

the molecular formula of A. So, these numbers (3, 3, 2) correspond to the actual number of protons present in the peaks (a, b, c).



Since all the signals are between 0 and 5 ppm so the molecule A has only aliphatic protons. The ratio of protons in the triplet (peak a) and quartet (peak c) is 3:2. This most likely shows the presence of an ethyl group. The furthest downfield signal (c) has a chemical shift ( $\delta$  4.1) indicating the presence of a methylene group

The signal b ( $\delta$  2.07) having an integrated intensity of 3 implies the presence of a CH<sub>3</sub> group. Its chemical shift value ( $\delta$  2.07) suggest that most likely the methyl group is adjacent to a carbonyl group.

bonded to an oxygen atom. So, the molecule A has the group OCH<sub>2</sub>CH<sub>3</sub>.

On the basis of the above, the compound A is most likely ethyl acetate,

There is another reasoning, by which it can be said that the methyl group (peak b) is bonded to oxygen. The chemical shift value ( $\delta$  2.07) of the signal b (having an integrated intensity of 3) suggest that the methyl group is bonded is an atom which has no proton. Adding the fragments  $OCH_2CH_3$  and  $CH_3$  (about which we are sure) gives the molecular formula  $C_3H_8O$ . This formula differs from the given formula  $(C_4H_8O_2)$  by CO  $[C_4H_8O_2 - C_3H_8O = CO]$ , which is a carbonyl group. Using this procedure, it is concluded that the compound A has  $CH_3$ ,  $OCH_2CH_3$  and C=O groups, which gives the formula  $CH_3 - CO - OCH_2CH_3$  or ethyl acetate.

2. An organic compound B has the molecular formula  $C_8H_9Br$ . It gives the following <sup>1</sup>HNMR spectrum (Fig. 25.22) in a 300 MHz instrument. The spin-spin splitting pattern in all peaks is J=7 Hz. Give structure to the compound B.

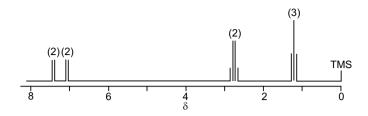


Fig. 25.22

- 1. In the above  $^{1}$ HNMR spectrum, the absorptions are in the region  $\delta$  7-7.5 and  $\delta$  1-3 indicating that the compound has both aromatic and aliphatic protons. This is also supported by the J value (7 Hz) of all the peaks.
- 2. The integrated intensity values for peaks from left to right is 2:2:2:3. The sum equals to 9, which is the actual number of protons in the given molecular formula (C<sub>8</sub>H<sub>0</sub>Br).
- 3. Two doublets in the aromatic region correspond to 4 protons; this corresponds to substitutions are attached to *para* position with respect to each other (*see p*-disubstituted benzene, page 310).
- 4. In aliphatic region ( $\delta$  1-3), the appearance of a triplet and a quartet in the ratio 3:2 indicates the presence of an ethyl group.
- 5. Thus, the compound B contains an aromatic ring (C<sub>6</sub>H<sub>4</sub>) and CH<sub>2</sub>CH<sub>3</sub> group and a Br atom.

On the basis of the above, the compound B is p-bromethylbenzene,  $p(Br) C_6H_4CH_2CH_3$ .

# 25.1.7 Interpretation of the <sup>1</sup>H NMR Spectra of Some Simple Molecules

### **Ethanol**

The <sup>1</sup>H NMR spectra of ordinary ethanol shows hydroxyl proton as a singlet ( $\delta$  3.9) and the protons of CH<sub>2</sub> group as a quartet ( $\delta$  3.7) (Fig. 25.23). In ordinary ethanol no signal splitting arising from coupling between the hydroxyl proton on the protons of the —CH<sub>2</sub>— group is observed, even though they are on adjacent atoms.

However, in the  $^1H$  NMR spectra of very pure ethanol, the signal from hydroxyl proton ( $\delta$  3.9) is split into a triplet, and that the singal from the protons of the —CH<sub>2</sub>—group is split into a multiplet of eight peaks. Thus, in very pure ethanol, the spins of the proton of the hydroxyl group is coupled with the spins of the protons of the —CH<sub>2</sub>—group.

The area under peaks a, b and c is in the ratio 3:2:1. In the above spectra (Fig. 25.23), the peak b for  $CH_2$  is shown as a quartet. However, it is a multiplet of eight peaks.

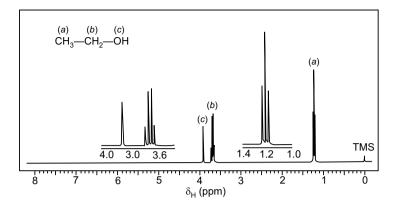


Fig. 25.23 <sup>1</sup>H NMR spectra of pure ethanol on a 300 MHz instrument

## Acetaldehyde

The NMR spectra of acetaldehyde,  $CH_3$ — $C_0$  shown a doublet centered at  $\delta$  2.14 corresponding to 3H(a) protons of methyl group (downfield due to carbonyl group) and a quartet at  $\delta$  9.78 due to 1H(d) corresponding to aldehydic proton (Fig. 25.24).

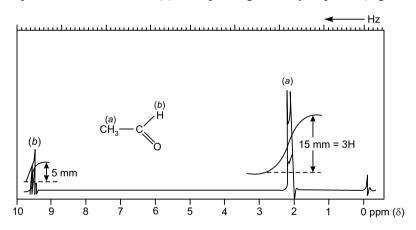


Fig. 25.24 <sup>1</sup>H NMR spectra of Acetaldehyde

### Acetophenone

The NMR spectra of acetophenone,  $c = \begin{pmatrix} c & b & O \\ c & -C & -C \\ b & c \end{pmatrix}$  is expected to give a singlet at  $\delta$  2.47 due to 3H (a) of methyl protons. Two downfield multiplets at  $\delta$  = 7.50

and 7.9 represent aromatic protons (c) due to 2H (m), 1H (p) and 2H ortho position (b), respectively (Fig. 25.25).

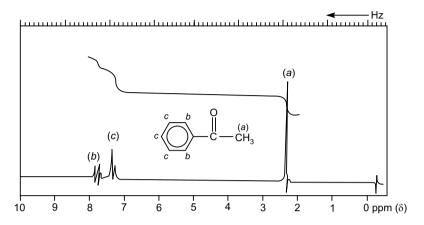


Fig. 25.25 <sup>1</sup>H NMR spectra of Acetophenone

## 25.1.8 Predicting the <sup>1</sup>H NMR Spectrum of an Organic Compound

Using the knowledge gained so for, the <sup>1</sup>H NMR spectra of an organic compound can be predicted. Two examples are given below.

(i) p-Methoxybenzaldehyde: p-Methoxybenzaldehyde has three parts: aldehyde group ( $\delta$  10), aromatic ring ( $\delta$  7—8) and an aliphatic group ( $\delta$  0—5) (use data given in Table 25.1).

Using the data given in Table 25.3, assign more specific values of chemical shifts and the integrated intensity value: CHO ( $\delta$  10, 1H), Ar H ( $\delta$  7.8, 4H) and Ar—CH<sub>3</sub> ( $\delta$  2.3, 3H).

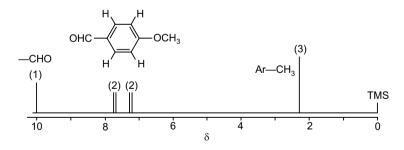


Fig. 25.26

Finally take into account the spin-spin coupling. The aldehyde proton will appear as a singlet since it has no other proton within three bonds. The aromatic protons of *p*-disubstituted ring will appear as two doublets with integrated

intensity value of 2 H. Finally, the methyl group will appear as a singlet since it has no other proton within three bonds. So, the expected <sup>1</sup>H NMR spectrum of *p*-methoxybenzaldehyde is (Fig. 25.26).

(ii) Isopropyl Methyl Ether: In isopropyl methyl ether all protons are aliphatic and will appear in the range δ 0—5. Using the Table 25.2, to find the precise chemical shift values and the intensity values of the different substituents: —CH—O—R (δ 3.7, 1H), —CH<sub>3</sub>—O—R (δ 3.3, 3H), (CH<sub>3</sub>)<sub>2</sub>—C—O (δ 1.5, 6H).

Finally take into account spin-spin coupling the protons of the isopropyl group [(CH<sub>3</sub>)<sub>2</sub>CH] will appear as a septet and doublet and the protons of the methyl group will appear as a singlet. Thus, the following <sup>1</sup>H NMR spectrum is expected (Fig. 25.27).

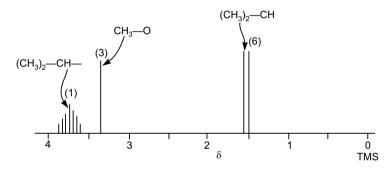


Fig. 25.27

# 25.1.9 Complicated <sup>1</sup>HNMR Spectra

Most of the <sup>1</sup>H NMR spectras are more complicated than the ones discussed so far. These are first interpreted in the same way as discussed earlier, *i.e.*, to find how many protons the molecule has, how many kinds of protons the molecule has and so on. Some times it is not possible to decide whether a molecule contains two methyl group or one ethyl group. Instrumental techniques are now available which help to analyses a complicated <sup>1</sup>H NMR spectra. One such technique is **double resonance** or **double irradiation**. In this technique, the spins of two sets of protons can be decoupled and a simpler spectrum is obtained. The molecule is irradiated with two radio frequency beams. The usual one whose absorption is being measured and a second, much stronger beam whose frequency differs from that of the first. In the second case, when the field strength is reached at which the protons in which we are interested absorbs and gives a signal, the splitting protons are absorbing the other

310

very strong radiation. In this way, the spins are decoupled and the signal appears as a simple, unsplit peak.

Another useful technique is the use of **deuterium labelling**. Since deuterium has a much smaller magnetic moment than a proton, it absorbs at a much higher field and give no signal in the <sup>1</sup>H NMR spectrum. So, the replacement of a proton by deuterium removes from the <sup>1</sup>H NMR spectra both the signals from that proton and the splitting by it of signals of other protons. In this case, it looks as there were no hydrogen at that position in the molecule. An interesting example is:

In this way, deuterium labelling is used to find out which signal is produced by which proton or protons. The disappearance of a particular signal is observed when a proton in a known location is replaced by deuterium.

## Two-Dimensional (2D) NMR Spectroscopy

The interpretation of NMR spectra is greatly simplified using multi-dimensional NMR spectroscopy, particularly two-dimensional (2D) NMR spectroscopy. This type of spectroscopy require an NMR spectrometer of the pulse (Fourier transform) type. The two commonly used 2D NMR spectroscopy are known as COSY ( $^{1}H_{-}^{1}H$  correlation spectroscopy) and HETCOR (2D NMR spectra which indicate coupling between hydrogens and the carbons to which they are attached; the word HETCOR stands for heteronuclear correlation spectroscopy (or C—H HETCOR). These two techniques are very helpful when ambiguities are present in one-dimensional  $^{1}H$  and  $^{13}C$  NMR spectra (the  $^{13}C$  NMR spectroscopy forms the subject-matter of a subsequent section).

### COSY

In a COSY spectra, the ordinary one-dimensional <sup>1</sup>H spectra of a compound is shown along both the horizontal and vertical axis. The COSY spectra of 1-chloro-2-propanol CICH<sub>2</sub>—CH—CH<sub>3</sub>) is given in the Fig. 25.28. The important and new

information from COSY spectrum is obtained from the correlation peaks that appear off the diagonal (called "cross peaks").

In case of 1-chloro-2-propanol, even though the usual  ${}^{1}H$  NMR spectra is readily interpreted. However, the COSY spectra of this compound should be understood as an example. A good reference point in the COSY spectrum of 1-chloro-2-propanol (Fig. 25.28) is the peak for methyl group (which appears as a doublet at  $\delta$  1.2). This methyl peak from both the horizontal and vertical axis is drawn to meet at the diagonal line at a point labelled  ${}^{c}H$ . An imaginary line can be drawn parallel to the vertical axis that intersects a correlation peak (labelled  ${}^{b}H$ – ${}^{c}H$ ) in the X—Y

field off the diagonal from this point, a perpendicular line can be drawn back to its intersection with the diagonal peaks. At its intersection, we observe this diagonal peak is directly above the one-dimensional peak at  $\delta$  3.9. Thus, we say that the methyl hydrogens at  $\delta$  1.2 are coupled to the hydrogens whose signal appear at  $\delta$  3.9. So, the peaks at  $\delta$  3.9 are due to the hydrogen on the alcohol carbon in 1-chloro-2-propanol ( ${}^bH$  on  $C_2$ ).

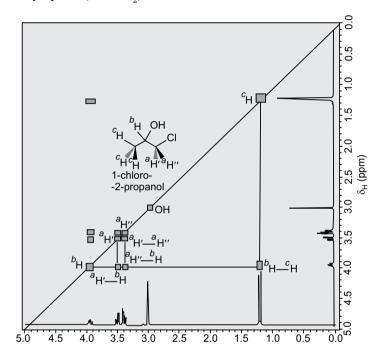


Fig. 25.28 COSY spectrum of 1-chloro-2-propanol

(Source: Organic Chemistry, G. Soloman and C. Fryhle)

On close examination of the COSY spectra (Fig. 25.28), it is seen that hydrogens whose signal appear at  $\delta$  3.9 is coupled to the hydrogens whose signals appear at  $\delta$  3.4 and  $\delta$  3.5. Thus, the hydrogens at  $\delta$  3.4 and  $\delta$  3.5 couple with each other by the cross peak; they have in common between them right next to their diagonal peaks.

Thus, using COSY spectrum, one can see which hydrogens are coupled to each other.

Answer

### Solved Problems Based on <sup>1</sup>H NMR

(i) How many kind of hydrogens are there in

(a) CH <sub>3</sub> CH <sub>3</sub>	one (all equivalent)
(b) CH <sub>3</sub> CH <sub>2</sub> CH <sub>3</sub>	Two CH <sub>3</sub> CH <sub>2</sub> CH <sub>3</sub>

(c) 
$$(CH_3)_2CHCH_2CH_3$$
 ... Four  $(CH_3)_2CHCH_2CH_3$  ... One (all equivalent)

(e) 
$$CH_3CH=CH_2$$
 ...  $H_3C = C = C H_d$  (Four)

In this case, the hydrogens  $H^c$  and  $H^d$  are not equivalent, since one is *cis* to the  $CH_3$  and the other in *trans*. Replacement of  $H^c$  by X gives the *cis* diastereomer. Replacement of  $H^d$  gives the *trans*-diastereomer.

(f) 
$$C_6H_5NO_2$$
 Three: Two ortho, two meta and one para

$$c \bigotimes_{b=a}^{b} NO_2$$

(g) 
$$C_6H_5$$
— $CH_3$  Two: the ring hydrogens are little affected by alkyl groups and are equivalent

$$b \stackrel{b}{\longleftrightarrow} CH_3$$

$$\begin{array}{c} {\rm CH_3} & {\rm CH_3} \\ {\rm I} & {\rm CH_3} \\ {\rm C} - {\rm OCH_3} & {\rm Two.~CH_3} - {\rm C} - {\rm OCH_3} \\ {\rm I} & {\rm CH_3} \\ {\rm CH_3} & {\rm CH_3} \\ \end{array}$$

(ii) How many kinds of non-equivalent hydrogens are there in the following(a) p-CH<sub>3</sub>CH<sub>2</sub>C<sub>6</sub>H<sub>4</sub>CH<sub>2</sub>CH<sub>3</sub>

Ans. Three 
$$H_3^a CCH_2 \longrightarrow H_2^b CH_2 - CH_3$$

The methylene hydrogens are enantiotopic and are magnetically equivalent.

(b)  $Cl_2CH CH_2CH_2CH_2Cl$ 

**Ans.** Four 
$$Cl_2CH - CH_2 - CH_2 - CH_2CI$$

(c) CH<sub>3</sub>CHCl CH<sub>2</sub>CH<sub>3</sub>

Ans. Five

The methylene hydrogens  $H^c$  and  $H^b$  are not equivalent because of the presence of chiral carbon. Replacing these diastereotopic hydrogens by X gives two diastereomers.

(iii) Give the structure for each of the following compounds:

(a) C<sub>2</sub>H<sub>4</sub>Cl<sub>2</sub>, only one kind of H

**Ans.** ClCH<sub>2</sub>CH<sub>2</sub>Cl: the isomeric 1, 1-dichloroethane has two kinds of hydrogens.

(b) C<sub>8</sub>H<sub>18</sub>, only one kind of H

**Ans.** (CH<sub>3</sub>)<sub>3</sub> C—C(CH)<sub>3</sub>

(c)  $C_0H_{12}$ , two type of hydrogens.

Ans. 1,3,5 Trimethyl benzene (mesitylene)

(d) Non-cyclic C<sub>6</sub>H<sub>12</sub>, only one kind of H

Ans. One double bond; all H's must be in Me's

$$(CH_3)_2C = C(CH_3)_2$$

(e) C<sub>3</sub>H<sub>7</sub>Cl, three kinds of H

**Ans.** CH<sub>3</sub>CH<sub>2</sub>CH<sub>2</sub>Cl; the isomeric 2-chloropane CH<sub>3</sub>CHCl CH<sub>3</sub> has only two kinds of hydrogen.

(iv) Explain the splitting pattern for the unit  $\begin{bmatrix} 1 & b \\ -CH - CH_3 \end{bmatrix}$ 

**Ans.** The single  $H^a$  split  $H^b$  into a doublet integrating for three protons.  $H^a$  appears as quartet due to splitting by three protons of —CH<sub>3</sub>; the entire quartet integrates for one H.

- (v) In which of the following molecules does spin-spin coupling occur? In case splitting is observed give the multiplicity of each kind of H.
  - (a) ClCH<sub>2</sub>CH<sub>2</sub>Cl splitting is not observed; it has only one equivalent proton.
  - (b) ClCH<sub>2</sub>CH<sub>2</sub>I the H's of CH<sub>2</sub> are non-equivalent and each is split into a triplet.

(c) 
$$\frac{H}{Br}C=C$$
  $\frac{H}{Br}$  Splitting is not observed; it has only one equivalent H.

- (d)  $\frac{H}{Br}C=C \frac{CI}{H}$  The two hydrogens are not equivalent, and each is split into a doublet.
- (e) CI C=C H The vinylic hydrogens are non-equivalent since one is *cis* to Cl and the other is *cis* I. Each is split into a doublet; in this case the interacting hydrogens are on the same carbon.
- (f) CH<sub>3</sub>CH<sub>2</sub>—CH<sub>2</sub>CH<sub>3</sub> Gives a singlet for the four equivalent, equivalent uncoupled H's, a quartet for the H's of two equivalent CH<sub>2</sub> coupled to CH<sub>3</sub>, and a triplet for the two equivalent CH<sub>3</sub> groups coupled with CH<sub>2</sub>.
- (vi) In neopentyl chloride (1-chloro-2, 2-dimethyl propane)
  - (A) there is no splitting but in 2-methyl propene. (B) there is splitting, explain why?

**Ans.** Although  $H^a$  and  $H^b$  in (B) and are not on adjacent C's, they are close enough to couple due to shorter C=C bond. However, in case of A,  $H^a$  and  $H^b$  are on non-adjacent C's are too far away to couple, the C's being joined by longer single bond.

(vii) How will you find out whether a particular signal arises from the following groups, —OH, —NH, —SH.

**Ans.** Add excess  $D_2O$ , shake the tube and rerun the spectrum. The H's in these groups is replaced by D's.

$$\mathbf{R} \mathbf{\longrightarrow} \mathbf{OH} + \mathbf{D_2O} \mathbf{\longrightarrow} \mathbf{R} \mathbf{\longrightarrow} \mathbf{OD} + \mathbf{DOH}$$

Since D does not resonate under the conditions used for H, the original peak for —OH (or —NH or —SH) disappears in the spectrum rerun in D<sub>2</sub>O. A New signal for DOH appears.

(*viii*) The three chlorobromo ethenes (C<sub>2</sub>H<sub>2</sub>BrCl) (A), (B) and (C) show two doublets. How will you differentiate between these

**Ans.** These can be differentiated by their J values which are 0.3 Hz (for A), 7–12 Hz (for B) and 13–18 Hz for (C).

(*ix*) How will you differentiate between ethane, ethylene and acetylene by NMR spectroscopy?

**Ans.** Each of these compounds contains only one type of proton and so it will show only one NMR signal. However, the environment of the proton in each compound is different. So, the relative position of the signals in each case will be different (as given below):

$H_3C$ — $CH_3$	δ 0.9	
$H_2C = CH_2$	δ 5	
НС≡СН	δ 2.5	

Thus, it is possible to differntiate between the three hydrocarbons.

(x) Give a structure of the compound consistent with the following data:

Molecular formula C<sub>8</sub>H<sub>9</sub>Cl

NMR data

Triplet,	δ 2.7 (2H)	
Triplet,	δ 3.4 (2H)	
Singlet,	δ 7.22 (5H)	

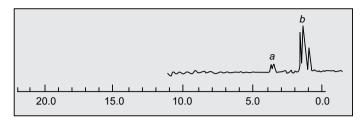
**Ans.** The compound contains three kinds of protons.

The singlet at  $\delta$  7.22 (5H) is due to a monosubstituted benzene ring. The side chain,  $C_2H_4Cl$  is attached to benzene ring consists four protons in the form of two different types. So, the structure is

(xi) How many NMR signals will be obtained in the <sup>1</sup>H NMR spectrum of methoxyacetonitrile, CH<sub>3</sub>OCH<sub>2</sub>CN?

**Ans.** No signal splitting occurs from enantiotropic protons OCH<sub>2</sub>CN of methoxyacetonitrile. So methoxyacetonitrile will give two <sup>1</sup>H NMR signal one each from CH<sub>3</sub> and CH<sub>2</sub>.

(xii) The <sup>1</sup>H NMR spectra of 1-heptanol gives the only interpetable signal of methylene adjacent to OH ( $\delta$  3.8 triplet) and the terminal methyl ( $\delta$  0.9 triplet)



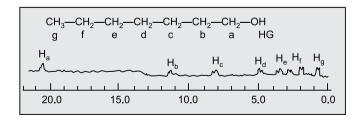
(a)  $\delta$  3.8(t) CH<sub>2</sub> adjacent to OH (b)  $\delta$  0.9(t) terminal CH<sub>3</sub>

The above spectra does not give information about the structure of the compound. How will you obtain the correct structure.

**Ans.** The detailed spectra of 1-heptanol can be obtained by the addition of **shift reagents**. The shift reagents are ions in the rare earth (lanthanide) series, coordinated to organic ligands. Two such reagents commonly used are tris-(dipivalomethanato) euronium  $[Eu(dpm)_3]$  and tris-1,1,1,2,2,3,3-heptafluoro-7,7-dimethyl-3,5-octane dionatoeuropium  $[Eu(fod)_3]$ .

$$\begin{array}{c} R \\ CH \\ CH \\ H_3C - C - CH_3 \\ CH_3 \end{array} \qquad \begin{array}{c} Eu(dpm)_3 \quad R = t\text{-butyl} \\ Eu(fod)_3 \quad R = CF_2CF_2CF_3 \\ CH_3 - CH_3 \\ CH_3 - CH_3 - CH_3 \\ CH_3 - CH_3 - CH_3 - CH_3 \\ CH_3 - CH_3 - CH_3 - CH_3 - CH_3 \\ CH_3 - C$$

An addition of the shift reagent, Eu(dpm)<sub>3</sub>. The signals of the methylene group closer to the OH group are moved downfield so that a separate signal is obtained for each of the methylene units.



Simplified spectra of 1-heptanol using shift reagent. Eu(dpm)<sub>3</sub>

While using shift reagent it is necessary to report the solvent and the concentration of the substrate and shift reagent. In the above case 0.40 mL of CDCl<sub>3</sub> solution containing 0.300 M 1-heptanol and Eu(dpm)<sub>3</sub> (0.79 moles per mole) (1-heptanol), temp. 30°C was used.

Thus, the use of shift reagent simplifies the spectrum.

# 25.1.10 Applications of Proton Magnetic Resonance Spectroscopy

(i) Structure elucidation of organic compounds. On the basis of the NMR spectrum of a compound, it is possible to find.

The number of signals, which indicate how many kinds of protons are present in the compound.

Position of signals give us an idea of the nature of protons i.e., whether they are aromatic, aliphatic, primary, secondary, tertiary, benzylic, vinylic, acetylenic and adjacent to halogens or other hetero atoms or functional groups.

Intensities of the signals give the ratio of each type of protons in the molecule.

Splitting of the signals gives us an idea about the environment of a proton with respect to other nearby protons.

On the basis of the above, it is possible in most of the cases to assign the structure to a compound.

(ii) Distinction between isomeric structures can be made on the basis of NMR spectra. Thus, a compound having molecular formula C<sub>3</sub>H<sub>7</sub>Cl can be either propyl (I) or isopropyl chloride (II).



The NMR spectra of propyl chloride will give three signals: 3H, t, methyl protons; 2H sextet methylene protons; and 2H, t, chlorine bearing methylene protons. On the other hand, isopropyl chloride gives only two signals, 6H, d, two methyl groups; and 1H, septet methine proton.

(iii) Determination of relative amounts of compounds in a mixture. For example, in a mixture of aspirin and phenacetin, the relative amounts of each can be determined as molar ratio

$$\begin{array}{c} O \\ O \\ C \\ C \\ COOH \end{array}$$

$$\begin{array}{c} O \\ I \\ OC_{2}\\ H_{5} \\ \end{array}$$

$$\begin{array}{c} O \\ I \\ OC_{2}\\ H_{5} \\ \end{array}$$

$$\begin{array}{c} O \\ I \\ OC_{2}\\ H_{5} \\ \end{array}$$

$$\begin{array}{c} O \\ I \\ OC_{2}\\ I \\ OC_{2}\\ \end{array}$$

In aspirin and phenacetin the acetyl protons appear as singlets at  $\delta$  2.3 and  $\delta$  2.1, respectively. The integration showed the two peaks in the ratio 1:1. Therefore, aspirin and phenacetin are present in equimolar amounts.

(iv) Distinction between cis and trans isomers. NMR is helpful to determine the cis and trans isomers of alkenes. Thus, 1-bromo-2-chloro ethylene can exist as cis and trans forms.

The protons in the *trans* structure couple more strongly with a J value of 16 Hz; the *cis* coupling constant is generally half of this value, about 8 Hz; it is to be noted that coupling between terminal methylene

much smaller of the order of about 3 Hz.

(v) In a chemical reaction, performed in the laboratory, it is possible to find the structure of the product. As an example, oxidation of benzyl alcohol with MnO<sub>2</sub> gives benzaldehyde. Thus, in the <sup>1</sup>H NMR spectrum of the formed product, the signals for CH<sub>2</sub>OH (a doublet due to CH<sub>2</sub> and a triplet for H) will disappear and a signal for aldehydic proton (δ 9—10) will appear.

In a similar way, in the formation of isopropyl methyl ether in the following Williamson's ether synthesis:

$$\rightarrow OH \xrightarrow{1. \text{ NaH, DMF}} \xrightarrow{H_3C} H_2C -O - CH_3$$

The signal for H of OH will disappear and a signal ( $\delta$  3.7, heptat) will appear. Besides a signal for CH<sub>3</sub> will appear.

(vi) Differentiation between the three possibe isomers of a compound,  $C_2H_2$  BrCl.

The protons of the *trans* isomer (3) couple strongly with J value of 16 Hz. The *cis* compling constant is normally half this value (about 8 Hz) (isomer 2). However, coupling between terminal methylene proton (geminal coupling) is much smaller (about 3Hz) (isomer 1)

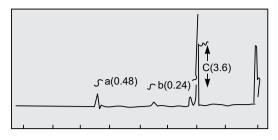
(vii) Characterisation of vinyl acetate on the basis of the NMR spectra

Vinyl acetate has three hydrogens ( $H_a$ ,  $H_b$  and  $H_c$ ) on the double bond. Each of these protons displays a different chemical shift ( $\delta$  4.5, 5.0 and 7.2 for  $H_a$ ,  $H_b$  and  $H_c$  respectively) and a different coupling costant to each at the other protons  $J_{bc}$  (trans) in larger than  $J_{ac}$  (cis) and  $J_{ab}$  (gemimal) is very small]. The CH<sub>3</sub> proton at vinyl actate appear at  $\delta$  2.1.

(ix) Differentiation between cis and trans stilbene.

In *trans* stilbene, the two olefinic protons ( $H_A$ ) are deshielded by both aromatic rings. However, in the *cis* stilbene, each of the hydrogen ( $H_B$ ) is deshielded by only one adjacent aromatic ring. Therefore  $H_A$ 's appear at a lower field ( $\delta$  7.0) than  $H_B$ 's ( $\delta$  6.0).

(x) Determination of the relative amounts of the tautomers in a tautomeric mixture



NMR spectra of acetyl acetone

Signals for keto and enol forms occur separately. From the integrated intensities of Keto and enol forms (assuming that the integral in the methyl region is six protons), the percentage of keto forms can be calculated as shown:

% Keto form = 
$$\frac{3 \times b}{c} \times 100 = \frac{3 \times 0.24}{3.6} = 20$$
  
%Enol =  $\frac{6 \times a}{c} \times 100 = \frac{6 \times 0.48}{3.6} = 80$ 

## (xi) Study of Rotational Isomerism.

In 1,2-dibromnoethane, BrCH<sub>2</sub>—CH<sub>2</sub>Br, all the four protons are chemically equivalent and it is expected that all the four protons will give the same chamical shift. However, it we consider Newman diagram, it is found that the proton (marked astrick) is flanked by H and Br in conformation (a), whereas in conformation (b) the said proton is flanked by H and H. In case, the NMR spectrum is recorded at lower temperature, the rapid rotation araound C—C is reasonably frozen for these differences to be observed.

### (xii) Influence of chirality on the chemical shifts of the enantiomers

The <sup>1</sup>H NMR spectra of enantiomers are identical and are exactly the same as that of the racemate. However, by using otpically active solvents, it has become possible to engender in a racemate a chemical shift between the chemical shifts of D and L-forms. As an example, a solution of phenyl isopropyl carbinol in D-1-naphthylethyl amine, two intense doublets are observed for the resonance of the tertiary proton

(H<sub>t</sub>). Their separation is of the order 1.6 and 2.5 Hz at 60 and 100 MHz respectively. The splitting is the result of a difference in resonance frequencies as the spin-spin coupling is independent of the of the field strength

The different spectra of the enantiomers in presence of an optically active solvents is due to the formation of diastereomeric complexes

# 25.2 CARBON-13 NMR (13C NMR) SPECTROSCOPY

### 25.2.1 Introduction

Like the proton, one of the isotopes of carbon, <sup>13</sup>C also has spin number 1/2. It given NMR spectra commonly known as carbon-13 NMR (<sup>13</sup>C NMR) spectra. The principles involved in the generation of <sup>13</sup>C NMR spectrum is the same as in the case of <sup>1</sup>H NMR spectrum. A special feature of <sup>13</sup>C NMR spectra is that each carbon atom (non-equivalent) in an organic molecule produces only one <sup>13</sup>C NMR peak. This greatly simplifies the interpretation procedure. It is to be noted that the major isotope of carbon, <sup>12</sup>C, with natural abundance of about 99% has no net magnetic spin and so cannot produce NMR signals.

We have known that in <sup>1</sup>H NMR spectra, hydrogen that is near to other hydrogen (the two hydrogen can be separated within a few bonds) couple with each other causing the signal for each hydrogen to give rise to a multiplet of peaks. In case of <sup>13</sup>C NMR spectra, this multiplicity of peaks due to neighbouring carbon does not occur. This is because only one carbon atom out of 100 carbon atoms is a carbon-13 nucleus (1% natural abundance). Thus, the probability of two <sup>13</sup>C atoms being adjacent to each other in a molecule is only about 1 in 10,000, thereby practically eliminating of possibility of two neighbouring carbon atom splitting each others signal into a multiplet of peaks. The sensitivity of modern spectrometers (particularly pulse FT NMR spectrometers, where signal averaging is possible) makes the low level (1%) sufficient for the measurement of <sup>13</sup>C NMR spectra.

Though carbon-carbon signal splitting does not occur in <sup>13</sup>C NMR spectra, hydrogen atoms attached to the carbon split the <sup>13</sup>C NMR signals into multiple peaks. Thus, for each carbon, the multiplicity of the signal depends on how many protons are attached to it.

In this case also (as in the case of  ${}^{1}H$  NMR) the n+1 rule applies, where n is the number of hydrogens on the carbon atom in question.

It is however possible to eliminate signal splitting by  ${}^{1}H^{-13}C$  coupling by choosing instrumental parameters for the NMR spectrometer that decouple the proton-carbon interactions. A  ${}^{13}C$  NMR spectrum in which  ${}^{1}H^{-13}C$  coupling is eliminated is known as **broadband (BB) proton decoupled**  ${}^{13}C$  NMR spectrum. In such a spectrum, each type of carbon atom produces only one peak.

#### Comparison of <sup>13</sup>C-NMR and <sup>1</sup>H-NMR Spectroscopy

- (i) As already stated <sup>1</sup>H-NMR spectroscopy gives indirect information about the nature of carbon in an organic molecule; this is because most of the carbons have atleast one H attached to it. On the other hand <sup>13</sup>C—NMR spectra gives signals arising from **all** carbon atoms in a organic molecule. So direct information is obtained about the carbon skeleton.
- (ii) <sup>13</sup>C spectras are much simpler compared to <sup>1</sup>H spectras due to very low natural abundane of <sup>13</sup>C. It is rather impossible that a <sup>13</sup>C nucleus in a molecule will have a second <sup>13</sup>C nucleus in immediate neighbour. Thus coupling of <sup>13</sup>C with neighbouring <sup>13</sup>C is unlikely.
- (iii) The signals in <sup>13</sup>C-spectras are spread over chemical range of 200 pm compared to a range less than 20 pm in case of proton spectra.

### 25.2.2 Interpretation of <sup>13</sup>C NMR Spectra

The information obtained from  $^{13}$ C NMR spectra is of the same kind as in case of  $^{1}$ H NMR spectra. In  $^{13}$ C NMR spectra, the information relates to the carbon skeleton. It includes the following:

- Number of signals
- Splitting of signals
- Chemical shift

In <sup>13</sup>C NMR spectra, the intensities of the signals (as is the case of <sup>1</sup>H NMR spectra) is not taken into account.

We will look into the above three set of information.

#### **Number of Signals**

The number of signals is a <sup>13</sup>C NMR spectra tell us how many different carbons or different sets of equivalent carbons are there in a molecule. The equivalence or non-equivalence of carbon is judged in the same way as in the case of protons (*see* section 25.1.3).

#### **Splitting of Signals**

As has already been stated a <sup>13</sup>C NMR spectra does not ordinary show carbon-carbon splitting (unless a compound is prepared using starting materials that are isotopically enriched with <sup>13</sup>C). Due to this, the <sup>13</sup>C NMR spectras are very much simple.

In <sup>13</sup>C NMR spectras, the peak for proton is not seen; this is because these signals are far off the scale. However, as has already been stated there is splitting of a carbon signal by the proton attached to the carbon itself. Thus, a proton decoupled spectra [Ususally known as broad band (BB) proton decoupled <sup>13</sup>C NMR spectra] consists of a set of single peak, one for each carbon or each set of equivalent carbons in the molecule.

As an example, in case of secondary butyl chloride, there are four different (or non-equivalent) carbons and we see four peaks in the proton-decoupled <sup>13</sup>C NMR spectra (Fig. 25.29).

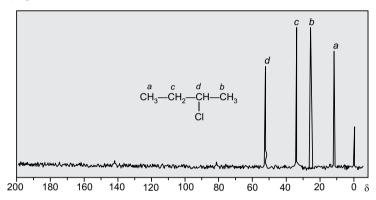


Fig. 25.29 Proton-decoupled CMR spectrum of sec. butyl chloride a,  $\delta$  12.1, b,  $\delta$  26.1, c,  $\delta$  34.3 and d,  $\delta$  52.8

However, proton coupled <sup>13</sup>C NMR spectra of sec. butyl chloride shows four multiplets (Fig 25.30).

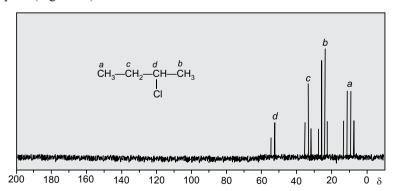


Fig. 25.30 Proton-coupled CMR spectrum of sec. butyl chloride a,  $\delta$  12.1 (q); b,  $\delta$  26.1 (q); c,  $\delta$  34.3 (t); d,  $\delta$  52.8 (d)

Thus, the proton-decoupled spectra (Fig. 25.29) tell us how many different carbons are there, and the proton-coupled <sup>13</sup>C NMR spectra (Fig. 25.30) tell us how many protons are attached to each of the carbon atoms. Both the spectras together give us a detailed picture of the structure of the molecule.

#### 25.2.3 Chemical Shift

In <sup>13</sup>C NMR, chemical shift arise virtually in the same way as in <sup>1</sup>H NMR. Each carbon nucleus has its own electronic environment, different from the environment of other, non-equivalent nuclei; it feels a different magnetic field, and absorbs at a different applied field strength. Carbon atoms that are attached only to other carbon and hydrogen atoms are relatively shielded from the magnetic field by the density of the electrons around them, and so the carbon atoms of this type produce peaks that are upfield in <sup>13</sup>C NMR spectra. On the other hand, carbon atoms bearing electronegative groups are deshielded from the magnetic field due to electron-withdrawing nature of these groups and so produce peaks that are downfield in the <sup>13</sup>C NMR spectra.

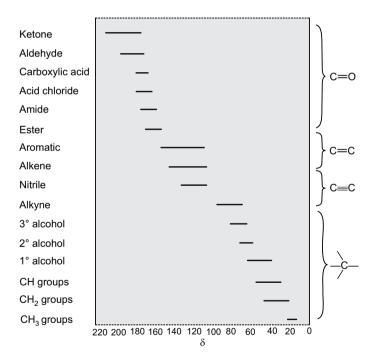


Fig. 25.31 Chemical shifts ranges for <sup>13</sup>C NMR spectra

The chemical shifts in  $^{13}$ C NMR differ in a number of ways from those in  $^{1}$ H NMR spectra. Thus, in  $^{13}$ C NMR, the chemical shifts are much larger than in  $^{1}$ H NMR; the scale in  $^{13}$ C extends from  $\delta 0$  to about 200. Finally,  $^{13}$ C NMR spectras

are normally not interpretated as in the case of  $^{1}H$  NMR. However, the reference standard assigned as zero ppm in  $^{13}C$  NMR spectra is also tetramethylsilane (TMS),  $(CH_{3})_{4}Si$  (as in the case of  $^{1}H$  NMR).

The approximate carbon-13 chemical shifts ranges are given in Fig. 25.31.

The  $^{13}$ C-chemical shifts of different types of compounds are in the order: C = 0 (ketones and aldehydes) > C=O (carboxylic acids, esters and amides) > C = C, C = N and aromatic carbons > C=C > C-O (alcohols and ethers) > C-X (X=Cl, Br, N $\checkmark$ ) > alkanes.

Electronegative groups like halogens, hydroxyl and other electron withdrawing functional groups deshield the carbon to which these groups are attached; this results the <sup>13</sup>C peaks to occur further downfield compared to those of unsubstituted carbon atoms. Following reference table (Table 25.7) gives the approximate chemical shifts for carbons bearing different substituents.

Table 25.7 Approximate Carbon-13 Chemical shifts

Type of Carbon Atom	Chemical Shift (δ, ppm)
1° Alkyl, RCH <sub>3</sub>	0–40
2° Alkyl, RCH <sub>2</sub> R	10–50
3° Alkyl, RCHR <sub>2</sub>	15–50
Alkyl halide or amine, —C—X (X=Cl, Br, or N—)	10–65
Alcohol or ether, —C—O	50–90
Alkyne, —C≡	60–90
Alkene, C=	100–170
Aryl, C_	100–170
Nitriles, —C≡N	120–130
O       Amides, —C—N—	150–180
Carboxylic acids, esters, —C—O, chlorides	160–185
Aldehydes, ketones, —C—	182–215

Let us consider the <sup>13</sup>C NMR spectra of 1-chloro-2-propanol

This compound has three unique carbons and so produces only three peaks in its broadband decoupled  $^{13}$ C NMR spectrum (Fig 25.32). Besides three singlet peaks, the spectrum shows three peaks at  $\delta$  77; these peaks arise from deutrochloroform (CDCl<sub>3</sub>) used as a solvent; in this case, the C of CDCl<sub>3</sub> is split into three peaks due to effect of the attached deuterium. The CDCl<sub>3</sub> peaks are disregarded for interpretation of  $^{13}$ C spectra.

The separation of the three peaks (a, b and c) is due to the differences in shielding by the circulating electrons in the local environment of each carbon. It has already been stated that lower is the electron density in the vicinity of a carbon, the less that carbon will be shielded and the more downfield will be the signal for that carbon. The oxygen of the hydroxyl group being most electronegative (in the structure  $CICH_2CH\_CH_3$  withdraws electrons most effectively. So the carbon (b) bearing the

—OH group is most deshielded and gives signal at the furthest downfield ( $\delta$  67). Chlorine being less electronegative than oxygen causes the peak for that carbon (a) to which it is attached to occur more upfield ( $\delta$  51). The methyl group (c) has no electronegative group directly attached to it and so it occurs at the furthered upfield ( $\delta$  20). Thus, by using the approximate chemical shift in Table 25.7, it is possible to assign <sup>13</sup>C NMR signal to each carbon in the molecule depending on the group attached to each carbon.

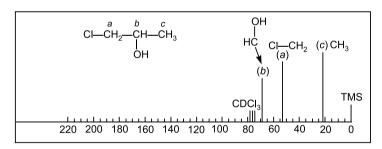
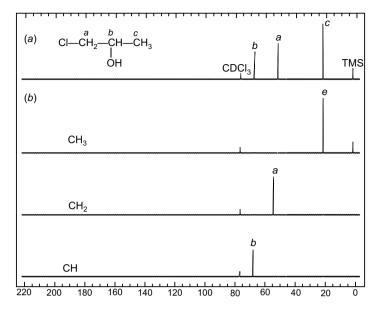


Fig. 25.32 Broadband decoupled <sup>13</sup>C NMR spectrum of 1-chloro-2-propanol

Now, it is possible to obtain <sup>13</sup>C NMR spectra known as DEPT <sup>13</sup>C spectrum (distortionless enhanced polarization transfer spectrum). Such a spectra consists of several spectra depicting one spectrum for each type of carbon atom. Thus, CH, CH<sub>2</sub> and CH<sub>3</sub> carbons are printed out on spectra with a <sup>13</sup>C spectrum in which all types of carbon are shown (Fig. 25.33). By such a spectrum, each of carbon is unambiguously identified.



**Fig. 25.33** (a) The broadband proton-decoupled <sup>13</sup>C NMR spectrum of 1-chloro-2-propanol. (b) These three spectra show the DEPT <sup>13</sup>C NMR data from 1-chloro-2-propanol

The spectrum shown above (a) is similar to that shown in Fig. 25.32.

## 25.2.4 Identification of Peaks in <sup>13</sup>C NMR Spectra on the Basis of Hybridization of Each Carbon Atom

The most important shifts in  $^{13}$ C NMR spectra are determined by the hybridization of the carbon atom. If we look at the spectra of 1-octene, it is found that the  $sp^3$ -hybridized carbons upfield ( $\delta$  14.1— $\delta$  34.1), and for the  $sp^2$ -hybridized carbons over  $\delta$  100, downfield from these at  $\delta$  113 and  $\delta$  140.

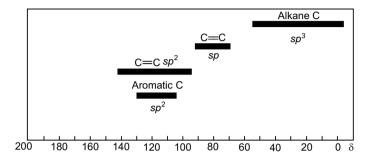
The aromatic carbons beings  $sp^2$ -hybridized also absorbs downfield in the same region as alkene carbons. Thus, in the spectrum of ethylbenzene, there are two sets of peaks—one upfield, a set from the side chain ( $sp^3$ -hybridized) carbon, and downfield a set from the ring ( $sp^2$ -hybridized) carbons.

The absorption by *sp*-hybridized carbons (as in the case of 1-hexyne) falls between the regions for  $sp^3$ -hybridized carbons and  $sp^2$ -hybridized carbons ( $\delta$  68— $\delta$  85).

13.7 22.1 30.9 18.3 84.5 68.4 
$$CH_3$$
— $CH_2$ — $CH_2$ — $CH_2$ — $C$ — $CH$ 
 $sp^3$ 

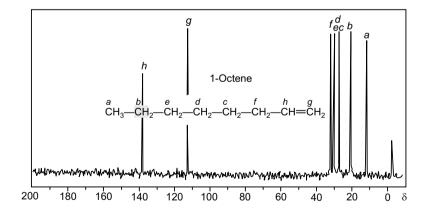
1-Hexyne
 $sp \rightarrow$ 

The regions of absorptions by  $sp^3$ -,  $sp^2$  -and sp-hybridized carbons are shown below (Fig. 25.34).

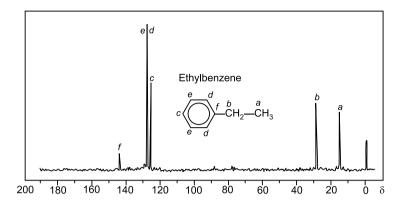


**Fig. 25.34** Chemical shift regions for  $sp^3$ -,  $sp^2$ - and sp-hybridized carbons in <sup>13</sup>C NMR spectra

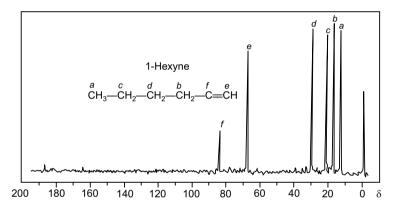
For the sake of convenience the <sup>13</sup>C NMR spectras of 1-octene, ethyl benzene and 1-hexyne are shown below (Figs. 25.35, 25.36 and 25.37). All these spectra show decoupled proton peaks; the protons coupled peaks are given at the bottom of each spectra showing the coupling of each of the carbons with the protons on each



**Fig. 25.35** Proton decoupled <sup>13</sup>C NMR spectra of 1-octene. In proton coupled spectra, the peaks at *a* to *h* appear as *q*, *t*, *t*, *t*, *t*, *t* and *d* respectively



**Fig. 25.36** Proton decoupled spectra of ethylbenzene. In proton coupled spectra, the peaks at *a* to *e*. appear as *q*, *t*, *d*, *d*, *d*, *s*, respectively



**Fig. 25.37** Proton decoupled spectra of 1-Hexyne. In proton coupled spectra, the peaks at *a* to *f* appear as *q*, *t*, *t*, *d* and *s* respectively

Let us now see the effect of carbon atoms of a substituted chain by chlorine. The <sup>13</sup>C NMR signals in case of *n*-pentane and 1-chloropentane are given below:

It is found that in signal for CH<sub>3</sub> ( $\delta$  13.7) shifts to  $\delta$  44.3 for CH<sub>2</sub>Cl. Thus, there is a difference of  $\pm$  30.6 ppm. Such a downfield shift is called an  $\alpha$ -effect. In case of C—2, chlorine again effects a downward shift from  $\delta$  22.6 to  $\delta$  32.7, a difference of  $\pm$  10.1 ppm; such a shift is called a  $\beta$ -effect. However, in case of C—3, absorption is shifted upfield from  $\delta$  34.5 to  $\delta$  29.20 a difference of  $\pm$  5.3 ppm. Such a shift is called a  $\alpha$ -effect. Beyond  $\alpha$ -carbon, the effect of chlorine is very small. In fact, all

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substituents (like F, Br, NH<sub>2</sub>, OH, NO<sub>2</sub>) on  $sp^3$ -hybridized carbon atom follow the same pattern, *i.e.*,  $\alpha$ - and  $\beta$ -effects downfield ( $\alpha$  having greater effect than  $\beta$ ) and  $\gamma$ -effect upfield.

#### 25.2.5 Two-Dimensional (2D) <sup>13</sup>C NMR Spectroscopy

As in the case of <sup>1</sup>H NMR spectra, two-dimensional (2D) spectra is also useful in <sup>13</sup>C NMR spectroscopy. In the case of <sup>13</sup>C NMR spectra, this technique is known as HETCOR (indicating coupling between hydrogens and the carbons to which they are attached). As already stated the word HETCOR stands for heteronuclear correlation spectra or C—H HETCOR.

In HETCOR spectrum, a <sup>13</sup>C spectrum is presented along one axis and a <sup>1</sup>H spectrum is presented along the other axis. The cross peaks in a HETCOR spectrum indicate which hydrogens are attached to which carbon in a molecule. There is no diagonal spectrum in the X—Y field like that found in the COSY. From the HETCOR spectra, it is apparent which hydrogens are attached to which carbon. The HETCOR spectrum of 1-chloro-2-propanol is given below (Fig. 25.38).

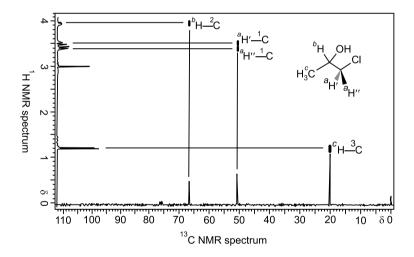


Fig. 25.38 HETCOR spectrum of 1-chloro-2-propanol

### 25.2.6 Applications of <sup>13</sup>C Spectra

(i) Identification of Functional Groups Containing Carbon Atoms: The carbon containing functional group can be easily identified using <sup>13</sup>C NMR spectra (*see* Table 25.7 and Fig. 25.31). The presence of functional group like OH, SH, NH, NH<sub>2</sub>, N=O, NO<sub>2</sub>, S=O and SO<sub>2</sub> cannot be detected.

- (ii) Number of Protons Attached to Carbon Atoms: Using proton coupled <sup>13</sup>C NMR spectra, the number of protons attached to carbon atoms can be determined.
- (iii) The hybridization state of each atom can be inferred on the basis of the chemical shifts of each carbon atom.
- (iv) <sup>13</sup>C NMR spectra can be used to confirm other assignments that have been made from other types of spectras. These data are summarized in Table 25.8.

**Table 25.8** Expected <sup>13</sup>C resonances for confirmation of structural assignments

Functional or structural group		Expected peak(s) in the broadband decoupled  13C NMR spectrum (δ)			
Acid chloride (—CO—CI)	ı	1 peak, 185–165			
Alcohl (—C—OH)	3°	1 peak, 85–70			
	2°	1 peak, 75–60			
	1°	1 peak, 70–40			
Aldehyde (—CHO)		1 peak, 205–175			
Alkene		2 peaks, 150–110			
Alkyne		2 peaks, 100–70			
Amide (—CO—NR <sub>2</sub> )		1 peak, 180–160			
Amine (—C—NR <sub>2</sub> )		1–3 peaks, 75–20			
Anhydride (—C—O—CO	—)	2 peaks, 175–150			
Benzenoid compounds		2–6 peaks, 155–110			
Carboxylic acid (—COOF	H)	1 peak, 185–160			
Ester (—COOR)		1 peak (C <del>—</del> O), 175–155			
		1 peak (R), 85–40			
Ether (—C—O—C—)		2 peaks, 85–40			
Ketone (—CO—)		1 peak, 225–175			
Nitrile (—C≡N)		1 peak, 130–110			
Methine group (CH)		1 peak, 60–30			
Methyl group (CH <sub>3</sub> )		1 peak, 30–20			
Methylene group (CH <sub>2</sub> )		1 peak, 45–25			

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### EXERCISES

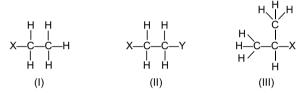
#### I. Multiple Choice Questions

1. Which of the following structures shows splitting in their NMR spectrum?



$$(c) \begin{array}{c} \mathsf{CH_3} \\ \mathsf{CH_3} \\ \mathsf{CH_3} \end{array} \\ \mathsf{C} = \mathsf{C} \\ \mathsf{H} \\ \mathsf{H} \\ (d) \begin{array}{c} \mathsf{CH_3} \\ \mathsf{Br} \\ \mathsf{C} = \mathsf{C} \\ \mathsf{H} \\ \mathsf{C} \\ \mathsf{$$

- 2. In the NMR spectra, of 2-chloropropane, CH<sub>3</sub>—CH—CH<sub>3</sub> the signal for tetriary proton will appears as:
  - (a) Doublet (b) Quartet (c) Triplet (d) Septet
- 3. In the following structures,



each structure has two types of protons. Which of the above structures will exhibit a quartet and a triplet (X and Y represent electron withdrawing groups that do not contain proton).

(a) I (b) II

(c) III (d) None of the above

**4.** In the above structures I, II, III (Q. No. 3), which of the structures will exhibit a doublet and a septet.

(a) I (b) II

(c) III (d) None of the above **5.** In the above structures I, II and III, (Q. No. 3), which one will exhibit two triplets?

in the above structures I, II and III, (Q. No. 3), which one will exhibit two triplets: (a) I (b) II

(c) III (d) None of the above

**6.** In the NMR spectrum of 1,2,4-trisubstituted benzene in which the substituents do not have any H, the number of peaks for H<sub>a</sub>, H<sub>b</sub> and H<sub>c</sub> proton will be

(a) d, d, d (b) d, d, t

(c) d, d, s (d) t, t, d

7. In the NMR spectra of p-nitrotoluene, besides a singlet for  $CH_3$  how many peaks (s, d or t) will be the (a) Two doublets (b) Two singlets (c) Two triplet (d) One singlet and one doublet **8.** Which of the following structures will show two doublets and one triplet? (The substituents x, y and z do not have any H.) (I) (II)(III)(a) I (b) II (d) None of the above (c) III **9.** Which of the following protons  $(H_a \text{ and } H_b)$  will have the highest J value? (a) I (b) II (d) IV (c) III **10.** How many signals will be obtained in the NMR spectra of 2-methyl propene? (a) Two (b) Three (c) Four (d) One 11. Which of the following is the best spectroscopic technique for confirming the presence of a phenolic hydroxyl group? (a) IR (b) UV (c) <sup>1</sup>H NMR (d) All can confirm

12. Which of the following are helpful in <sup>1</sup>H NMR spectra to determine the structure of an organic compound?

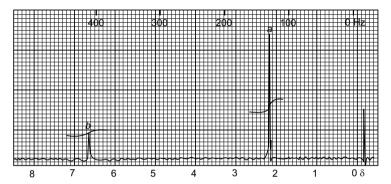
(a) Number of signals (b) Position of signals (c) Intensities of signals (d) Splitting of the signals

13.	Which of the following has spin number	$\frac{1}{2}$ ?			
	(a) <sup>1</sup> H	( <i>b</i> ) <sup>19</sup> F			
	$(c)^{13}$ C	$(d)^{2}H$			
14.	Which of the following will have spin nu	mber 1?			
	$(a)^{14}N$	(b) <sup>2</sup> H			
	(c) <sup>35</sup> Cl	$(d)^{11}B$			
15.	Which of the following will show three <sup>1</sup> H NMR signals?				
	(a) CH <sub>3</sub> —CHCl—CH <sub>3</sub>	$(b) \operatorname{CH}_3$ — $\operatorname{CH}_2$ — $\operatorname{CH}_2$ — $\operatorname{CH}_3$			
	$(c)  ext{ CH}_3$ — $ ext{CH}_2$ — $ ext{CH}_2$ Cl	(d) CH <sub>3</sub> —CHCl—CH <sub>2</sub> Cl			
16.	The two protons of $\mathrm{CH}_2$ group in $\mathrm{CH}_3\mathrm{CH}_2$	I <sub>2</sub> Cl are			
	(a) Enantiotopic	(b) Diastereotopic			
	(c) Can be (a) or (b)	( <i>d</i> ) None of the above			
17.	. Which of the following protons will have maximum downfield absorption?				
	(a) R—O <u>H</u>	( <i>b</i> ) Ar O <u>H</u>			
	(c) R—C <u>H</u> O	(d) RCOO <u>H</u>			
18.	Which of the following proton will have maximum upfield absorption?				
	(a) R—O $\underline{\mathbf{H}}$	$(b) C\underline{H}_3 - N <$			
	$(c)$ — $C \equiv C \underline{H}$	( <i>d</i> ) Ar <u>H</u>			
19.	Which of the following protons will have				
	(a) Methyl protons	(b) Methylene protons			
	(c) Methine protons	(d) All will have same $\delta$ value			
20.	Which of the following compounds will	show splitting in their spectras			
	(a) Benzaldehyde	(b) Aniline			
	(c) p-xylene	(d) All of the above			
21.	In case of <sup>13</sup> C NMR spectra, which of the following statement is correct?				
	(a) Each carbon whether equivalent or non-equivalent gives one peak				
	(b) Only non-equivalent carbons give one peak each				
	(c) Only equivalent carbons give one peak each				
	(d) None of the above is correct				
22.	Splitting of a <sup>13</sup> C NMR signal is due to the	ne			
	(a) Presence of neighbouring carbon				
	(b) Presence of neighbouring hydrogen				
	(c) Presence of hydrogen/hydrogens on the same carbon				
22	(d) There is no splitting				
23.	In case of CH <sub>3</sub> CH <sub>2</sub> OH, the <sup>13</sup> C NMR spectra shows splitting of the signal for carbon of CH <sub>3</sub> and CH <sub>2</sub> as				
	(a) $q$ and $t$	(b) t and $q$			
	(c) t and $t$	(d) q and $s$			

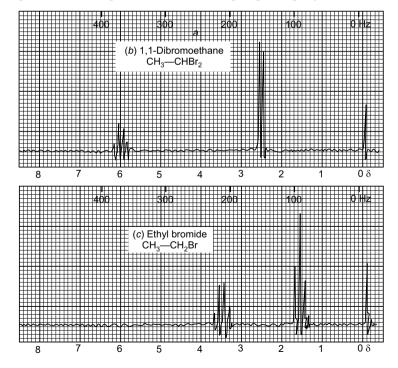
24.	<b>4.</b> A <sup>13</sup> C NMR spectra can be interpretated on the basis of				
	(a) Number of signals only		(b) Splitting of signals	s only	
	(c) Chemical shifts only		(d) A combination of	(a), (b) and (c)	
25.	In case of secondary butyl br	omide, CH <sub>3</sub> -	-CHCH <sub>2</sub> CH <sub>3</sub> , how i	many signal will	
			l Br		
	be obtained in its <sup>13</sup> C NMR s	spectra?			
	(a) 1		(b) 2		
	(c) 3		(d) 4		
26.	<b>26.</b> In the proton coupled <sup>13</sup> C NMR spectra of secondary butyl chloride, the s				
	C in the group —CH—will a	ppear as			
	ĊI				
	(a) Singlet		(b) Doublet		
	(c) Quartet		(d) Triplet		
27.	27. Which of the following signal will came upfield in the <sup>13</sup> C NMR spectra?				
	(a) Ketones		(b) Alkenes		
	(c) Alcohols		(d) CH <sub>3</sub> groups		
28.	8. In case of <sup>13</sup> C NMR spectra of 1-chloro-2-propanol, which of the C signal				
	expected to come as furtherest upfield?				
	(a) ClCH <sub>2</sub>	(b) —CH—     OH			
			011		
	(c) —CH <sub>3</sub>		(d) All will be at the s		
29.	<b>29.</b> The chemical shift of which of the following carbon will appear at max upfield				
	(a) sp		$(b) sp^2$		
	$(c) sp^3$		(d) All will be at the s	ame δ value	
	- <u>-</u> -	NSWER	s ‡		
			_		
1.	(d) <b>2.</b> (d)	<b>3.</b> ( <i>a</i> )	<b>4.</b> (c)	<b>5.</b> ( <i>b</i> )	
6.	(c) 7. (a)	<b>8.</b> (b)	<b>9.</b> (b)	<b>10.</b> ( <i>a</i> )	
11.	, , , , , , , , , , , , , , , , , , , ,	<b>13.</b> (a), (b)		<b>15.</b> ( <i>c</i> )	
16.	, ,	<b>18.</b> (a)	<b>19.</b> (c)	<b>20.</b> (c)	
21.		<b>23.</b> (a)	<b>24.</b> (d)	<b>25.</b> ( <i>d</i> )	
26.	(b) <b>27.</b> (d)	<b>28.</b> (c)	<b>29.</b> (c)		
II. Shor	rt Answer Questions				

- 1. What are enantiotopic protons and diastereotopic protons? How do they behave in <sup>1</sup>H NMR spectra? Explain with the help of examples.
- 2. What do you understand by splitting of signals? Explain with reference to the <sup>1</sup>H NMR spectra of pure ethyl alcohol.

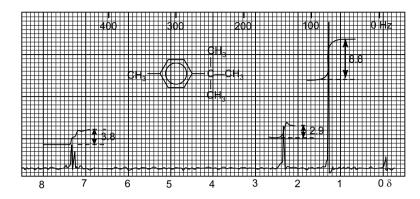
- **3.** Explain the term coupling constant. How it is useful for structure elucidation of compounds? Give examples.
- 4. How many kinds of protons are there in nitrobenzene and toluene?
- 5. In case of 2-chloro-3-methyl-2-butene, how many NMR signals will be obtained?
- **6.** What type of peaks are expected to be observed in the <sup>1</sup>H NMR spectrum of ethylacetoacetate.
- 7. An organic compound exhibits a doublet at  $\delta$  8.9 (6 H) a multiplet at  $\delta$  8.1 (1 H) and doublet at  $\delta$  6.55 (2 H). What is the probable structure of the compound?
- **8.** Assign a probable structure of the compound, whose NMR spectrum is given below:



**9.** The NMR spectras of 1, 1-dibromoethane and ethyl bromide are given below. Explain the different peaks with reference to spin-spin coupling.



10. The NMR of p-tert-butyl toluene is give below. Assign the different peaks.



- 11. Give a structure consistent with the NMR data of the following two compounds:
  - (i)  $C_{10} H_{14} \text{ singlet } \delta 0.88 \text{ (9H)}$ singlet  $\delta 7.28 \text{ (5H)}$
  - (ii)  $C_9H_{11}Br$  quartet  $\delta$  2.15 (2H) triplet  $\delta$  2.75 (2H) triplet  $\delta$  3.38 (2H) singlet  $\delta$  7.25 (5H)
- **12.** With the help of NMR spectra, how will you differentiate between the following structures:

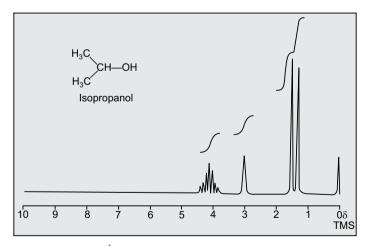
**13.** Describe, how will you determine the equivalence of protons. Illustrate with each hydrogen in

- 14. A compound having the molecular formula  $C_9H_{11}$ Br exhibited in its NMR spectra a singlet ( $\delta$  7.25), two doublets ( $\delta$  2.75 and  $\delta$  1.45, 2H and 3H respectively) and a multiplet ( $\delta$  3.4, 1H). Assign a structure to this compound.
- **15.** How many different types of protons are there in 1, 3, 5-trimethylbenzene and what will be their splitting pattern?
- 16. An organic compound,  $C_9H_{10}O_2$  showed three singlets in the NMR at  $\delta$  7.31 (5H), 5.08 (2H) and 2.06 (3H). It also showed a peak at 1730 cm<sup>-1</sup> in the IR spectra. Identify the organic compound.
- 17. Write note of
  - (i) Fourier Transform (FT) NMR spectrometer
  - (ii) Magnetic anisotropy effects

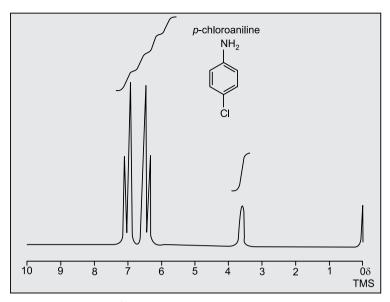
- (iii) n + 1 Rule
- (iv) Deuterium exchange.
- **18.** How is plane of symmetry and centre of symmetry is helpful in determining whether the protons are equivalent or not? Give examples.
- **19.** How will you proceed to interpret the <sup>1</sup>H NMR spectra of a compound? Give one example.
- **20.** For complicated <sup>1</sup>H NMR spectras, how does double resonance and deuterium labelling are useful?
- **21.** For the following compound, give approximate chemical shift range for each proton or set of protons.

H C=C—H CH<sub>3</sub> 
$$Ans. \quad {}^{\delta \, 5-6}_{\text{Cl}} + {}^{\text{C}}_{\text{Cl}} = {}^{\text{C}}_{\text{CH}_3} \, {}^{\delta \, (1.5-2)}$$

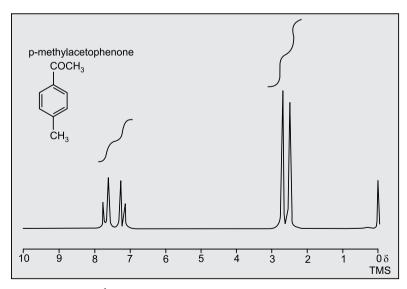
- 22. Estimate the chemical shift values for the protons in 1,1-dichloroethane.
- **23.** Following are given the <sup>1</sup>H NMR spectras of isopropanol, *p*-chloroaniline, *p*-methyl acetophenone, cinnamic acid, ethyl methyl ketone, 2-bromobutyric acid, phenacetin and isopropyl benzene. Assign different peaks in each of the spectras.



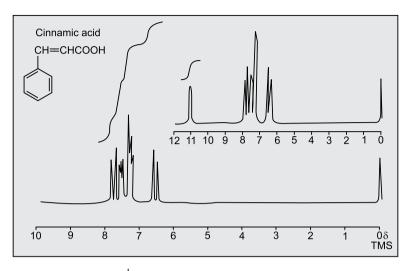
<sup>1</sup>H NMR Spectrum of Isopropanol



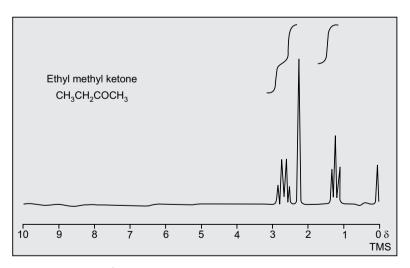
<sup>1</sup>H NMR Spectrum of *p*-Chloroaniline



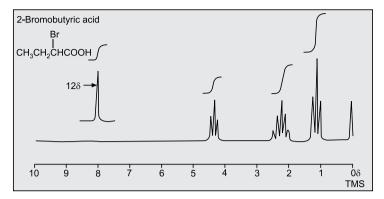
<sup>1</sup>H NMR Spectrum of *p*-Methylacetophenone



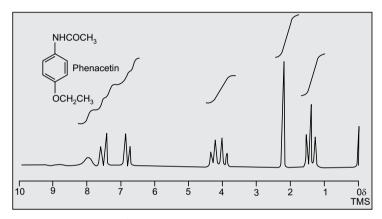
<sup>1</sup>H NMR Spectrum of cinnamic acid



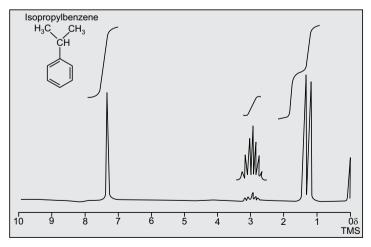
<sup>1</sup>H NMR Spectrum of ethyl methyl ketone



<sup>1</sup>H NMR Spectrum of 2-bromobutyric acid

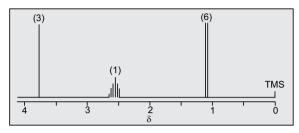


<sup>1</sup>H NMR Spectrum of phenacetin



<sup>1</sup>H NMR Spectrum of Isopropylbenzene

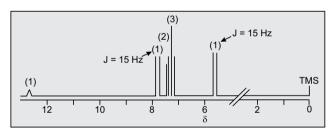
- 24. How many chemically different kinds of protons are there in see. butyl alcohol? How many <sup>1</sup>H NMR signals are expected in the spectrum of sec. butyl alcohol? [Hint: Sec. butyl alcohol as you know exists in two enantiomers.]
- 25. A compound, C<sub>5</sub>H<sub>10</sub>O<sub>2</sub> has the <sup>1</sup>H NMR spectra given below.



Give a structure to the compound.

26. Give the expected <sup>1</sup>H NMR spectrum for 1-bromo-1-(4-bromophenyl) ethane.

A compound,  $C_9H_8O_2$  has the  $^1H$  NMR spectra given below. The signal at  $\delta$  12.4 disappears when the sample is treated with  $D_2O$ . The J value for the doublet at  $\delta$  5.45 and  $\delta$  7.77 is about 15 Hz.



Give a structure to the compound.

**27.** For the compound (structure given below), give the approximate chemical shift range for each proton or set of protons.

$$H$$
  $C=C$   $C\equiv C-H$   $CH_3$ 

[Hint: The molecule contains three types of protons–alkyne proton and proton attached to alkene carbon and alkane carbon adjacent to a  $\pi$  bond. Using the data given in Tables 25.2 and 25.3).

[Ans. 
$$^{(\delta \, 5-6)}$$
 H C=C  $^{\text{C}\equiv\text{C}-\text{H} \, (\delta \, 2.5-3)}$   $^{\text{C}}$   $^{\text$ 

- 28. The major isotope of carbon, <sup>12</sup>C does not produce a NMR signal. Explain.
- **29.** In the case of <sup>13</sup>C NMR spectra, the multiplicity of peaks due to neighbouring carbon normally does not occur. Explain why?
- **30.** In <sup>13</sup>C NMR spectra, the peak for proton is not seen. Why?
- **31.** In what ways, the chemical shifts in <sup>13</sup>C NMR spectra differ from those in <sup>1</sup>H NMR spectra?
- **32.** Normally CDCl<sub>3</sub> is used as a solvent in <sup>13</sup>C NMR spectrum. Its peak occurs as a triplet. Explain.
- 33. In the  $^{13}$ C NMR spectra (decoupled) of methyl methacrylate give the carbon atoms in the decreasing order of  $\delta$  value (upfield) (you may refer to Table 25.6). Explain.

$$\begin{array}{c|c}
H & O \\
C = C - C - OCH_3
\end{array}$$

$$\begin{array}{c|c}
D & d \\
C = C - C - OCH_3
\end{array}$$

[**Ans.** *a*, *b*, *c*, *d*, *e*]

**34.** An organic compound,  $C_5H_{11}Cl$  gave the following  $^{13}C$  NMR data  $\delta$  9.5 (q),  $\delta$  32.0 (q),  $\delta$  39.0 (t) and  $\delta$  70.9 (s). Give the structural formula of the compound.

**35.** An organic compound  $C_5H_{11}$ Br on  $^{13}$ C NMR analysis gave the following signals  $\delta$  26.0 (CH<sub>3</sub>),  $\delta$  32 (CH),  $\delta$  33 (CH<sub>2</sub>) and  $\delta$  42 (CH<sub>2</sub>). Give the structural formula of the compound.

[Ans. Br 
$$CH_2CH_2$$
  $CH_3$   $CH_3$ 

- **36.** What information you can obtain from <sup>13</sup>C NMR spectra?
- **37.** The proton decoupled <sup>13</sup>C NMR spectra of *p*-bromotoluene will exhibit what types of pattern.

[Hint: The six aromatic carbons will show six singlets in the region  $\delta$  120-140. The CH<sub>3</sub> carbon will appear as a singlet at  $\delta$  2.0.

**38.** Disuss the <sup>13</sup>C NMR spectra of carboxylic acids and their derivatives.

**[Hint:** The carbonyl carbon of carboxylic acids and their derivatives occur far douwnfield in the region  $\delta$  160—180. The nitrile carbon is not shifted so downfield and absorbs in the region  $\delta$  115-120. The downfield shift of these derivatives is not as much for aldehydes and ketones ( $\delta$  180—220)]



# Chapter 26

## Electron Spin Resonance (ESR) Spectroscopy

#### **26.1 INTRODUCTION**

It is a type of absorption spectroscopy and is also called 'electron paramagnetic resonance' (EPR) and 'electron magnetic resonance' (EMR) spectroscopy. It consists in the absorbance of radiation having frequency in the microwave region by paramagnetic substances, which induces transition between magnetic energy levels of electrons with unpaired spins. The magnetic energy spliting is achieved by the application of a static magnetic field. The unpaired electrons are normally present is odd molecules, free radicals, triplet electronic states and transition metals and rare earth ions. The unpaired electrons of free radicals are mostly studied. These are generally obtained by the homolytic fission of a covalent bond.

$$A : \overset{\mathsf{A}}{\xrightarrow{\mathsf{B}}} \xrightarrow{\mathsf{Homolytic fission}} \overset{\dot{\mathsf{A}}}{\xrightarrow{\mathsf{hv}}} \overset{\dot{\mathsf{b}}}{\xrightarrow{\mathsf{Free radicals}}}$$

The electron spin resonance spectroscopy concerns with the magnetic behaviour of the spinning electrons. ESR spectrum (like the NMR spectrum) results from transition from one spin state to the other state of an electron; the transion occurs under the resonation conditions in magnetic field. The ESR differs from NMR is that in ESR the transition is induced by radiation of microwave frequency rather than by radiowave (in NMR)?

As already stated ESR is useful for paramagnetic substances. These are of two types:

(a) Stable Paramagnetic Substances: These can be easily studied by ESR. Examples of such substances include molecules like NO, O<sub>2</sub> and NO<sub>2</sub> and ions of rare earth elements and of transition metals.

(b) Unstable Paramagnetic Substances: These are formed as intermediates in a chemical reaction or are produced by the irradiation of stable molecules with uv light or X-ray radiation. The species produced are also called free radicals or radical ions. These can be studied by ESR provided their life times are greater than 10<sup>-6</sup> second. In case the life times are less than 10<sup>-6</sup> second, these should be produced at low temperatures.

#### **Principle of ESR Spectroscopy**

In ESR, interaction of the magnetic moment of an unpaired electron in a molecule ion with an applied magnetic field produce energy levels. The transitions between these energy levels by absorbing radiations of microwave frequency gives an ESR spectrum.

The spin angular momentum quatum number of an electron spin  $s=\frac{1}{2}$ , will have a value of  $m_S=\pm\frac{1}{2}$ . In the absence of an external magnetic field, the free electron may exist in one of the two states,  $+\frac{1}{2}$  or  $-\frac{1}{2}$ , of equal energy (degenerate energy levels). However, in presence of magnetic field  $(H_o)$ , the degeneracy is removed giving two non-degenerate energy levels. The lower energy state having the spin magnetic moment alligned with the field corresponds to quantum number,  $m_S=-\frac{1}{2}$  and the higher energy state will have magnetic moment opposed to the applied field and corresponds to the quantum number,  $m_S=+\frac{1}{2}$  (Fig. 26.1).

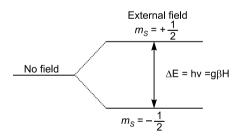


Fig. 26.1

The two states (Fig. 26.1) possess energies that are split up from original state in the absence of applied magnetic field by the amount  $+\mu_e H$  and  $-\mu_e H$  for the low energy and high energy state respectively [H is the magnetic field applied on the unpaired electron and  $\mu_e$  is the magnet moment of the spinning electron]. The transition between the two different energy levels takes place by absorption of aquantum of radiation of frequency in the microwave region. Therefore, the ESR

spectrum of a free electron will be a single peak corresponding to transition between the two energy levels. When absorption takes place, following relation holds good.

$$2\mu H = h v \qquad ...(i)$$

where v is the frequency of radiation absorbed in cycles per second.

The above relation is valid for free electrons. The energy of transition ( $\Delta E$ ) is given accurately by the relation

$$\Delta E = hv = g\beta H \qquad ...(ii)$$

Where h is Planks constant, H is the applied magnetic field,  $\beta$  is the Bohr's magneton, a factor used for converting angular momentum into magnetic moment.

$$\beta = \frac{eh}{4\pi mc} = 0.9723 \times 10^{-23} \text{ erg/gauss}$$

where e is the charge of the electron, m is the mass of the electron and C is the velocity of light.

Finally g [in equation (ii)] is the proportionately factor, a function of the electron's environment. It is also called Lande's splitting factor or spectroscopic splitting factor. For free electron the value of g is 2.0023. However, this value is slightly modified for electrons in molecules.

#### **26.2 INSTRUMENT**

An ESR spectrometer consists of the following components as shown in Fig. 26.2.

- (i) Klystron: It is a vacuum tuble to produce microwave oscillations and gives frequency on a small range. The frequency of the monochromatic radiation is dependent on the applied voltage to klystron. Normally, a Klystron oscillator operates at 9500 Mc/sec.
- (ii) Isolator: It is a strip of ferrite material. Its function is to minimise vibrations in the frequency of microwaves produced by Klystron oscillator.
- (iii) Wavemeter: It is normally calibrated in frequency units (megahertz) and is used to know the frequency of microwaves produced by Klystron oscillator.
- (*iv*) **Attenuator:** It adjusts the level of the microwave power emitting from the wavemeter. It has an absorption element (a neutral fillter)
- (v) Circulator: From the attenuator, the microwave radiations passed through a four-port circulator. The microwave radiations enter arm 1. The directions of microwave transmission along the various arms are shown in the figure.
- (vi) Sample Cavity: The sample whose ESR spectrum is to be determined is taken in the sample cavity. The sample cavity is such so as to maximize the applied magnetic field along the sample. Normally 0.15 to 0.5 mL

solution of the sample is taken in the sample cavity. In almost most of the ESR spectrometers, two sample cavities are used, one for the sample and the other for the reference. This arrangement is helpful in compensating the sources of error by comparing relative heights of the signal.

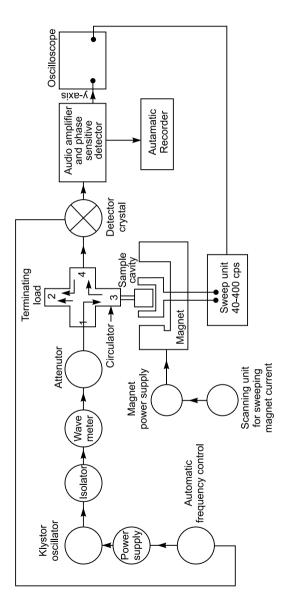
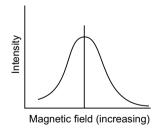


Fig. 26.2 Schematic diagram of ESR spectrometer showing various components

- (vii) Magnet: An electromagnet, which provides a homogeneous magnetic field is used (the sample cavity is placed between the poles of an electromagnet). The homogeneous magnetic field can be varied from zero to 500 gauss. Highly regulated power supply helps to stabilize the field.
- (viii) Crystal Detector: It is a silicon crystal and acts as a microwave racitifer and converts microwave power into a direct-current output.
  - (ix) Autoamplifier and Phase Sensitive Detector: The signal after been detected by crystal detector undergoes narrow band amplification. Any noise in the amplified signal is rejected.
  - (x) Oscillioscope and Pen Recorder: The signal is finally recorded by an oscillisocope or a pen-recorder.

#### 26.3 RECORDING AN ESR SPECTRA

- 1. The sample, whose ESR sepctra is to be recorded is taken in the sample cavity.
- 2. The Klystron oscillator is set. The produced microwaves pass through the isolator, wave meter and attenuator. These microwaves are received by the circulator. The direction of microwave transmission is shown in Fig.26.2. The dector acting as a ractifier, converts the microwave power into direct current. In case the magnetic field around the sample cavity is changed to a value required for resonance, the recorder will show an absorption peak (Fig. 26.3). However, if the main magnetic field is swept slowly (2-5 minutes), the recorder will show the derivative of the microwave absorption spectrum against field (Fig. 26.4).



Magnetic field (increasing)

Fig. 26.3

Fig. 26.4

**3. Sensitivity of the ESR Spectrometer**: The sensitivity of an ESR spectrometer is given by the expression:

$$N_{\min} = 1 \times 10^{11} \, \frac{\Delta H}{\sqrt{t}}$$

where  $N_{\min}$  is the minimum number of detectable spins per gauss

 $\Delta H$  is the width between deflection points on the derivative absorption curve

t is the time constant of the detecting system, which is inversely proportional to the band width of the detection circuit.

The sensitivity of the ESR spectrometer is increased with higher magnetic field.

- **4.** Concentration of the Sample: The concentration of the sample should be of the order 10<sup>-9</sup>M for samples having very small dielectric loss. 10<sup>-7</sup>M for aqueous solutions and 10<sup>-6</sup>M for structure determination and quantitative analysis.
- 5. Choice of Solvent: High dielectric constant solvents like water, alcohol etc. should not be used, since these strongly absorb microwave power. In certain cases, solvents with high dielectric constants can be usual provided the sample to be analysed has strong absorbance and is contained in a specially designed cell (narrow sample tube). ESR spectras can also be obtained for gases, solutions, powders, single crystals and fozen solutions. The frozen solvents are obtained when the solvent freezes to form a glass. Some examples of solvents which form a glass are methylcyclohexane, glycerol, toluene, Nujol and Iso-octane. Mixtures of solvents which form glass include Propane + propene; ethanol + methanol; Ethanol+ diethyl ether; toluene + acetone; Toluene + Chloroform.
- **6. The ESR Spectrum**: The ESR spectrum is similar to NMR spectrum and is obtained by plotting intensity against strength of the magnetic field. A better way is to represent the ESR spectrum as a derivative curve (see Figs. 26.3 and 26.4). Each negative slope in the derivative curve represents a peak or shoulder in the absorption spectrum. Crossing of the derivative axis with a negative slope indicates a maximum. On the other hand, crossing with positive slope indicates a minimum. In fact, the number of peaks or shoulders in an absorption curve is determined from the number of minima or maxima in the derivative curve.

The total area covered by either the absorption or derivative curve is proportional to the number of unpaired electrons in the sample. The number of electrons in the sample can be calculated by comparison with a standard sample having known number of unpaired electrons. A commonly used standard is 1, 1-diphenyl-2picrylhydrazyl free radical (DPPH).

DPPH is chemically stable and has a spinning factor g = 2.0036. It contains  $1.53 \times 10^{21}$  unpaired electrons per gram.

DPPH cannot be used as standard for other free radicals since there is only a very small variation in the g-values. Thus, it is not possible to distinguish from the standard substance. In case of free radical, the internal standard used is a trace of Cr (III) entrapped in a tiny chip of ruby crystal cemented to the sample cell. This standard gives a strong resonance and its *g* value is 1.4. Quantitative measurement can be done on the integral of the ESR curve.

#### 26.4 HYPERFINE SPLITTING

ESR spectrum exhibits hyperfine splitting. It is caused by the interaction between the electron and the nuclear spin of the adjacent spinning magnetic nuclei. The number of splitting is equal to 21 + I (where I is the spin quantum number of the nucleus) when are single electron interacts with one nucleus. In case, a single election intracts magnetically with n equivalent nuclei, the electron signal splits into (2n + I + 1) multiplet.

#### 26.4.1 ESR Spectra of Hydrogen Atom

As an example, is case of a hydrogen atom having one proton and one electron  $\left(I=\frac{1}{2} \text{ for the proton}\right)$ , in absence of magnetic field, the single electron of spin  $\left(S=\frac{1}{2}\right)$  gives a doubly degenerate spin energy state. However, on application of magnetic field the degeneracy is removed and the two energy lives  $\left(m_s=-\frac{1}{2}\right)$  and  $m_s=+\frac{1}{2}$  are alligned with the field (in case  $m_s=\frac{1}{2}$ ) or opposing the field (in case  $m_s=+\frac{1}{2}$ ) (Fig. 26.5).

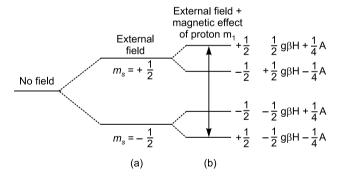


Fig. 26.5 (a) Effect of external magnetic field on energy states of an electron, (b) Effect of a nuclear spin  $\frac{1}{2}$  of proton on energy states

As seen (Fig. 26.5), there are four energy levels. Their energies are:

$$E_{\frac{1}{2},\frac{1}{2}} = \frac{1}{2}g\beta H + \frac{1}{4}A \qquad ...(i)$$

$$E_{\frac{1}{2},-\frac{1}{2}} = -\frac{1}{2} g\beta H - \frac{1}{4} A \qquad ...(ii)$$

$$E_{-\frac{1}{2},-\frac{1}{2}} = -\frac{1}{2} g\beta H + \frac{1}{4} A$$
 ...(iii)

$$E_{-\frac{1}{2},\frac{1}{2}} = -\frac{1}{2} g\beta H - \frac{1}{4} A$$
 ...(iv)

The four energy levels [equations (i) to (iv)] are given by the general expression

$$E = g\beta H m_c + A m_c m_I \qquad ...(v)$$

where A is the hyperfine coupling constant. Substituting the appropriate values of  $m_s$  and  $m_I$  in the above equation ( $\nu$ ), the values of energies as indicated in Fig. 26.5 are obtained. The selection rules in ESR are

$$\Delta_{mI} = 0$$
 and  $\Delta m_s = \pm 1$ 

The ESR spectrum of a free electron will consist of a single peak corresponding to transition between these levels (Fig. 26.5).

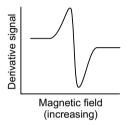


Fig. 26.5 ESR spectrum of free electron

Consideration of the interaction between the two energy states and the nuclear spin due to proton, each energy state is further split into two energy levels corresponding to  $m_I = +\frac{1}{2}$  and  $m_I = -\frac{1}{2}$  (where  $m_I$  is the nuclear spin angular momentum quantum number). Thus, four different energy levels are obtained (Fig. 26.6). In this situation, the ESR spectrum will consist of two peaks (Fig. 26.6) corresponding to two transitions shown by two arrows in Fig. 26.6.

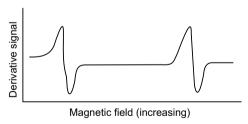


Fig. 26.6 ESR spectrum of hydrogen

#### 26.4.2 ESR Spectra of Deuterium

In case of deuterium, the energy levels are calculated as in the case of hydrogen atom. Thus, corresponding to  $m_s = +\frac{1}{2}$ , there are three values of  $m_I = 1$ , 0, -1 and corresponding to  $m_s = -\frac{1}{2}$ , there are also three values of  $m_I$ , i.e.,  $m_I = -1$ , 0 and +1. (Fig. 26.7).

$$\begin{split} E_{\frac{1}{2},1} &= \frac{1}{2}g\beta H + \frac{1}{2}hA \\ E_{\frac{1}{2},0} &= \frac{1}{2}g\beta H \\ E_{\frac{1}{2},-1} &= \frac{1}{2}g\beta H - \frac{1}{2}hA \\ E_{-\frac{1}{2},-1} &= -\frac{1}{2}g\beta H - \frac{1}{2}hH \\ E_{-\frac{1}{2},0} &= \frac{1}{2}g\beta H \\ E_{-\frac{1}{2},1} &= \frac{1}{2}g\beta H - \frac{1}{2}hA \end{split}$$

Using the selection rules ( $\Delta m_s = \pm 1$  and  $\Delta m_I = 0$ ), three allowed transitions are possible and thus three lines corresponding to these transitions are obtained in the ESR spectrum of deuterium (Fig. 26.7).

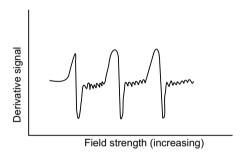


Fig. 26.7 ESR Spectra of deuterium

Various energy states in case of deuterium are shown in Fig. 26.8.

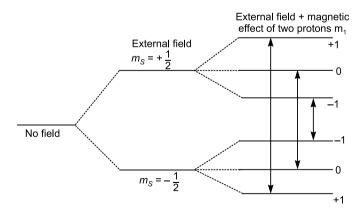


Fig. 26.8 Various energy states in deuterium

#### 26.4.3 ESR Spectra of Methyl Radical

In case of methyl radical (CH<sub>3</sub>), we consider the interaction between the unpaired electron and three hydrogen nuclei, it is found that there are four values corresponding to  $m_s = +\frac{1}{2} \left( m_I = +\frac{3}{2}, +\frac{1}{2}, -\frac{1}{2}, -\frac{3}{2} \right)$  and four values corresponding to  $m_s = -\frac{1}{2} \left( m_I = -\frac{3}{2}, -\frac{1}{2}, +\frac{1}{2}, +\frac{3}{2} \right)$  (Fig. 26.9).

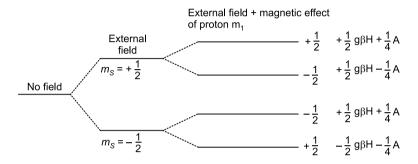


Fig. 26.9 Energy states in methyl radical

Application of selection rules ( $\Delta m_{\rm I} = 0$  and  $\Delta m_s = +1$ ), it is observed that four transitions are possible giving four lines in the ESR spectra of methyl radical. The relative intensities for the four lines are in the ratio 1:3:3:1 (Fig. 26.10).

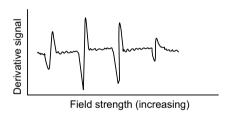


Fig. 26.10 ESR spectra of methyl radica

Thus, in case an unpaired electron undergoes interaction with n equivalent nuclei of equal spin I, the number of lines observed in the ESR spectrum will be (2nI+1). However, interaction involving a set of n equivalent nuclei of spin  $I_i$  and a set of equivalent nuclei m of spin  $I_j$ , the number of line observed in the ESR spectrum will be  $(2n+I_i+1)(2mI_i+1)$ .

#### 26.5 DETERMINATION OF G-VALUE

The value of g is determined by measuring the field spearation between the centre of the unknown spectrum and that of a reference whose g-value is correctly known.

Generally, DPPH is used as a standard, its g-value is 2.0036. The standard substance is placed along with the unknown in the same chamber of a dual cavity cell of the ESR spectrometer. The ESR spectra shows two signals with field separation of  $\Delta H$ . The g-value for the unknown is given by

$$g = g_s \left( 1 - \frac{\Delta H}{H} \right)$$

where H is the resonance frequency. In case, the known has its centre at higher field,  $\Delta H$  is positive.

#### 26.6 LINE WIDTH

The line width of an ESR signal is dependent on the relexation time of the state under study. The relexation time is usually  $10^{-7}$  sec.

Substitution of the relexation time in the Heisenberg's uncertainty relation gives a frequency uncertainty of 1 gauss

$$\Delta E + \Delta t = h/2\pi$$

The widening of the signal is the result of shorter relexation time. The line width in ESR spectrum is more than in NMR.

The advantage of wider lines in ESR spectrum is that a lower magnetic field homogeneous to 1 in  $10^5$  over the sample is employed compared to 1 in  $10^8$  is sufficient. However, a disadvantage associated with wider lines is these are comparatively more difficult to observe and measure than sharp lines. It is due to this reason that the ESR spectrometers are normally operated in the derivative mode.

The increase in width of spectral lines in ESR spectra is called broadening of the lines. The broadening of the lines can be remedied by working at very low temperatures.

It is difficult to observe spectrum of titanium (III)  $(d^1)$ . This is because in this case, the ground state has another orbital level about it and not having different energy.

The broadening of the spectral lines in ESR also occurs if the concentration of the paramagnetic species is high. In such cases only a trace quantity is incorporated as an impurity on a diamagnetic compound, which is isomorphous with the product under study.

#### 26.7 HYPERFINE STRUCTURE IN ESR SPECTRA

As already stated, in ESR spectra there is interaction between the electron spin and the nuclear spins of the adjacent magnetic nuclei. Due to this the ESR signals are split into several lines resulting is a hyperfine structure.

The selection rule for hyperfine transition is

$$\Delta m_s = \pm 1; \Delta m_I = 0$$

The interpretation of selection rule is made by saying that the nuclear motion is slower than the electronic motion so that during the time the electron changes its spin orientation, the nucleus (being heavier than the electron) has no time to reorient its spin and so  $\Delta m_{\rm I} = 0$ .

The hyper structure interaction is discussed in some typical cases.

#### (i) Hydrogen atom

Hydrogen atom has an unparied electron  $\left(S=\frac{1}{2}\right)$  and a proton  $\left(\text{nuclear spin }I=\frac{1}{2}\right)$ . Hence  $m_s=\pm\,\frac{1}{2}$  and  $m_I=\pm\,\frac{1}{2}$ .

In the absence of magnetic field (H=0), the electron spin energy level have the same energy, *i.e.*, the elergy levels are degenerate. However, application of magnetic field removes the degenaracy, the  $m_s=-\frac{1}{2}$  sublevel goes down and the  $m_s=\pm\frac{1}{2}$  sublevel going up (Fig. 26.11). Each of the electron sublevel interacts with the nucleus gives four sublet levels designated by values of  $m_I$ . This is known as hyperfine interaction. Using the selection rules, a doublet (two ESR lines) are observed.

The spacing between the two signals is called hyperfine coupling constant, a, which is expressed in units of tesla or millitesla.

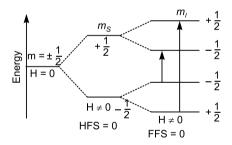


Fig. 26.11 Hyperfine Structure in ESR Spectrum of hydrogen atom

#### (ii) Methyl radical (CH<sub>3</sub>)

Methyl radical has an unpaired electron  $\left(S = \frac{1}{2}\right)$  and three equivalent protons  $\left(I = \frac{1}{2}\right)$ . For three equivalent protons,  $m_I = +\frac{3}{2}, +\frac{1}{2}, -\frac{1}{2}, \frac{3}{2}$ . Also  $m_s = \pm \frac{1}{2}$ . The hyperfine structure is shown in Fig. 26.12. The ESR spectrum has four equally spaced lines (quartet) (Fig. 26.13).

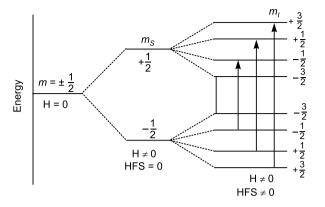


Fig. 26.12 Hyperfine spectrum of methyl radical



Fig. 26.13 ESR Spectra of methyl radical

#### (iii) 1, 4-Benzoquinone radical anion



Fig. 26.14

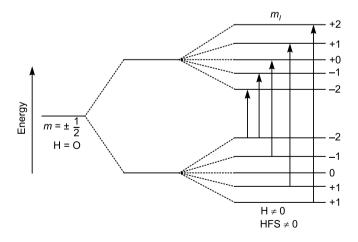


Fig. 26.15 Hyperfine structure, in the ESR spectrum of *p*-benzoquinone anion

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It has one unpaired electron  $\left(S = \frac{1}{2}\right)$ , which interacts with four equivalent protons. For a proton,  $I = \frac{1}{2}$  and four equivalent protons  $m_I = +2, 1, 0, -1, 2$ . The hyperfine structure of 1, 4 benzoquinone radical ion is shown in Fig. 26.15. The ESR spectra of the benzoquinone contants a quintet (Five equally spaced lines) (Fig. 26.16).



Fig. 26.16 ESR Spectra of 1, 4-benzoguinone radical anion

#### 26.8 APPLICATIONS OF ESR SPECTROSCOPY

The ESR spectroscopy has not wider applications as in NMR spectroscopy, its applications are specific and are increasing consistantly. It has been used to study minute quantities of para magnetic species. ESR spectrum is helpful to give the following information:

- (i) The site of unpaired electrons can be known.
- (ii) The number and type of nuclei present in the vicinity of the odd electron can be found.
- (iii) The type of nuclei that is responsible for the splitting patern can be determined by the relative intensities of the spectrum lines in the ESR spectra.
- (iv) The value of g can be found by comparing the position of the line with that of a reference e.g., DPPH has a g value = 2.0036.

The interpretations of the ESR spectrum is easy provided the spectrum is completely resolved.

Various application of ESR spectroscopy are given below:

1. Study of Free Radicals: ESR is used for the study of free radicals even in low concentrations. As an example DPPH radical can be in as low as  $10^{-12}$  gm amount. For study, the radicals can be produced photochemically, chemically or by high energy radiation. For good results, free radicals should be produced in  $10^{-13}$  mold  $dm^{-3}$  concentration. The number of free radicals can be determined from the intensity of the ESR spectrum.

The ESR spectra of methyl radical and 1,4-benzoquinone radicals has been shown in Figs. 26.12 and 26.15 respectively.

A typical application of ESR is the attachment of unpaired electron to macromolecules like proteins to obtain information about their structures in the ESR spectra.

**2. Determination of Rreaction Mechanisms:** A number of reactions proceed by radical mechanism. The radicals formed are very reactive but are unstable. For ESR study the rapid-flow system is used. In this procedure, a specially designed cell is used, which permits, the compounds whose interation yields paramagnetic intermediates to flow immediately through the observation area. This procedures maintain a steady concentration of the radicals. Using this technique radicals of life time as low as 0.01 second can be characterised. A typical example is the characterisation of  $\dot{CH}_2$  OH redical.

This radical contains a set of two equivalent protons and is produced in a flow system by the reaction of methyl alcohol with  ${\rm Ti}^{3+}$  and  ${\rm H_2O_2}$ 

$$\begin{split} T_i^{3+} + H_2O_2 & \longrightarrow T_i^{4+} + \dot{O}H + O\overline{H} \\ T_i^{3+} + \dot{O}H & \longrightarrow T_i^{4+} + OH^- \\ \dot{O}H + H_2O_2 & \longrightarrow H_2O + \dot{O}_2H \\ CH_3OH + \dot{O}H & \longrightarrow \dot{C}H_2OH + H_2O \end{split}$$

The  $\dot{\text{CH}}_2\text{OH}$  radical is produced outside the reaction cavity. In case the pH is low the OH proton does not contribute to hyperfine splitting as this proton is rapidly exchanged (*see* equations given above). The net results is 1:2:1 triplet from  $\dot{\text{CH}}_2$  protons (hiperfine splitting is 17.2*G*) (Fig. 26.17).



Fig. 26.17 First derivative ESR spectrum of CH2OH

The above procedure can also be used for the detection of  $CH_3\dot{C}HOH$  radical, which is generated by reaction an acidified solution of  $C_2H_5OH$  with hydrogen peroxide. The ESR spectra of  $CH_3\dot{C}HOH$  is shown in Fig. 26.18.



Fig. 26.18 First derivative ESR spectra of  $\mathrm{CH_{3}\dot{C}HOH}$ 

**3. Study of Inorganic Compounds:** ESR has also been used successfully in the study of Inorganic compounds.

A typical example is study of  $[NO(SO_3)_2]^{2-}$ . Its ESR spectrum in CHCl<sub>3</sub> yields a triplet, which arises by the interaction of the spin of unpaired electron with the spin of <sup>14</sup>N nucleus (I = 1). The results confirm that the electron is located on nitrogen atom. The ESR spectra of  $[NO(SO_3)_2]^{2-}$  is given in Fig. 26.19.

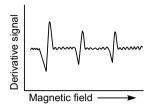


Fig. 26.19 ESR Spectra of [NO(SO<sub>3</sub>)<sub>2</sub>]<sup>2-</sup>

Another example is the determination of the structure of the complex  $[(NH_3)_5$  Co—O—Co $(NH_3)_5]^{5+}$ . Its ESR spectrum shows fifteen lines. On the basis of the study it is inferred that the structure is a mixture of:

- (i) Two cobalt (III) atoms are connected by an  $O_2^-$  bridge, and
- (ii) There are two equivalent cobalt atoms, the unpaired electron is undergoing equal interaction with both the cobalt atoms.
- **4. Analytical Applications: Determination of Mn<sup>2+</sup>:** Its ESR spectra shows six lines. The multiplicity is given by 2I + 1. For Mn<sup>2+</sup> ions, I is  $\frac{5}{2}$ . So the multiplicity is  $2 \times \frac{5}{2} + 1 = 6$  (Fig. 26.20).

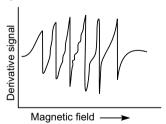


Fig. 26.20 ESR spectrum of Mn<sup>2+</sup>

ESR studies has also been used for determination of vanadium (in petroleum products) and polynuclear hydrocarbons and for the estimation at copper (II), chromium (III), iron (III) and titanium (III).

#### 5. Biological Systems

(i) The presence of free radicals can be studied in diseased and healthy tissues.

(ii) The presence of oxidative enzymes which function via. one electron redox reactions and produce enzyme bound free radical can be conformed by ESR studies.

#### 6. Miscellaneous Applications

- (i) It has been confirmed by ESR studies that the electrons in impurities are responsible for the conducting properties of semiconductors.
- (ii) Detection of electrons in solution of alkali metals in liquid ammonia. alkaline earth metals etc. is possible by ESR studies.
- (iii) Complexes of copper, nickel, cobalt and sodium with malenonitrile dithiolate can be studied by ESR.

#### 26.9 ELECTRON NUCLEAR DOUBLE RESONANCE (ENDOR)

This technique has been used for improving the resolution of an ESR spectrum. The method involves the irradiation of a sample simultaneously with microwave frequency (which is suitable for electro resonance) and a radio frequency (which is suitable for nuclear resonance). Subsequently the radio frequency is swept and ESR sprectrum under microwave saturation is observed. For this type of studies a spectrometer of high intensity is required. ENDOR procedure is best in the following cases:

- It is suitable for cases where there is broadening of normal electron resonance lines.
- Suitable for cases where hyperlines are resolved but precise values of the hyperfine coupling are the need.
- Useful in cases where quadrupole couplings are to be measured in a system.

## 26.10 ELECTRON DOUBLE RESONANCE (ELDOR)

This technique involves simultaneous irradiation of the sample with two microwave frequencies. In this case, the ESR signal height is a function of the difference of two microwave frequencies. ELDOR is useful to study relaxation mechanisms. In fact, precise coupling constants of DPPH can be made by this technique.

# EXERCISES

- 1. What is the principle of ESR? In what respect ESR spectroscopy differ from NMR spectroscopy.
- 2. What type of substances can be analysed by ESR spectroscopy?
- 3. Describe the various components at an ESR instrument explaining their functions giving a schematic diagram.

4. Which of the following will show ESR spectrum?

(a) 
$$Cu^+$$
, (b) H, (c) H<sub>2</sub>, (d)  $K^+$  (e)  $Cl^-$ , (f)  $\dot{C}H_3$ , (g)  $NO_2$ , (h)  $CO_2^-$ .

**Ans.** H<sub>2</sub>, K<sup>+</sup>, Cl<sup>-</sup> will not show ESR spectrum due to absence of unpaired electrons.

H, CH<sub>3</sub> will show ESR spectrum as both these have unpaired electron.

 $NO_2$  has an odd electron in the  $4a_1$  orbital

$$[(1a_1)^2 (1b_1)^2 (2a_1)(2b_1)^2 (1b_2)^2 (3a_1)^2 (1a_2)^2 (3b_1)^2 (4a_1)^1]$$

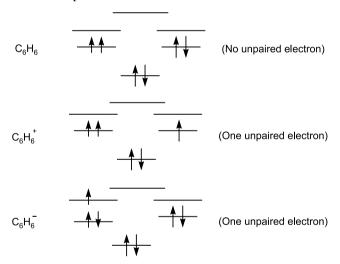
So it will show ESR spectrum.

CO<sub>2</sub> being isoelectronic with NO<sub>2</sub> has one unparied electron and so will show ESR spectrum.

- 5. Which of the following will show ESR spectrum?
  - (a) Benzene,  $C_6H_6$ , (b) Benzene cation  $C_6H_6^+$ , (c) Benzene anion  $C_6H_6^-$ .

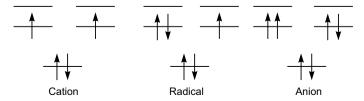
**Ans.** Out of these benzene cation and benzene anion will show ESR spectrum since these have one unpaired electron each.

On the basis at HMO calculations,  $C_6H_6$ ,  $C_6H_6^+$  and  $C_6H_6^-$ , the *p*-electron distribution is represented as:



**6.** In case of the species, cyclopentadiene, which of the systems, *viz.*, cation, radical or anion will show ESR spectrum.

The MHO energy level of cyclopentadiene is given below.



The anion has no unpaired electron, the radical has one and the cation has two unpaired electrons. So the anion does not show ESR spectrum but the cation and radical do show ESR spectrum.

7. Explain ESR spectra of H atom, CH<sub>3</sub> and 1, 4-benzosemiquinone anion—whether the spractra is a doublet, triplet or triplet?

**Ans.** H atoms-doublet of equal intensity;  $\dot{C}H_3$  radical—a quartet of intensity 1:3:3:1 and 1,4-benzosemiquinone anion—a quintet in the intensity ratio 1:4:6:4:1.

- **8.** Write notes on:
  - (a) Determination of g value
  - (b) Hyperfine splitting
  - (c) Paramagnetic substances
  - (d) Sensitivity of ESR spectrometer.
- **9.** Discuss the applications of ESR spectroscopy.





# Chapter 27

# **Mass Spectrometry**

#### 27.1 INTRODUCTION

Mass spectrometry is a useful technique for determination of molecular weight and molecular formula of an unknown compound. In this technique, mass spectrometer bombards a substance under investigation with electron beam. The result is recorded as a spectrum of positive ion fragments. This record is known as mass spectrum. The positive ion fragments are separated on the basis of mass.

In mass spectrometer, the molecule weight of the compound under investigation is recorded as the nearest whole number. In fact, the recording should be such that a peak at say mass 400 can be distinguish from a peak at mass 401 or at mass 399. It has been arbitarily decided that a valley between two adjacent peaks should not be more than about 10 per cent of the height of the larger peak. This is termed "unit" resolution. The resolution of an instrument can be determined by considering two adjacent peaks of approximately equal intensity. The selection of the peaks should be such that the height of the valley between the peaks is about 10 per cent of the intensity of the peaks (Fig. 27.1).

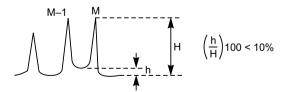


Fig. 27.1 Determination of resolution of an mass spectrometer

The Resolution (R) is

$$R = \frac{M}{\Lambda M}$$

where, M is the higher mass number of the two peaks and  $\Delta M$  is the difference between the two mass numbers.

Mass spectrometers, in general are of two types, viz., low resolution and high resolution. The former, *i.e.*, low resolution instruments sparate unit mass up to m/e 2000; in this case R = 2000/(2000 - 1999) = 2000. Such instruments give low resolution spectras. The latter type viz., high resolution instruments can separate two ions differing in mass by at least one part in ten thousand to fifteen thousand; in this case, R = 10,000 - 15,000. Such an instrument of resolution 10,000 can separate an ion of mass 500.00 from one at mass 499.95 (R = 500/0.05 = 10,000).

#### 27.2 THE MASS SPECTROMETER

A diagrammatic representation of a mas spectrometer is given in Fig. 27.2.

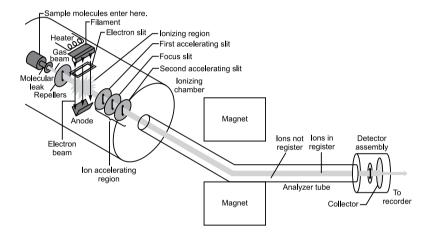


Fig. 27.2 A diagrammatic representation of various parts of a mass Spectrometer

A mass spectrometers has five components:

- (i) Sample Handling Component: This has a device for the introduction of the sample, a device (micrometer) for knowing the amount of the sample injected, a device for metering the sample to the ionization chamber and a pumping system.
- (ii) Ionization and Accelerating Chambers: The sample in the form of gas stream enters the ionization chamber at a pressure of about 10<sup>-6</sup> to 10<sup>-5</sup> Torr, where it is bombarded at right angles by an electron beam from a heated filament. This results in the formation of positive ions, which passes through first accelerating slit due to a small electrostatic field between the repellers and the first accelerating slit. A stronger electrostatic field between the first and second accelerating slits accelerates the positive ions to the final required velocities. A magnetic

field applied to the analyser tube are focused at the collector slit as a function of mass (mass/charge).

- (iii) Analyzer Tube and Magnet: The ion beam passes through the analyser tube (made of metal) evacuated at  $10^{-7}$  to  $10^{-8}$  Torr to the collector. The analyser tube passes through a uniform and stable magnet field.
- (*iv*) **Ion Collector and Recorder:** It consists of one or more collimating slits. The ion beam strikes the collector and the signal is amplifield and recorded in the form of a graph.

A mass spectrum is a plot of the distribution of ion masses corresponding to the formula weight of a molecule and fragments derived from the molecule. The height of each peak (measured against the *y*-axis) indicates the relative amount of each ion produced. Peaks along the *x*-axis of a mass spectrum give the distribution of masses produced from the molecule of the compound.

Mass spectrometry can be used for analysing very small samples (of the order of  $10^{-12}$ g). Even complex mixtures (that are isolatated as natural products) can be analysed; in the latter case the mixture is separated by gas or liquid chromatography and then data obtained for each component.

In a mass spectrometer following three basic tasks are accomplished:

- (i) Vapourization of the Molecule: This is accomplished by heating a sample under high vacuum (low pressure). Under these conditions, the collision between the ions and molecules are minimum; this procedure makes the analysis simple.
- (ii) Production of Ions from the Molecules in Gas-phase: In mass spectrometer, the molecules in the gaseous phase are bombarded with a beam of energetic electrons, 70 electron volts (eV). The simplest event that occurs is the formation of a molecular ion by removal of one electron from the parent molecule.

$$M \rightarrow M^+ + e^-$$

The molecular ion is in fact a radical cation M<sup>+</sup>.

- (iii) Fragmentation: It is caused by the energy of the electron impact process (70 eV), which imparts enough vibrational energy to the molecular ion (For more details about fragmentation section 27.7).
- (iv) Separation of the Ions by their Mass-to-Charge Ratio: The mass spectrometer finally separates the various ions on the basis of their mass-to-charge ratio. These are recorded as peaks and a mass spectrum is obtained. A typical mass spectrum is given below (Fig. 27.3).

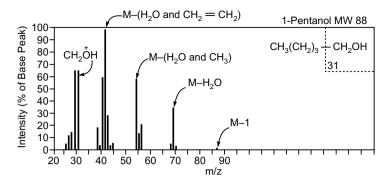


Fig. 27.3 Mass spectrum of 1-pentanol. (Adapated from Silverstein, R.M.; Webster F.X., Spectrometric Identification of Organic Compounds, 6th ed.; Wiley: New Yourk, 1998; p 19.)

#### 27.3 THE MASS SPECTRUM

A mass spectrum represents the masses of the positively charged fragments (including the molecular ion) versus their relative concentration. Mass spectras are usually obtained by an electron beam energy of 70 electron volts. The simplest event that occurs is the removal of an electron from the molecule in the gas phase. For example, ammonia forms a molecular ion:

$$H: \mathring{\mathbf{N}}: \mathbf{H} + \mathbf{e}^{-} \longrightarrow \left[ H: \mathring{\mathbf{N}}: \mathbf{H} \right]^{+} + 2\mathbf{e}^{-}$$

$$\mathbf{molecular ion}$$

The molecular ion, as soon as it is formed undergoes fragmentation as shown below:

H:
$$\dot{\vec{N}}$$
: + H°  
H: $\dot{\vec{N}}$ : + H°  
 $m/z$  15  
H: $\dot{\vec{N}}$ : + H°

The fragmentation is normally represented as:

In the mass spectrometer, only the positive ions are detected. The radicals since they are not charged, are not detected by the magnetic field. The mass spectra of ammonia is given in Fig. 27.4. The mass spectras are usually represented as bar

graphs or in tabular form (*see* Fig. 27.4). The most intense peak (in either of the representation) is called the base peak; it is arbitrarily assigned an intensity of 100%. All other peaks are given proportionate values as percentages of the base peak. In mass spectrum, the masses of the ions are calculated on the bases of whole number masses, which are 1, 12, 14, 16 and 19 for H, C, N, O and F respectively.

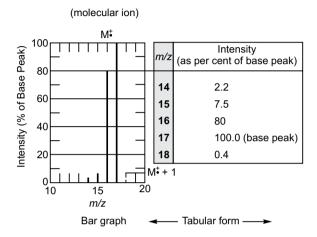


Fig. 27.4 Mass Spectra of Ammonia

In case of ammonia (Fig. 27.4), the base peak is due to the molecular ion ( $M^{+}$ ). This is not always the case. As we will see subsequently, a base peak (the most intense peak) will be different from the molecular ion. This is because in these cases the molecular ion fragments so rapidly that some other ion at a smaller m/z value produces the most intense peak. At times, in a few cases, the molecular ion peak is too small and sometimes it is absent.

A special feature in the spectrum of ammonia (Fig. 27.4) is that a small peak occurs at m/z 18; this is due to  $M^{+} + 1$  (unit mass greater than the molecular ion). The  $M^{+} + 1$  peak normally appears in the spectrum as most elements (*e.g.*, N and H) have more than one naturally occurring isotopes (Table 27.1). The ammonia molecule consists mainly of  $^{14}N^{1}H_{3}$ . However, a very small fraction of ammonia consists of  $^{15}N^{1}H_{3}$  and also of  $^{14}N^{1}H_{2}^{2}H$ . These molecules ( $^{14}N^{1}H_{3}$  or  $^{14}N^{1}H_{2}^{2}H$ ) produce molecular inos at m/z 18, *i.e.*, at  $M^{+} + 1$ .

The mass spectrum helps to establish the structure of a new compound by determination of molecular formula and molecular weight. If we take the mass spectrum of an unknown compound and find it identical with the mass spectrum of a previously reported compound of known structure, then it is concluded almost beyond doubt that the two compounds are identical.

Element	Common Isotope		Isoto	ural Abund pes as per toms of Co	cent (bas	sed on
Carbon	<sup>12</sup> C	100	<sup>13</sup> C	1.11		
Hydrogen	<sup>1</sup> H	100	<sup>2</sup> H	0.016		
Nitrogen	<sup>14</sup> N	100	<sup>15</sup> N	0.38		
Oxygen	<sup>16</sup> O	100	<sup>17</sup> O	0.04	<sup>18</sup> O	0.20
Fluorine	<sup>19</sup> F	100				
Silicon	<sup>28</sup> Si	100	<sup>29</sup> Si	5.10	<sup>30</sup> Si	3.35
Phosphorus	<sup>31</sup> P	100				
Sulphur	<sup>32</sup> S	100	<sup>33</sup> S	0.78	<sup>34</sup> S	4.40
Chlorine	<sup>35</sup> CI	100	<sup>37</sup> CI	32.5		
Bromine	<sup>79</sup> Br	100	<sup>81</sup> Br	98.0		
lodine	127	100				

Table 27.1 Principal Stable Isotopes of common elements<sup>a</sup>

#### 27.4 DETERMINATION OF MOLECULAR FORMULA

### 27.4.1 Molecular Formula from Isotopic Peaks

We know that most of the common elements found in organic compounds have naturally occurring heavier isotopes (*see* Table 27.1). The principal heavier isotope in carbon, hydrogen and nitrogen is one mass unit greater than the common isotope. So, the presence of these elements (C, H and N) in a compound gives a small peak with mass one unit greater than the molecular ion. This peak is called  $M^{\dagger} + 1$  peak. However, in case of oxygen, sulphur, chlorine and bromine, the principal heavier isotope is two mass units greater than the common isotope. So the presence of these elements (O, S, Br, Cl) gives rise to an isotopic peak at  $M^{\dagger} + 2$ .

Molecular formulae can be determined from the isotopic peak. This can be easily understood from the following example. Let us consider the mass spectrum of methane (Fig. 27.5).

The isotopic abundance in Table 27.1 are based on 100 atoms of the normal isotope. Let us now consider that we have 100 molecules of methane (CH<sub>4</sub>). On an average, there will be 1.11 molecules that contain  $^{13}$ C and  $4 \times 0.016$  molecules that contain  $^{2}$ H. These heavier isotopes ( $^{13}$ C and  $^{2}$ H) together contribute an M<sup>+</sup> + 1

<sup>&</sup>lt;sup>a</sup>Data taken from Silverstien, R.M.; Webster, F.X. *Spectrometric Identification of Organic Compounds* 6<sup>th</sup> ed.; Wiley: New York, 1998; p. 7.

peak, whose intensity is about 1.17% of the intensity of molecular ion peak (1.11 +  $4 \times 0.016$ ). This figure is in agreement with the actual spectrum given in Fig. 27.5.

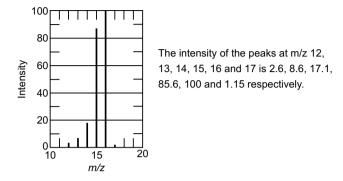


Fig. 27.5 Mass spectra of methane

In case  $M^{\dagger}$  peak is not the base peak, the molecular formula is determined as follows: Consider the mass spectra of an unknown compound (which gives the peaks at different m/z) in which the molecular ion peak ( $M^{\dagger}$ ) is different from the base peak (Fig. 27.6).

m/z	Intensity (as per cent of base peak)
27	59.0
28	15.0
29	54.0
39	23.0
41	60.0
42	12.0
43	79.0
44	100.0 (base)
72	73.0 M <sup>‡</sup>
73	3.3 M <sup>‡</sup> + 1
74	0.2 M <sup>†</sup> + 2

Fig. 27.6 Mass spectra (Tabular form) of an unknown compound

The first step is to recalculate the intensities of the  $M^{+} + 1$  and  $M^{+} + 2$  peaks so as to express them as the percentage of the  $M^{+}$  peak. This is done by dividing each

372

intensity by the intensity of the  $M^{\dagger}$  peak, which is 73% and multiplying by 100. The results obtained are given below.

m/z	Intensity (% of M*)
72	73.0/73 × 100 = 100
73	3.3/73 × 100 = 4.5
74	0.2/73 × 100 = 0.3

m/z	Intensity (as per cent of M•)		
72	M*	100.0	
73	M* + 1	4.5	
74	M* + 2	0.3	

The molecular formula is finally determined by the following steps:

- (i) Presence of Nitrogen: The presence of nitrogen in an organic compound is determined by Nitrogen rule. According to this rule, if M<sup>†</sup> is even then the compound must contain an even number of nitrogen atoms (zero is an even number). For the unknown compound, M<sup>†</sup> is even and so the compound must have an even number of nitrogen atoms (Remember that zero is an even number.).
- (ii) Number of Carbon Atoms: The number of carbon atoms is determined by the relative abundance of  $M^{+} + 1$  peak. In the unknown compound the relative abundance of  $M^{+} + 1$  peak is 4.5 %.

∴ Number of C atoms = 
$$\frac{\text{Relative abundance of } M^+ + 1 \text{ peak}}{1.1}$$
  
=  $\frac{4.5}{1.1} = 4$ .

( $^{13}$ C is the most important contributor to the  $M^{\dagger}+1$  peak having natural abundance of about 1.1%).

- (iii) The presence or absence of S, Cl and Br is indicated by the relative abundance of M<sup>+</sup> + 2 peak, which in the case of unknown compound is only 0.3% (see Table 27.1 for the natural abundance of isotopes of S, Cl and Br). In view of the above, it is assumed that S, Cl and Br are absent.
- (*iv*) The compound is expected to contain C, H and or oxygen. For the unknown compound, the M<sup>†</sup> peak at *m/z* 72 given the molecular weight. Since N is absent (or even number) [*see* step (*i*) above)], the compound contains four carbons (as established in (*ii*) above) and two nitrogen (to get even molecular weight), the molecular weight (76) is greater than that for the unknown compound (M<sup>†</sup> is 72).

For a compound having only C and H

$$H = 72 - (4 \times 12) = 24.$$

So, the formula  $C_4H_{24}$  is not possible.

For a compound having C, H and one O.

$$H = 72 - (4 \times 12) - 16 = 8$$

 $\therefore$  The unknown compound has the molecular formula  $C_4H_8O$ .

The above calculations can be easily done using computers and tables are available that give the relative values for the  $M^{\ddagger}+1$  and  $M^{\ddagger}+2$  peaks for all combinations of common elements with molecular formulae up to mass 500. Table 27.2 gives part of the data obtained from these tables. Using data in the results of the molecular formula ( $C_4H_8O$ ) in the above example can be checked.

<b>Table 27.2</b> Relative Intensities of M <sup>+</sup> + 1 and M <sup>+</sup> + 2 peaks for various
combinations of C, H, N, and O for masses 72 and 73

		Percentage of M <sup>‡</sup> Intensity				Percent M.+ Inte	_
M <sup>+</sup>	Formula	M <sup>+</sup> +1	M <sup>+</sup> +2	M <sup>+</sup>	Formula	M <sup>+</sup> +1	M <sup>‡</sup> +2
72	CH <sub>2</sub> N <sub>3</sub> O	2.30	0.22	73	CHN <sub>2</sub> O <sub>2</sub>	1.94	0.41
	CH <sub>4</sub> N <sub>4</sub>	2.67	0.03		CH <sub>3</sub> N <sub>3</sub> O	2.31	0.22
	C <sub>2</sub> H <sub>2</sub> NO <sub>2</sub>	2.65	0.42		CH <sub>5</sub> N <sub>4</sub>	2.69	0.03
	C <sub>2</sub> H <sub>4</sub> N <sub>2</sub> O	3.03	0.23		C <sub>2</sub> HO <sub>3</sub>	2.30	0.62
	C <sub>2</sub> H <sub>6</sub> N <sub>3</sub>	3.40	0.04		C <sub>2</sub> H <sub>3</sub> NO <sub>2</sub>	2.67	0.42
	$C_3H_4O_2$	3.38	0.44		C <sub>2</sub> H <sub>5</sub> N <sub>2</sub> O	3.04	0.23
	C <sub>3</sub> H <sub>6</sub> NO	3.76	0.25		C <sub>2</sub> H <sub>7</sub> N <sub>3</sub>	3.42	0.04
	$C_3H_8N_2$	4.13	0.07		C <sub>3</sub> H <sub>5</sub> O <sub>2</sub>	3.40	0.44
	C₄H <sub>8</sub> O	4.49	0.28		C <sub>3</sub> H <sub>7</sub> NO	3.77	0.25
	C <sub>4</sub> H <sub>10</sub> N	4.86	0.09		C <sub>3</sub> H <sub>9</sub> N <sub>2</sub>	4.15	0.07
	C <sub>5</sub> H <sub>12</sub>	5.60	0.13		C₄H <sub>9</sub> O	4.51	0.28
					C <sub>4</sub> H <sub>11</sub> N	4.88	0.10
					C <sub>6</sub> H	6.50	0.18

<sup>&</sup>lt;sup>a</sup>Data from Beynon, J.H. *Mass Spectrometry and its Applications to Organic Chemistry*; Elsevier, Amsterdam, 1960; 489.

# 27.4.2 Molecular Formula Using High-Resolution Mass Spectrometry

Compared to the usual low-resolution mass spectrometers (which measure m/z values to the nearest whole-number mass unit), the high-resolution mass spectrometers can measure m/z values to three or four decimal places. This provides an accurate method for determination of molecular weights. Thus, the determination of accurate molecular weight can be used to determine the molecular formula.

Using the data given in Table 27.2, giving the exact masses of the nuclides, the molecular wieght and molecular formula can be determined. As an example, the three molecules  $O_2$ ,  $N_2H_4$  and  $CH_3OH$  all have atomic masses of 32 using the normal values for the atomic weights of O, N, H and C as 16, 14, 1 and 12 respectively. However, the actual atomic masses of the three molecules are different.

$$O_2 = 2(15.9949) = 31.9898$$
  
 $N_2H_4 = 2(14.0031) + 4(1.00783) = 32.0375$   
 $CH_4O = 12.00000 + 4(1.00783) + 15.9949 = 32.0262$ 

Thus, high-resolution spectrometers can easily distinguish among the three molecules (O<sub>2</sub>, N<sub>2</sub>H<sub>4</sub>, CH<sub>4</sub>O) and this is helpful for the determination of the molecular formula.

Isotope	Mass	Isotope	Mass
<sup>1</sup> H	1.00783	<sup>19</sup> F	18.9984
<sup>2</sup> H	2.01410	<sup>32</sup> S	31.9721
<sup>12</sup> C	12.00000 (std)	<sup>33</sup> S	32.9715
<sup>13</sup> C	13.00336	<sup>34</sup> S	33.9679
<sup>14</sup> N	14.0031	<sup>35</sup> Cl	34.9689
<sup>15</sup> N	15.0001	<sup>37</sup> Cl	36.9659
<sup>16</sup> O	15.9949	<sup>79</sup> Br	78.9183
<sup>17</sup> O	16.9991	<sup>81</sup> Br	80.9163
<sup>18</sup> O	17.9992	127	126.9045

Table 27.3 Exact Masses of Nuclides

#### 27.5 RECOGNITATION OF THE MOLECULAR ION PEAK

Recognitation of the molecular ion peak in mass spectra is most important in order to find the molecular mass of the compound. It has already been stated that removal of one electron from the parent molecule gives the molecular ion (*see* Section 27.2).

$$M \rightarrow M^{+} + e^{-}$$

The m/z value of the moecular ion peak is the molecular wieght of the compound. It is found that some times the  $M^{\dagger}$  peak is the base peak and is easily recognised (*see* Fig. 27.2). However, in certain cases the molecular ion peak is not the base peak and is very small. In such cases some effort has to be made to locate the molecular ion peak.

It is known that most elements occur naturally as several isotopes (*see* Table 27.1)–generally the lightest one greatly predominates and the heavier ones occur to a lesser extent. Thus, the molecular ion peak,  $M^+$  is due to the molecules containing only the commonest isotope of each element. Let us consider the case of benzene. In this case, the  $M^+$  peak, m/z 78 is due only to ions of the  $C_6H_6^+$ . There is also a peak at m/z 79, the  $M^+$  + 1 peak, which is due to  $C_5^{13}CH_6^+$  and  $C_6H_5D^+$ . In addition, a  $M^+$  + 2 peak at m/z 80 also appears; this is due to  $C_4^{13}C_2H_6^+$ ,  $C_5^{13}CH_5D^+$  and  $C_6H_4D_2^+$ . Due to low natural abundance of most heavy isotopes, these isotopic peaks are generally much less intense than the  $M^+$  peak. In the case of benzene, the  $M^+$  1 and  $M^-$  2 peaks are respectively 6.75% and 0.18% as intense as the  $M^+$  peak. In case of monochloro compound, the  $M^+$  2 peak is about one third as intense as the  $M^+$  peak (*see* Table 27.1) and a monobromo compound will give  $M^+$  + 2 peak of about equal intensity (Table 27.1).

Thus, on the basis of the isotopic peaks,  $M^+ + 1$  and  $M^+ + 2$ , it is possible to recognise the molecular ion peak.

The molecular ion peak can also be identified from the Fragmentation of the molecule (see section 27.7). Thus the presence of M-15 peak (loss of  $CH_3$ ) or an M-18 peak (loss of  $H_2O$ ) or an M-31 peak (loss of  $OCH_3$  from methyl esters) etc. is taken as a confirmation of a molecular ion peak).

Besides the methods described above for the recognitiation of the molecular ion peak, few other methods are also available, some of which are described below.

**Chemical ionization method:** The method consist in introduction of the sample at about 1 Torr pressure with carrier gas like methane. The methane ionizes by the impact of electron to give primary ions  $(CH_4^+, CH_3^+ \text{ etc.})$ , which in turn react with excess methane to give secondary ions  $(CH_5^+, C_2H_5^+ \text{ etc.})$ .

$$CH_4^+ + CH_4 \rightarrow CH_5^+ \text{ and } CH_3^-$$
  
 $CH_3^+ + CH_4 \rightarrow C_2H_5^+ \text{ and } H_2$   
 $CH_4 + C_2H_5^+ \rightarrow C_3H_5^+ + 2H_2$ 

Finally the sample reacts with the secondary ions.

$$\begin{aligned} \mathrm{RH} + \mathrm{CH_5}^+ &\to \mathrm{RH_2}^+ \, \mathrm{and} \, \, \mathrm{CH_4} \\ \mathrm{RH} + \mathrm{C_2H_5}^+ &\to \mathrm{RH_2}^+ \, \mathrm{and} \, \, \mathrm{C_2H_4} \end{aligned}$$

The formed M+1 ions (quasi – molecular ions) normally give prominent peaks. The quasi-molecular ions undergo fragmentation by loosing  $H_2$  to give prominent M-1 ions. As, the M+1 ions (produced chemically) do not have excess of energy associated with ionization by electron impact, they undergo much less framentation and can be easily recognised.

**Field Ionization Technique:** This technique requires reasonable concentration of the sample in the gas phase. The molecules is the gaseous phase approach a

metal surface having high positive potential ( $10^8$  volts/cm); this causes an electron to "tunnel" from the molecule of the sample to the metal surface. In this process there is no excitation at the sample molecules and the molecular ion is quickly accelerated. In this way there is less fragmentation and a more intense molecular ion peak results.

**Field desorption technique:** In this technique, the solid sample is directly placed on the field ion emitter, which is operated at slightly above room temperature. The ions thus produced have little or no internal energy. This results in the formation of molecular ions predominately. These ions are desorved from the solid by a strong electric field.

#### 27.6 USE OF THE MOLECULAR FORMULA

As we know, the molecular formula gives the kinds and the number of atoms present in a compound. Besides these, the molecular formula gives the index of hydrogen deficiency. It is defined as the number of pairs of hydrogen atoms that must be removal from a 'saturated' formula (e.g.,  $C_nH_{2n+2}$  for alkanes) to produce a molecular formula of the compound of interest. Hydrogen deficiency can also be due to cyclic structure and multiple bonds. Thus, the index is the sum of rings, number of double bonds and twice the number of triple bonds.

For compounds containing C, H, N, halogen, O and S, the index of deficiency can be calculated from the formula

$$Index = carbons - \frac{hydrogens}{2} - \frac{halogens}{2} + \frac{nitrogen}{2} + 1$$

As as example, a compound  $C_7H_7$  NO has an index of 7 - 3.5 + 0.5 + 1 = 5.

In the above calculations divalent atoms (oxygen and sulfur are not counted).

For a compound containing monovalent atoms (H,D), bivalent atoms (O,S), trivalent atoms (N,P) and tetravalent atoms (C,Si), the hydrogen deficiency index is calculated as follows:

For a generalised molecular formula  $A_I B_{II} C_{III} D_{IIV}$ 

$$Index = IV - \frac{1}{2} + \frac{III}{2} + 1$$

where.

A is a monovalent atom (H, D or halogen)

B is a bivalent atom (O, S)

C is a trivalent atom (N, S)

D is tetravalent atom (C, Si)

*I, II, III, IV* designate the numbers of mono, di-, tri-, and tetravalent atoms respectively. Thus for a compound  $C_{13}H_9 N_4O_4$  Br S

Index = 
$$13 - \frac{10}{2} + \frac{2}{2} + 1 = 10$$

The index of hydrogen deficiency is the above case  $(C_{13}H_2 N_4O_4BrS)$  is in consistent will the structure

$$O_2N$$
  $\longrightarrow$   $S$   $\longrightarrow$   $CH_2$   $\longrightarrow$   $Br$ 

(Index of hydrogen deficiency = 4 for each benzene ring and 1 for each  $NO_2$  group).

#### 27.7 FRAGMENTATION

The first step in mass spectrometry is the formation of a molecular ion  $(M^{\dagger})$ . This molecular ion can break apart in a number of ways. We say that the molecular ion undergoes fragmentation. The fragmentation and assigning the structure to the fragmentated species is an important step in the structure elucidation of an organic compound. The fragmentation of a molecular ion can take place in a number of ways. Some of the ways are discussed below:

#### 27.7.1 Fragmentation by Cleavage of a C—C Single Bond

As an example, the molecular ion obtained from propane can fragment in two ways (two possible modes of cleavage). A fish-hook arrow (\( \sigma \)) is the shift of a single electron

In the mass spectrum of propane, the peak at m/z 29 (CH<sub>3</sub>CH<sub>2</sub><sup>+</sup>) is most intense and the peak at m/z 15 (CH<sub>3</sub><sup>+</sup>) has an intensity of only 5.6%. Thus, the two modes of cleavage do not take place at equal rates. The mode which results in the formation of more stable carbocation is the one which takes place in preference to the route which gives less stable carbocation. This reflects greater stability of CH<sub>3</sub>CH<sub>2</sub><sup>+</sup> compared to CH<sub>3</sub><sup>+</sup>. It is to be noted that the peaks due to free radicals (CH<sub>3</sub> CH<sub>2</sub><sup>-</sup> and CH<sub>3</sub><sup>-</sup>) are not recorded in the mass spectra. This is because the radicals are not charged and are not deflected by the magnetic field and so are not detected.

In a long chain hydrocarbons (more than 3 carbons), it is not possible to localize the charge and the odd electron arbitrarily. So, as a matter of convention, the formula for the radical cations put in brackets and the charge and the odd electrons are placed outside as shown below:

$$[CH_3CH_2CH_3]^{\ddagger} \longrightarrow CH_3CH_2^{\phantom{2}\ddagger} + \cdot CH_3$$
$$[CH_3CH_2CH_3]^{\ddagger} \longrightarrow CH_3CH_2 \cdot + {\phantom{1}}^{\ddagger}CH_3$$

However, in case a compound contains an oxygen, or nitrogen, the charged and the odd electrons are placed on the hetero atoms.

Which electron is removed from the parent molecule? The electron which is most loosely held is removed. Which electron is this, is usually estimated from the ionization potentials (Table 27.4). The ionization potential of a molecule is the amount of energy (in eV) required to remove an electron from the molecule.

Compound	Ionization Potential (eV)
CH <sub>3</sub> (CH <sub>2</sub> ) <sub>3</sub> NH <sub>2</sub>	8.7
C <sub>6</sub> H <sub>6</sub> (benzene)	9.2
$C_2H_4$	10.5
CH₃OH	10.8
$C_2H_6$	11.5
CH <sub>4</sub>	12.7

Table 27.4 Ionization potentials of selected molecuels

In case of branched chain alkanes, the cleavage occurs at a branch point because a more stable carbocation results. Thus, 2-methylbutane a peak at  $M^+$  – 15 is observed due to loss of a methyl radical from the molecular ion.

In a similar way, neopentane (2,2-dimethyl propane) gives a peak at m/z 57 (M<sup>+</sup> – 15) due to loss of methyl radical from the molecular ion; in this case the peak (m/z 57) is attributed to a tertiary carbocation.

Another example of a tertiary carbocation being formed is in the mass spectra of 3, 3-dimethyl heptane.

In such a case (illustration cited above), the larger group is normally lost as the radial portion.

The fragmentation of the hydrocarbon portion of a molecule occurs to produce the more stable carbocation  $(3^{\circ} > 2^{\circ} > 1^{\circ})$ . In the mass spectrum, the carbocations that are resonance stabilized are produced. Some examples are given below:

(a) Allylic carbocations are obtained by the fragmentation of alkenes.

$$CH_2 \xrightarrow{+\cdot} CH \xrightarrow{\cdot} CH_2 \xrightarrow{\cdot} R \xrightarrow{\cdot} CH_2 \xrightarrow{\cdot} CH = CH_2 + \cdot R$$

$$m/z \ 41$$

(b) Carbon-carbon bonds, which are next to an atom with an unshared electron pair usually undergo cleavage as the resulting carbocation is resonance stabilized.

$$R - \overset{+}{Z} \stackrel{\longleftarrow}{P} CH_2 \overset{\bullet}{\downarrow} CH_3 \longrightarrow R - \overset{+}{Z} = CH_2 + \overset{\bullet}{\bullet} CH_3$$

$$R - \overset{+}{Z} - \overset{+}{C}H_2$$

Z = N, O, or S; R may also be H

(c) Carbon-carbon bonds next to carbonyl groups, (as in case of aldehyde or ketone) cleave readily. In this case resonance stabilized ions known as acylium ion are produced.

This type of bond cleavage in case of carbonyl compounds is especially important.

Thus, for aliphatic ketones,  $\alpha,\beta$ -cleavage is the most common fragmentation pathways. These give rise to three fragment peaks as show below:

$$\alpha$$
,β-Cleavage

m/z = 114

 $m/z = 57$ 

or

$$m/z = 114$$

and

 $m/z = 85$ 

380

(d) Fragmentation of molecular ion peak obtained from alkyl substituted benzenes gives a prominent peak at m/z 91 due to the formation of tropylium ion.

$$CH_3$$

$$-H \rightarrow CH_3$$

$$Tropylium ion$$

$$CH_3$$

$$-CH_3$$

$$Tropylium ion$$

$$CH_3$$

$$Tropylium ion$$

The monosubstituted benzenes also loose their substituent and yield a phenylcation (m/z 77).

Y = halogen, 
$$-NO_2$$
,  $-CR$ ,  $-R$ , etc.

#### 27.7.2 Fragmentation by Cleavage of More than One Bond

Alcohols, cycloalkenes and carbonyl compounds with a hydrogen on  $\gamma$  carbon undergo fragmentation by cleavage of more than one bond.

(a) Fragmentation of molecular ion derived from alcohols gives a prominent peak at  $M^+$  – 18 due to loss of a water molecule.

$$R \xrightarrow{\vdots} OH \\ R \xrightarrow{\downarrow} CH_{2} \longrightarrow R \xrightarrow{} R \xrightarrow{-CH} CH_{2} + H \xrightarrow{} OH$$
or
$$[R - CH_{2} - CH_{2} - OH]^{\ddagger} \longrightarrow [R - CH = CH_{2}]^{\ddagger} + H_{2}O$$

$$M^{\ddagger} - 18$$

(b) Cycloalkenes (e.g., cyclohexene) undergo **retro-Diels-Alder reaction** to produce an alkene and alkadienyl radial cation.

$$\frac{70 \text{ eV}}{\text{CH}_2}$$

(c) Carbonyl compounds having a hydrogen on γ-carbon on subjecting to mass spectral analysis give a molecular ion, which undergo fragmentation known as **McLafferty rearrangement.** 

Y may be R, H, OR, OH, etc.

General fragmentation pathways of a number of classes of organic compounds are summarized in Table 27.5.

Table 27.5 Fragmentation pathways for some classes of organic molecules

Compound type	Fragmentation pathways
Alcohols Aldehydes Alkanes Alkenes Alkyl halides Carboxylic acids Esters Ketones	Loss of a neutral molecule (water, $m/z$ 18); $\alpha$ , $\beta$ -cleavage $\alpha$ , $\beta$ -Cleavage; McLafferty rearrangement (aliphatic) Cleavage at branches $\alpha$ , $\beta$ -Cleavage; rearrangement Loss of a neutral molecule (HX, $m/z$ 20, 36, 80 or 127) $\alpha$ , $\beta$ -Cleavage; McLafferty rearrangement (aliphatic) $\alpha$ , $\beta$ -Cleavage; McLafferty rearrangement (aliphatic) $\alpha$ , $\beta$ -Cleavage; McLafferty rearrangement (aliphatic)

On the basis of extensive studies a number of general rules for prediction of prominent peaks in electron impact spectra have been formulated as given below:

- (i) For straight chain compounds, the relative height of the molecular ion peak is greatest. The height decreases as the brancing increases
- (ii) In a homologues series, the relative height of the molecular ion peak, normally decreases with increasing molecular weight. However, fatty acids are exception.
- (iii) In alkyl substituted chains, the cleavage in favoured at the alkyl substituted carbons. The cleavage is more likely if the chain is more substituted. This is due to increased stability of a tertiary carbocation over a secondary, which in turn is more stable than primary

$$\begin{bmatrix} R - C \end{bmatrix}^{\frac{1}{2}} \longrightarrow \dot{R} + C - C$$

The stability of the cations is of the order:

$$CH_3^+ < RCH_2^+ < R_2CH^+ < R_3C^+$$

In general, the largest substituent at a branch is more readily eliminated as a radical. This is because a long-chain radical can achieve some stability due to delocalization of the lone electron.

- (*iv*) Compounds containing double bonds, cyclic structures, particularily aromatic (or heteroaromatic) rings stabilize the molecular ion. Due to this the molecular ion peak appear easily.
- (v) Allylic cleavage is favoured by the presence of double bonds giving resonance stabilized allylic carbonium ion.
- (vi) Compounds containing saturated rings, tend to lose side chain at the  $\alpha$ -bond; the positive charge staying with the ring fragment. However, unsaturated rings may undergo retro-Diels-Alder reaction.

$$\left[ \begin{array}{c} \begin{array}{c} \\ \\ \\ \end{array} \right]^{\frac{1}{2}} \rightarrow \left[ \begin{array}{c} \\ \\ \end{array} \right]^{\frac{1}{2}} \quad + \quad \begin{array}{c} \begin{array}{c} \\ \\ \\ \end{array} \end{array} \right]$$

(vii) In case of alkyl-substituted aromatic compounds, cleavage occurs at the bond beta to the ring giving resonance stabilized benzyl ion or tropylium ion (more likely formed).

$$\begin{bmatrix} CH_2 & R & -R' & CH_2 & CH$$

(viii) In case of compounds containing hetero atom (O, N or S), the C—C bond next to the heteroatom is cleaved leaving the charge on the fragment containing the hetero atom.

$$CH_{3} \xrightarrow{C} CH_{2} \xrightarrow{Y} \xrightarrow{Y} - R \xrightarrow{-CH_{3}^{*}} CH_{2} \xrightarrow{Y} - R \xrightarrow{+} CH_{2} \xrightarrow{Y} - R$$

$$Y = O, N \text{ or } S$$

$$R \xrightarrow{-C} - CH_{2} - R' \xrightarrow{-R^{*}} C - CH_{2}R' \xrightarrow{+} C - CH_{2} - R'$$

$$O: \qquad CH_{2} \xrightarrow{C} - CH_{2} - R'$$

(ix) In most of the cases, the cleavage is associated with the elimination of stable neutral molecules like carbon monoxide, olefins, water, ammonia, hydrogen sulfide, hydrogen cyanide, mercaptans, ketene or alcohols.

## 27.7.3 Rearrangements

In mass spectroscopy, we come across certain rearrangements, which involve migration of hydrogen atoms in a molecule containing a heteroatom. As has already been stated (P 388) a typical example is the so called **McLafferty rearrangement**. For a compound to undergo McLafferty rearrangement, it must have an appropriately located heteroatom (*e.g.* O), a  $\pi$  system (like a double bond) and an hydrogen which can be easily abstractable (*e.g.*, hydrogen  $\gamma$  to the C = O system)

The rearrangements of the types mentioned above often give prominent characteristic peaks. The rearrangement peaks can be recognised by the m/e value of the fragment ion and the corresponding molecular ion peak.

#### **Derivatives**

As already stated, a compound for mass spectrometric analysis must have reasonable volatility. However, if a compound has low volatility or if the parent mass cannot be determined, it is advisable to prepare a suitable derivative, which should have enhanced volatility, a predictable mode of cleavage and an increased stability of the parent ion.

Compounds like sugars, peptides and dibasic carboxylic acids have polar groups and have very low volatility. The volatility can be increased by acetylation of hydroxyl and amino groups and methylation of the free acids. Such derivatives give characteristic peaks. Trimethyl silyl derivatives of sugars and amino acids can also be used. In these case M-15 peak due to the cleavage of one of the  $S-CH_2$  bond is always prominent.

Ketones can be reduced to hydrocarbons to increase the volatility.

# 27.8 MASS SPECTRA OF SOME TYPICAL CLASSES OF COMPOUNDS

Mass spectra of some typical class of compounds is discussed in this section. A table of uncharged fragments that are normally eliminated is given in Table 27.6.

## 27.8.1 Saturated Hydrocarbons

Most of the early work in mas spectrometry has been on hydrocarbons related to petroleum industry. In general rules 1 to 3 (P. 389) predict the fragmentations. Though rearrangements peaks also occurs but these are random rearrangements.

In straight chain, saturated hydrocarbons, the molecular ion peak are of low intensity for long chain compounds. A cluster of peaks and the corresponding peaks of each cluster and 14 ( $\mathrm{CH}_2$ ) mass units apart. In the cluster, the largest peak correspond to  $\mathrm{C}_n\mathrm{H}_{2n+1}$  fragment accompanied by  $\mathrm{C}_n\mathrm{H}_{2n}$  and  $\mathrm{C}_n\mathrm{H}_{2n-1}$  fragments. Identification depends on the molecular ion peaks. The mass spectra of n-hexadecane is shown in Fig. 27.7.

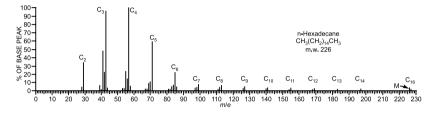


Fig. 27.7 Mass spectra of n-hexadecane

The mass spectra of branced saturated hydrocarbons are similar to that of straight chain compound. The mass specra of 5-methyl pentadecane is shown in Fig. 27.8. The peaks at m/e 169 and 8.5 shows cleavage on either side of the branch with charge retention on the substituted carbon atom. Cleavage at a branch loses a single hydrogen atoms to give  $C_nH_{2n}$  peak, which is prominent and is more intense than the corresponding  $C_nH_{2n+1}$  peak.

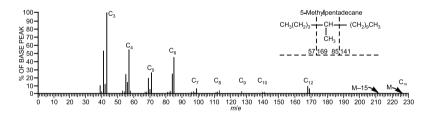


Fig. 27.8 Mass spectra of 5-methyl pentadecane

A saturated ring in a hydrocarbon as in the case of cyclohexane increases the intensity of the molecular ion peak favouring cleavage at the bond connecting the ring to the rest of the molecule. This is in accordance to rule vi on page 390. In this case, fragmentation of the ring is generally characterised by the loss of two carbon as  $C_2H_4$  (28) and  $C_2H_5$  (29). The mass spectra of cyclohexane is shown in Fig. 27.9. As seen, the base peak at m/e 56 (due to loss of  $C_2H_4$ ) and a larger peak at m/e 41.

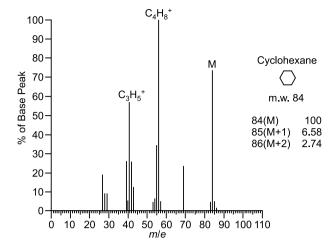


Fig. 27.9 Mass spectra of cyclohexane

#### 27.8.2 Unsaturated Hydrocarbons

The location of the double bonds is a cyclic olefins present defficulties due to facile migration of the fragments. However, in polycyclic olefins, the location of the double bond is possible due to tendency for allylic cleavage without the migration of the double bond. This is in accordance with rule 5 (P. 390). The position of the double bond also fixes by conjugation with a carbonyl group. The acyclic olefins (as in the case of saturated hydrocarbons) are characterised by clusters of peaks at interval of 14 units. However, in these clusters, the  $C_nH_{2n-1}$  and  $C_nH_{2n}$  peaks are more intense than the  $C_nH_{2n+1}$  peaks. The mass spectra of  $\beta$ -myrcene is given in Fig. 27.10. The peaks at m/e 69, 67 and 93 are believed to be obtained as shown below:

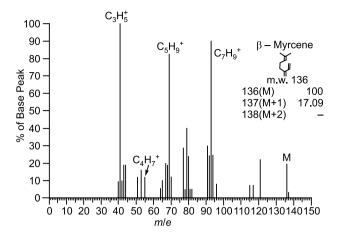
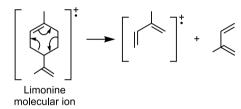


Fig. 27.10 Mass spectra of β-Myrcene

A distinct molecular ion peak is observed in case of **cyclic olefins**. A cleavage of the type of homolytic retro-Diels-Alder reaction is shown for limonine



In the mass spectrum of naphthalene (Fig. 27.11), the molecular ion peak is also the base peak. In this case, the largest fragment peak m/e 51 is about 12.5% as intense as the molecular ion peak.

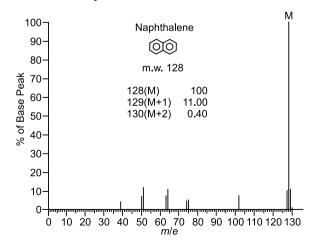


Fig. 27.11 Mass spectra of Naphthalene

In case of alkyl substituted benzene, like toluene a prominent peak at m/e 91 is also the base peak. Brancing at the  $\alpha$ -carbon gives masses higher than 91 by increments of 14; in this case the largest substituent is readily eliminate (Rule 3, P. 389). It should be noted that the presence of a peak at mass 91 does not preclude brancing at  $\alpha$ -carbon since this fragment may also result from rearrangements. A distinctive and prominant M-1 peak results from benzylic cleavage of a C—H bond.

It has been shown that in most cases, the ion of mass 91 is a tropylium rather than a benzylic cation. This accounts for ready loss of a methyl group from xylenes (although toluene does not lose a  $CH_3$  group). The molecular ion from xylenes rearranges to tropylium radical ion, which subsequently cleaves to tropylium ion  $(C_7H_7^+)$ . At times, the tropylium ion  $(C_7H_7^+)$  eliminates a molecule of acetylene to give a peak at m/e 65 as shown below:

In case in alkylbenzene, the alkyl group is longer than  $C_2$ , there is hydrogen miration with elimination of a neutral olefin giving peak at m/e 92

H 
$$CH_2$$
  $CH_2$   $CH_2$ 

#### 27.8.3 Alcohols

In case of alcohols, cleavage of the C – C bond next to oxygen atom takes place (Rule 8, P. 390). Thus, primary alcohols show a peak at m/e 31 due to  $CH_2 = \ddot{Q}H$  and secondary alcohols shows a peak at m/e 45, 59, 73 etc.) due to  $\ddot{R} = \ddot{Q}H$  and tertiary alcohols show a peak at m/e 59, 73, 87 etc. due to  $\ddot{R} = \ddot{Q}H$ . According to Rule 3, the largest substituent is readily explelled.

In case of alcohols, when R and or R' = H, an M - 1 peak is seen.

In addition to C—C cleavage next to oxygen, the primary alcohols show a homologous series at peaks resulting from the cleavage of C—C bonds successively removed from the O atom. In alcohols > C<sub>6</sub>, the pattern of fractionation is similar to that obtained in the corresponding olefin.

A distinctive feature in the mass spectra of alcohol is the M-18 peak due to loss of water. This is clearly noticeable in the spectra of primary alcohol.

The loss of water in the above case occurs between OH and  $\gamma$  -hydrogen (n = 1) or  $\delta$  hydrogen (n = 2). Elimination of water along with elimination of an olefin from primary alochols gives a peak at M – (olefin + H<sub>2</sub>O), *i.e.*, a peak at M – 46, M – 74, M – 102...

$$\begin{array}{c} H \\ O \\ CH_2 \\ R - CH \\ CH_2 \\ CH_2 \end{array} \xrightarrow{-CH = CH_2} \begin{bmatrix} CH_2 = CHR \end{bmatrix}^{\frac{1}{4}}$$

$$\begin{array}{c} H \\ O \\ CH_2 \\ CH_2 \\ \end{array} \xrightarrow{-H_2O} \begin{bmatrix} CH_2 = CHR \end{bmatrix}^{\frac{1}{4}}$$

The  $[M - olefin - H_2O]^{+}$  ion decomposes eliminating ethylene.

In case the alcohol has a branched  $CH_3$  group (as in the case of terpene alcohols), strong peak results at M-33 resulting from the loss of  $CH_3$  and  $H_2O$ .

Figs. 27.12-27.14 gives the mass spectra of the isomeric pentanols ( $C_5H_{11}OH$ ) *viz.*, 1-pentanol, 2-pentanol and 2-methyl-2-butanol.

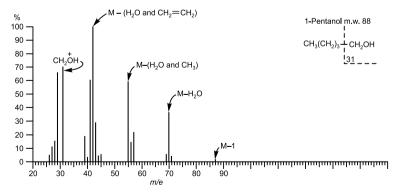


Fig. 27.12 Mass spectra of 1-pentanol

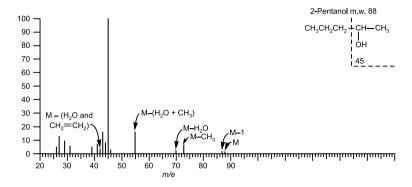


Fig. 27.13 Mass spectra of 2-pentanol

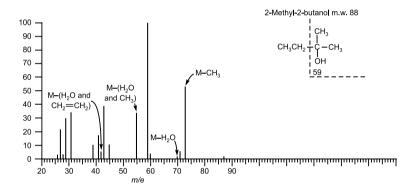


Fig. 27.14 Mass spectra of 2-methyl-2-butanol

A distinct type of primary alcohols are benzyl alcohol and their homologues. In these cases, the parent peak is strong. Beta cleavage to the ring gives the benzylic peak  $(m-\mathrm{OH})$  other peaks include  $\mathrm{M}-1$ ,  $\mathrm{M}-2$  and  $\mathrm{M}-3$ . Benzyl alcohol fragments to give  $\mathrm{M}-1$  ion,  $\mathrm{C}_6\mathrm{H}_7$  ion by the loss of CO and  $\mathrm{C}_6\mathrm{H}_5$  ion by the loss of  $\mathrm{H}_2$ .

 $\rm M-18$  peak by the loss of  $\rm H_2O$  is common in orthosubstituted benzyl alcohols.

Along with the M-18 peaks, the aromatic cluster (m/e 77, 78 and 79) is also prominent. This results from the complex degradation.

#### 27.8.4 PhenoIs

Phenols are identified by a conspicuous molecular ion peak. In case of simple phenol, the molecular ion peak is the base peak and the M-1 peak is small.

However, in cresols, M-1 peak is larger than the parent peak due the facile benzylic C—H cleavage. This is accompanied by a rearrangement peak at m/e 77 and also peaks resulting from loss of CO (M-28) and CHO (M-29).

The mass spectrum of o-ethylphenol, a typical alcohol is shown in Fig. 27.15. In the spectrum there is loss of CH<sub>3</sub> group and an  $\alpha$ -hydrogen.

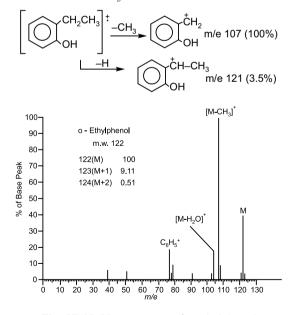


Fig. 27.15 Mass spectra of o-ethylphenol

#### 27.8.5 Ethers

In case of **aliphatic ether** (taking the case of ethyl sec-butyl ether,  $CH_3$ — $CH_2$ — $CH_2$ — $CH_3$ ), the molecular ion or the M+1 peak is obvious, The  $CH_3$ 

presence of O is deduced from peaks at m/e 31, 45, 59, 73 (see Fig. 27.16). The fragmentation occurs in two ways.

(i) Cleavage of the C—C bond next to O atom (α, β bond, Rule 8, Page 390)

$$\begin{array}{c} \text{CH}_{3}-\text{CH}_{2} & \overset{\dagger}{\overset{\bullet}{\text{CH}}} - \overset{\dagger}{\overset{\bullet}{\text{C}}} - \text{CH}_{2}-\text{CH}_{3} \\ \text{CH}_{3} & & \text{CH}_{2}-\text{CH}_{3} \\ \text{CH}_{3} & & \text{CH}_{2}-\text{CH}_{3} \\ \text{Sec-butyl ether} & \text{(A)} & \textit{m/e} \ 73 \\ \text{molecular ion} & \text{or} & \\ \text{CH}_{3}-\text{CH}_{2}-\text{CH}-\overset{\dagger}{\overset{\bullet}{\text{C}}} - \overset{\bullet}{\text{CH}_{2}} - \text{CH}_{3} \\ \text{CH}_{3} & & \text{CH}_{3}-\text{CH}_{2}-\text{CH}_{2} \\ \text{CH}_{3} & & \text{CH}_{3}-\text{CH}_{2} - \text{CH}_{2} \\ \text{CH}_{3} & & \text{CH}_{3}-\text{CH}_{2}-\text{CH}_{2} \\ \text{CH}_{3} & & \text{CH}_{3}-\text{CH}_{2}-\text{CH}_{2}-\text{CH}_{2} \\ \text{CH}_{3} & & \text{CH}_{3}-\text{CH}_{2}-\text{CH}_{2}-\text{CH}_{2} \\ \text{CH}_{3} & & \text{CH}_{3}-\text{CH}_{2}-\text{CH}_{2}-\text{CH}_{2}-\text{CH}_{2}-\text{CH}_{2} \\ \text{CH}_{3} & & \text{CH}_{3}-\text{CH}_{2}-\text{CH$$

(ii) Cleavage of C—O bond with the charge remaining on the alkyl fragment

 $R \xrightarrow{\bigcap_{i=1}^{t} -R'} R' \xrightarrow{-ii} R'$ 

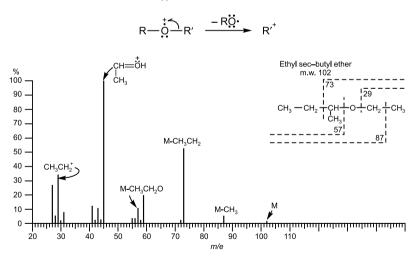


Fig. 27.16 Mass spectra of sec-butyl ether

In case of **aromatic ethers**, the molecular ion peak is prominent. Initially cleavage at the bond beta to the ring occurs followed by subsequent decomposition. Thus anisole (mole. wt. 108) gives ions at *m/e* 93 and 65.

$$m/e 108$$
 $CH_3 - CH_3$ 
 $m/e 93$ 
 $m/e 93$ 
 $m/e 65$ 

In case of anisole, aromatic peaks at m/e 78 and 77 result as shown below:

If the alkyl portion of the aromatic alkyl ether contains more than two carbons, β-cleavage to the ring occurs followed by hydrogen abstraction. In this case C—C cleavage next to O is negligible.

In case of diphenyl ethers, there is complex rearrangement giving peaks at M - H, M - CO and M - CHO

#### **27.8.6 Ketones**

**Aliphatic ketones** usually give a prominent molecular ion peak. Other fragments occur by cleavage of C - C bonds adjacent to O atom as in the case of alcohols and ethers giving peaks at m/e 43 or 57 or 71.

In case one of the alkyl group in Ketone is  $C_3$  or larger, the cleavage of the C-C bond once removed ( $\alpha\beta$  bond) from the C—O group occurs; the major peak is obtained by migration of H (McLafferty rearrangement).

In case R is C<sub>3</sub> or longer, the formed ion cleaves with migration of hydrogen

$$\begin{array}{c|c} H \overset{\overset{\leftarrow}{\circ}}{\overset{\leftarrow}{\circ}} - \overset{CH_2}{\overset{\leftarrow}{\circ}} + \overset{CH_2}{\overset{\leftarrow}{\circ}} - \overset{CH_2}{\overset{\leftarrow}{\circ}} + \overset{\overset{\leftarrow}{\circ}}{\overset{\leftarrow}{\circ}} - \overset{\overset{\leftarrow}{\circ}}{\overset{\leftarrow}{\circ}} \overset{\overset{\leftarrow}{\circ}}{\overset{\overset{\leftarrow}{\circ}}{\overset{\leftarrow}{\circ}} - \overset{\overset{\leftarrow}{\circ}}{\overset{\leftarrow}{\circ}} - \overset{\overset{\leftarrow}{\circ}}{\overset{\overset{\leftarrow}{\circ}}{\overset{\leftarrow}{\circ}} - \overset{\overset{\leftarrow}{\circ}}{\overset{\overset{\leftarrow}{\circ}}{\overset{\leftarrow}{\circ}} - \overset{\overset{\leftarrow}{\circ}}{\overset{\overset{\leftarrow}{\circ}}{\overset{\leftarrow}{\circ}} - \overset{\overset{\leftarrow}{\circ}}{\overset{\overset{\leftarrow}{\circ}}{\overset{\overset{\leftarrow}{\circ}}{\overset{\overset{\leftarrow}{\circ}}{\overset{\leftarrow}{\circ}} - \overset{\overset{\leftarrow}{\circ}}{\overset{\overset{\leftarrow}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\leftarrow}{\circ}}{\overset{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\overset{\overset{\smile}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}{\overset{\overset{\smile}{\circ}}{\overset{\overset{\smile}$$

In long chain ketones, the peaks due to hydrocarbon are indistinguishable (with using high-resolution procedures) from the acyl peak as the mass of CO unit and two methylene units is the same (CH $_2$ ). The identification becomes convenient, if the carbonyl group is reduced to CH $_2$  to give hydrocarbon whose fragmentation gives the carbon skeleton.

In **cyclic Ketones**, the molecular ion peak is prominent. As in the case of aliphatic ketones, the cleavage of cyclic ketones also takes place at adjacent to C = O group; the ion so formed undergoes further fragmentation.

In case of both cyclopentanone and cyclohexaneone is same is m/e 55 as shown below:

$$\begin{array}{c} \vdots \overset{\circ}{\bigcirc} \qquad \vdots \overset{\circ}{\bigcirc} \qquad$$

In case of cyclohexanone other peaks at m/e 83 and 42 are believed to be formed as shown ahead:

$$\begin{array}{c} \vdots \mathring{\mathsf{o}} \\ \mathsf{C} \\ \mathsf{H}_{2} \mathring{\mathsf{C}} \\ \mathsf{C} \\ \mathsf{C} \\ \mathsf{H}_{2} \\ \mathsf{C} \\ \mathsf{C} \\ \mathsf{C} \\ \mathsf{H}_{2} \\ \mathsf{C} \\ \mathsf{C} \\ \mathsf{H}_{2} \\ \mathsf{C} \\ \mathsf{C}$$

In **Aromatic Ketones**, a prominent molecular ion peak is obtained. In this case, cleavage occurs at the bond beta to the ring giving a characteristic fragment  $ArC \equiv O^+$  (which accounts for the base peak). Thus, in case of acetophenone, the loss of CO from this fragment gives aryl ion (m/e 77). The fragment  $RC \equiv Q^+$  is less important, but is enhanced by the presence of electron-withdrawing group and is diminished by the presence of electron donating groups in the para position of the Ar group.

In case of aromatic ketones, when the alkyl chain is  $C_3$  or longer, McLafferty rearrangement occurs (as in case it aliphatic ketone) via. the elimination of an olefin and formation of a stable ion.

$$Ar - C C CHR^{2}$$

$$Ar - C CHR^{2}$$

The mass spectrum of p-chlorobenzophenone, an unsymmetrical ketone is shown in Fig 27.17. As seen, a prominent molecular ion peak (m/e 216) is obtained. The M + 2 peak is less intense (intensity 33.99% reference to molecular ion peak). This shows the presence of chlorine in the structure (see mass spectra of halogen compounds section 27.8.17). The major peaks in the mass spectra of p-chlorobenzophenone (Fig. 27.17) are believed to be obtained as shown below:

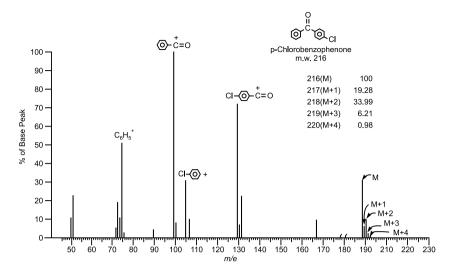


Fig. 27.17 Mass spectra of p-chlorobenzophenone

#### 27.8.7 Aldehydes

The molecule ion peak in case of aliphatic aldehydes is not easily recognised. However, cleavage of C—H bond next to the oxygen atom gives M-1 peak, which is a good diagnostic peak even for long chain aldehydes, In addition to M-1 peak, an M-R peak (m/e 29, CHO<sup>+</sup>) arises due to C—C bond cleavage next to oxygen (R-C). The m/e 29 peak in case of higher aldehydes is due to the ion  $C_2H_5^+$ .

In C<sub>4</sub> and higher aldehydes, McLafferty cleavage of the  $\alpha\beta$  C—C bond gives peaks at m/e 44, 58 or 72.... depending on the  $\alpha$ -substituent.

Other diagnostic peaks in case of straight chain aldehydes are M-18 (loss of  $H_2O$ ), M-18 (loss of  $CH_2=CH_2$ ), M-43 (loss of  $CH_2-CH=O$ ·) and M-44 (loss of  $CH_2=CH$  OH). With increase in chain length, the hydocarbon pattern (m/e 29, 43, 57, 71....) appear (see mass spectra of nonanal, Fig. 27.18).

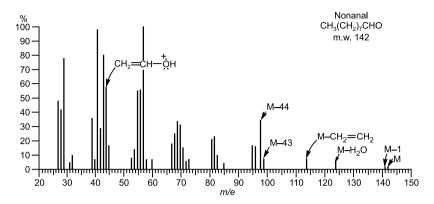


Fig. 27.18 Mass spectra of nonanal

### 27.8.8 Carboxylic Acids

The molecular ion peak of a straight chain monocarboxylic acid is small. A prominent and characteristic peak is at m/e 60, which is due to McLafferty rearrangement. Brancing at  $\alpha$ -carbon enhances the cleavage.

In carboxylic acids having short-chain prominent peaks are due to cleavage of bonds next to C = O giving M - OH and M - COOH peaks. However, in long chain carboxylic acids the mass spectrum consists of two series it peaks due to cleavage of each C—C bond with retention of charge either on alkyl fragment (m/e 29, 43, 57, 71, 85, .......) or on the oxygen containing fragment (m/e 45, 59,

73, 89.....). As discussed earlier, hydrocarbon pattern shows peaks at m/e 29, 28; 41, 42; 55, 56; 69, 70....). As an example, in the mass spectra of hexanoic acid (MW 116), the cleavage is as shown on previous page.

The dibasic acid, being non-volatile are converted into volatile derivatives like esters. The mass spectra of esters is discussed subsequently.

### **Aromatic Carboxylic Acids**

The molecular ion peak, unlike in aliphatic carboxylic acids is large. Some other prominent peaks arise by loss of OH (M-17) and of COOH (M-45). The typical M-18 peak  $(M-H_2O)$  occurs if a hydrogen bearing ortho group in present particularly in a 6-membered transition state to facilitate loss of molecules like  $H_2O$ , ROH or  $NH_3$ . This is an example of 'ortho effect'.

$$Z = OH, OR, NH_2$$

$$y = CH_2, O, M_7$$

# 27.8.9 Carboxylic Esters

Methyl esters of straight chain aliphatic acids give a weak molecular ion peak. A characteristic peak is due to McLafferty rearrangement and cleavage of one bond next to C = O group.

In short chain esters (up to  $C_4$ , the ion  $R^+$  is prominent. A diagnostic peak is given by the ion  $R - C \equiv O^+$ . In methyl esters, it is at M - 31. The other

ion, [OR']<sup>+</sup> and [C—OR']<sup>+</sup> are of no importance. The mass spectrrum of methyl octanoate is given in Fig. 27.19.

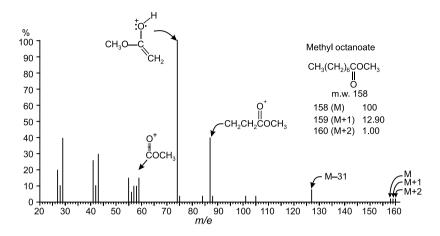


Fig. 27.19 Mass spectra of methyl octanoate

In some esters (*e.g.*, esters of fatty alcohols) (except methyl ester), the alcohol portion is predominant. Such esters eliminate a molecule of acid in a manner analogous to the elimination of water from alcohols. A plausible mechanism involves a hydride transfer to the carbonyl oxygen (McLafferty rearrangement).

Long chain esters give a diagnostic peak at m/e 61, 75, or 89 .... by the elimination of alkyl moiety and transfer of two hydrogens to the oxygen containing fragment

$$\begin{array}{c} H \\ \text{CH-CH}_2R' \\ \text{R-C} \\ \text{CH} \end{array} \xrightarrow{\text{CH-CHR'}} \begin{array}{c} H \\ \text{CH-CHR'} \\ \text{R-C} \\ \text{CH}_2H \end{array} \xrightarrow{\text{-R'CHCH=CH}_2} \begin{array}{c} H \\ \text{R-C} \\ \text{CH}_2H \end{array}$$

Benzyl and phenyl esters (like benzyl acetate and phenyl acetate) eliminate a molecule of ketene.

In case of benzyl acetate, the prominent peaks are at m/e 43 (CH<sub>3</sub>C  $\equiv$  O<sup>+</sup>) and m/e 91 (C<sub>7</sub>H<sub>7</sub><sup>+</sup>).

In methyl esters of aromatic carboxylic acids, the molecular ion peak is prominent, which decreases as the alcohol moiety increases (for  $C_5$  carboxylic ester m/e is zero). The base peaks results from elimination of  $\cdot$ OR (M – 31 for methyl ester) and elimination of  $\dot{\text{COOR}}$  (M – 59 for methyl ester); both these are prominant peaks. As the alkyl moiety increases in length, following three modes of cleavage are possible.

### (i) McLafferty rearrangement

(ii) Rearrangement of two hydrogens with elimination of an allylic radical

(iii) Retention of positive charge on alkyl group

$$Ar - C - O + R$$

$$-Ar - C - O + R$$

$$R^{\dagger}$$

Ortho-substituted benzoates eliminate alcohol (ROH) (ortho effect as described under aromatic acids). As as example, methyl salicylate gives a base peak at m/e 120, which eliminates carbon monoxide to give a strong peak at m/e 92.

#### 27.8.10 Lactones

Five membered ring lactones give a prominent molecular ion peak. However, if there is substitutent at  $C_4$ , the peak becomes weaker. Cleavage of the side chain at  $C_4$  (Rule 3 and 8, page 389 and 390) gives a strong M-alkyl.

 $\gamma$ -valerolactone gives a base peak at m/e 56. Same peak is obtained in case of butyrolactone.

#### 27.8.11 Amines

In aliphatic monoamines, the molecular ion peak is weak and is undetectable in logn chain or highly branced amines The C – C cleavage next ( $\alpha\beta$ ) to the nitrogen atom (Rule 8, Page 390) gives the base peak, In case of primary amines, unbranced at  $\alpha$ -carbon, the base peak is m/e 30 (CH<sub>2</sub>NH<sub>2</sub><sup>+</sup>). This cleavage is responsible for base peak in all primary amines and also for secondary and tertiary amines which are not branced at  $\alpha$ -carbon. The largest branch is lost from the  $\alpha$  – C atom.

The M-1 peak is usually visible when R and / or  $R^1 = H$ .

**Straight chain primary amines** show a homologous series of peaks of progressively decreasing intensity at m/e 30, 44, 58...... due to cleavage of C—C bonds successivley removed from the N atom with the retention of charge on the fragment carrying N. Other peaks follow the hydrocarbon pattern of  $C_nH_{2n+1}$ ,  $C_nH_{2n}$  and  $C_nH_{2n-1}$  ions. During the fragmentation of longer chain amines, cyclic fragments also occur.

$$R \leftarrow CH_2 \qquad NH_2 \qquad R \leftarrow CH_2 \qquad NH_2 \qquad NH_2 \qquad R \leftarrow CH_2 \qquad NH_2 \qquad NH$$

Though a peak at m/e 30 is considered to be good, but it is not conclusive evidence for straigh chain primary amines. The first formed ion from secondary or tertiary amine on decomposition give peak at m/e 30, 44, 58, 72 ..... This is similar to that for aliphatic alcohols and ethers.

R" = H, m/e 30, less intense R" = CH<sub>3</sub>, m/e 40, more intense

In case of amino acid esters, cleavage occurs at both C—C bonds (a, b shown) below) next to N, loss of carbalkoxy group is preferred (a). The fragment from aliphatic amine decomposes to give a peak at m/e 30.

**Cyclic amines** give a more intense molecular ion peak compared to acyclic amines. As an example, pyrrolidine gives a strong molecular ion peak. In fact, cleavage at both bonds next to N atom leads to either loss of  $\alpha$  – H to give a strong M – 1 peak or leads to opening of the ring; in the lather case there is elimination of ethylene to give  ${}^{\bullet}\text{CH}_2\text{N}^+\text{H} = \text{CH}_2$  (m/e 43, base peak) followed by loss of a H atom to give  ${}^{\bullet}\text{CH}_2 = \overset{+}{\text{N}} = \text{CH}_2$  (m/e 42). N-Methyl pyrrolidine also gives a  ${}^{C}_2\text{H}_4\text{N}^+$  (m/e 42) peak.

Strong molecular ion peak is also obtained from piperidine. This is followed by ring opening giving characteristic peaks at m/e 70, 57, 56, 44, 43, 42, 30, 29 and 28. The substituents are cleaved from the ring (Rule 6, P. 390)

The molecular ion peak (odd number) of an **aromatic amine** is intense. The M-1 peak is obtained by the loss of one of the amino hydrogen atom; loss of HCN followed by the loss of a H atom gives peaks at m/e 66 and m/e 65 respectively.

In the case of alkylaryl amines, the cleavage of C—C bond next to N atom occurs readily; in this case the hetero atom controls the cleavage (compare with alkyl aryl ethers, which involves rearrangement involving cleavage of ArO-R bond—the cleavage being controlled by the ring rather than *O* atom)

#### 27.8.12 Amides

In straight-chain monoamides, the molecular ion peak is not clear. The mode of cleavage depends on the length of acyl moiety and also on the lengths and number of alkyl groups attached to N atom.

In all straight chain primary amides, the base peak results from McLafferty rearrangement:

Homologous peak at m/e 73 or m/e 87 are obtained in case there is brancing at  $\alpha$ -carbon (CH<sub>3</sub> etc.)

Primary amides give a strong peak at m/e 44 resulting from the cleavage of R—CONH<sub>2</sub> bond:  $(\dot{\Omega} = C - \dot{N}H_2 \leftrightarrow \ddot{\Omega} = C = \dot{N}H_2)$ ; Such cleavage gives the base peak in  $C_1 - C_3$  primary amides and also in isobutrylamide. A peack at m/e 86 results by the  $\gamma\delta$  C—C cleavage and accompanied by cyclisation

Homologous peaks at m/e (40, 54, 68, 82 ....) are obtained from the ion  $(CH_2)_n C \equiv N^{++}$  by the simple cleavage of each C - C bond (except the one next to N atom). In addition, usual peaks of the hydrocarbon pattern.

# 27.8.13 Nitro Compounds

Aliphatic mononitro compounds mostly give a weak molecular ion peak (odd number). Nitro group is indicated by an peak at m/e 30 (NO<sup>+</sup>) along with a smaller peak at m/e 46 (NO<sub>2</sub><sup>+</sup>). However, aromatic nitro compounds give a strong molecular ion peak. A prominent peak is due to M – 46 (elimination of NO<sub>2</sub> radical) which is the base peak, along with a M – 30 peak (elimination of NO molecule with rearrangement). Both these peaks (M – 46 and M – 30) are diagnostic peaks. The M – 16 ion looses HC  $\equiv$  CH to give a strong peak at M – 72. And loss of CO from M – 30 ion peak gives M – 58 peak.

Nitroanilines (o, m-, and p-) give a strong molecular ion (even number), which give prominent peaks as shown below:

$$m/e$$
 138 (M)  $\xrightarrow{-NO_2}$   $m/e$  92  $\xrightarrow{-HCN}$   $m/e$  65  $\xrightarrow{-NO}$   $m/e$  108  $\xrightarrow{-CO}$   $m/e$  80

Ortho and para nitroanilines give a small peak at m/e 122 due to loss of O atom. On the other hand o-nitroaniline eliminates  $\Theta$ H to give a small peak at m/e 121.

#### 27.8.14 Nitrites

Aliphatic nitrites give a weak molecular ion peak (1N present). A large base peak is at m/e 30 (NO<sup>+</sup>). In addition, all nitrites unbranced at the  $\alpha$ -carbon give a large peak at m/e 60 (CH<sub>2</sub> =  $\stackrel{+}{\text{O}}$ NO); this is due to cleavage of the C—C bond next to the ONO group. A branch can however, be identified by a peak at m/e 74, 88 or 102.... Differentiation from nitro compounds can be made due of absence of a large peak at m/e 46. However, prominent hydrocarbon peaks are also obtained.

### 27.8.15 Nitrates

As in the case at aliphatic nitrites, the molecular ion peak (odd number) in aliphatic nitrates is weak or absent. A prominent peak (which is often the base peak) is obtained by the cleavage of C—C adjacent to  $ONO_2$  group; the heaviest alkyl group attached to  $\alpha$ -carbon is eliminated

A prominent peat at m/e 46 is due to  $NO_2^+$ . The hydrocarbon fragment ions are quite distinct as in the case of aliphatic nitrites.

# 27.8.16 Sulfur Containing Compounds

Sulphur containing compounds are generally recognised by the contribution (4.4%, see Table 27.1) of the  $^{34}$ S isotope to M + 2 peak, and also to a [fragment + 2] peak. From the size of the contribution of the  $^{34}$ S isotope to the M + 2 peak, the number of sulfur atoms can be determined. The mass of sulphur atom(s) present in the molecules is substracted from the molecular weight. The formula of the rest of the molecule is determined from the M + 1 peak after substracting the contribution of the  $^{33}$ S isotope. As an example, in di-isopentyl disulphide (mol.wt. 206) contains two sulfur atoms. The formula for the rest of the molecule is found under mass 142 (206 – 2 × 32); the corrected intensity of the M + 1 peak is 12.5 – (2 × 0.78) = 10.9.

Aliphatic mercaptans (except higher tertiary mercaptans) usually give a strong M + 2 peak. Cleavage of the C—C bond ( $\alpha\beta$  bond) next to SH group give a peak at m/e 47 due to the ion  $CH_2 = \stackrel{+}{S} \longleftrightarrow CH_2 - \stackrel{-}{S}H$ . Other peaks at m/e 61, m/e 75 and m/e 89 result from the cleavage of  $\beta\gamma$  bond,  $\gamma\delta$  bond and  $\delta\epsilon$  bond respectively.

As in the case of alcohols, primary mercaptans eliminate  $H_2S$  and give a M-34 peak, the resulting ion eliminate  $CH_2=CH_2$ .

$$\begin{array}{c} H \\ \vdots \\ S_{2}^{+} \\ CH_{2} \\ CH_{2} \\ CH_{2} \\ CH_{2} \end{array} \longrightarrow \begin{array}{c} -H_{2}S \\ -C_{2}H_{4} \\ \end{array} \qquad \begin{bmatrix} CH_{2} = CHR \end{bmatrix}^{+}$$

In case of aliphatic sulfides, the molecular ion peak is intense and so the M+2 peak can be measured. Cleavage of one of the two  $\alpha\beta$  C — C bond occurs in the elimination of the largest group as shown:

In sulfides, cleavage of C—S bond occurs; the resulting ion  $\stackrel{+}{R}$  gives peak at m/e 32 + CH<sub>3</sub>, 32 + C<sub>2</sub>H<sub>5</sub>, 32 + C<sub>3</sub>H<sub>7</sub> .... A ion m/e 103 is particularly formed due to formation of a rearranged cyclic ion

$$CH_3CH_2CH_2CH_2CH_2\overset{\overset{\bullet}{,}}{\overset{\bullet}{,}} \longrightarrow H_2\overset{\overset{\bullet}{,}}{\overset{\bullet}{,}} \stackrel{H_2}{\overset{\bullet}{,}} \stackrel{C}{\overset{\bullet}{,}} \stackrel{C}{\overset{\bullet}{,}} \stackrel{C}{\overset{\bullet}{,}} \stackrel{C}{\overset{\bullet}{,}} \stackrel{C}{\overset{\bullet}{,}} \stackrel{C}{\overset{\bullet}{,}} \stackrel{C}{\overset{\bullet}{,}} \stackrel{C}{$$

These features are illustrated in the spectrum of di-*n*-pentyl sulphide (Fig. 27.20).

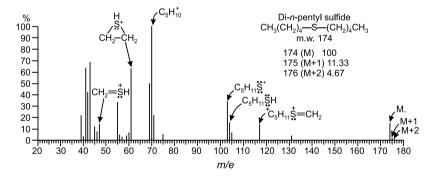


Fig. 27.20 Mass spectra of Di-n-pentyl sulfide

# 27.8.17 Compounds Containing Halogens

A compound containing one chlorine atom shows an M + 2 peak [intensity 33 percent of the molecular ion peak due to the presence of a molecular ion containing  $^{37}$ Cl isotope (Table 27.6)].

A compound containing one bromine atom shows an M+2 peak of intensity equal to that of molecular ion due to the presence of molecular ion containing <sup>81</sup>Br isotope. However, a compound containing two chlorines or two bromines, or one chlorine and one bromine shows a distinct M+4 peak in addition to M+2 peak, due to the presence of a molecular ion containing two atoms of heavy isotope. In fact, the number of chlorine and/or bromine atoms in a molecule can be ascertained by the number of alternate peaks beyond the molecular ion peak. Thus, a compound containing three chlorine atoms will give peaks at M+2, M+4 and M+6.

Combinations of Bromine and Chlorine							
Halogen Present	% M + 2	% M + 4	% M + 6	% M + 8	% M + 10	% M + 12	
Br	97.9						
Br <sub>2</sub>	195.0	95.5					
Br <sub>3</sub>	293.0	286.0	93.4				
CI	32.6						
Cl <sub>2</sub>	65.3	10.6					
Cl <sub>3</sub>	97.8	31.9	3.47				
CI <sub>4</sub>	131.0	63.9	14.0	1.15			
CI <sub>5</sub>	163.0	106.0	34.7	5.66	0.37		
CI <sub>6</sub>	196.0	161.0	69.4	17.0	2.23	0.11	
BrCl	130.0	31.9					
Br <sub>2</sub> Cl	228.0	159.0	31.2				
Cl <sub>2</sub> Br	163.0	74.4	10.4				

**Table 27.6** Intensities of Isotope Peaks (Relative to the Molecular Ion) for Combinations of Bromine and Chlorine

Data taken from Silverstein, R.M.; Webster F.X., Spectrometric Identification of organic compounds, Wiley, New York, 1998.

The **aliphatic monochlorides** (lower members) give a detectable molecular ion peak. Subsequent cleavage at the C — C bond adjacent to chlorine atom gives a small peak at m/e 49 and also a isotopic peak at m/e 51.

$$\mathsf{R} \overset{-}{-} \mathsf{C} \mathsf{H}_2 \overset{\cdot}{-} \mathsf{C} \mathsf{H}_2 \overset{\cdot}{-} \mathsf{C} \mathsf{H}_2 \overset{+}{-} \mathsf{C} \mathsf{H}_2$$

A small  $Cl^+$  peak and  $R^+$  peak is also obtained by the cleavage of C-Cl bond.

Chlorides (straight chain) longer than  $C_6$  give the ions  $C_3H_6Cl^+$ ,  $C_4H_8Cl^+$  and  $C_5H_{10}Cl$ ; the last ion forms the most intense peak due to the formation of a stable 5 membered cyclic structure.

The bromides behave in the same way as chlorides. The iodides give a strong molecular ion peak. As iodine is monoisotopic there is no isotope peak. Iodides cleave as the chlorides or bromides. However, the ion  $\mathrm{C_4H_8I^+}$  is not as evident as the corresponding bromide or chloride ions. Fluorides give the weakest molecular ion peak.

Benzyl halides give detectable molecular ion peak. In this case the formation of tropylium ion due to loss of halide (Rule 8, P. 390) is favoured in comparison to  $\beta$  bond cleavage.

The molecular ion peak of aromatic halides is apparent. The M — X peak is large, in compounds in which X is directly attached to the ring.

# 27.8.18 Heterocyclic Compounds

Heterocyclic compounds give intense molecular ion peak. Five membered heterocyclics (Furan, thiophene and pyrrole) show fragmentations as shown below:

$$y = O, S, NH$$

$$y = S, NH$$

$$y = S, NH$$

Following peaks are observed:

Furan: Two principal Peaks:  $C_3H_3^+$  (m/e 39) and  $HC \equiv \mathring{O}$  (m/e 29).

Thiophene: Three peaks; m/e 39 (C<sub>3</sub>H<sub>3</sub><sup>+</sup>), 45 (HC= $\stackrel{\stackrel{+}{\circ}}{=}$ ) and 58 (C<sub>2</sub>H<sub>2</sub> $\stackrel{\stackrel{+}{\circ}}{=}$ )

Pyrrole: Three peaks: m/e 39 (C<sub>3</sub>H<sub>3</sub><sup>+</sup>), 28 (HC $\equiv$ NH) and 41 (C<sub>2</sub>H<sub>2</sub>NH)

In alkylpyridines, the cleavage of  $\beta$  C — C bond depends on the position of alkyl substitutent. Cleavage is more pronounced in case of 3-alkyl substituent. In case, the alkyl group is more than 3 carbons in position 2, the fragmentation takes place as follows:

Table 27.7 gives characteristic fragmentation pattern of various classes of compounds.

Table 27.7 Common losses of fragments from molecular ions

Table 21.1 Common losses of fragments from molecular lons				
SI. No.	Mass of Fragment lost	Formula of the Fragment		
1.	1	Ĥ		
2.	15	CH <sub>3</sub>		
3.	16	Ö, NH <sub>2</sub>		
4.	17	ÖH,NН <sub>3</sub>		
5.	18	H <sub>2</sub> O		
6.	19	Ė		
7.	20	HF		
8.	26	$C_2H_2$ , $CN$		
9.	27	HCN, $C_2\dot{H}_3$		
10.	28	CO, C <sub>2</sub> H <sub>4</sub> , (HCN + H)		
11.	29	CHÖ, $C_2\dot{H}_5$		
12.	30	$\dot{\text{CH}}_2\text{NH}_2, \dot{\text{CH}}_3\dot{\text{N}}\text{H}, \dot{\text{CH}}_2\text{O}, \dot{\text{NO}}$		
13.	31	ÖCH <sub>3</sub> , ĊH <sub>2</sub> OH, CH <sub>3</sub> NH <sub>2</sub>		
14.	32	CH₃OH, S		
15.	33	$H\dot{S}$ , ( $CH_3 + H_2O$ ), $\dot{C}H_2F$		
16.	34	$ m H_2S$		
17.	35	Cl		
18.	36	HCI, 2H <sub>2</sub> O		
19.	38	$C_3H_2,F_2$		
20.	39	C <sub>3</sub> H <sub>3</sub> , H <sub>2</sub> CCN		
21.	40	$\mathrm{CH_2CN}$ , $\mathrm{CH_3C}$ $\equiv$ $\mathrm{CH}$		
22.	41	$CH_2 = CH\dot{C}H_2$		
23.	42	$CH_2 = C = O, C_3H_6$		
24.	43	$CH_3\dot{C}O, C_3H_7, CH_2 = CH - \dot{O}$		

SI. No.	Mass of Fragment lost	Formula of the Fragment
25.	44	$CO_2$ , $C_2H_5NH$ , $CH_2 = CH OH$ , $N_2O CONH_2$ , $C_3H_8$
26.	45	$C_2H_5\dot{O}$ , COO $\dot{H}$ , CH $_3$ CHOH
27.	46	$C_2H_5OH, \overset{\scriptscriptstyle+}{N}O_2$
28.	47	ĊH <sub>2</sub> SH, CH <sub>3</sub> Ś
29.	48	CH₃SH, SO
30.	49	ĊH <sub>2</sub> CI
31.	52	$C_4H_4$ , $C_2N_2$
32.	57	$C_2H_5\dot{C}O,C_4\dot{H}_9$
33.	58	$C_4H_{10}$ , NCS, (NO + CO), $CH_3COCH_3$
34.	59	$CH_3O\dot{C} = O, CH_3CO NH_2,$ $\dot{C}H_2COOH$
35.	60	СН <sub>3</sub> СООН, С <sub>3</sub> Н <sub>7</sub> ОН
36.	61	CH <sub>3</sub> CH <sub>2</sub> S
37.	63	ĊH <sub>2</sub> CH <sub>2</sub> CI
38.	64	$S_2$ , $SO_2$ , $C_5H_4$
39.	65	$C_5H_5$
40.	69	ĊF <sub>3</sub> , C <sub>5</sub> Ĥ <sub>9</sub>
41.	71	$\dot{C}_5H_{11}, C_4H_7O$
42.	73	$CH_2CH_2 - O - C = O$
43.	75	$C_6H_3$
44.	76	$C_6H_4$ , $CS_4$
45.	77	$C_6H_5$ , $CS_2H$
46.	78	$C_6H_6$ , $C_5H_4N$
47.	79	$\dot{Br}, C_{6}H_{5}N, C_{6}\dot{H}_{7}$
48.	80	HBr
49.	91	C <sub>7</sub> H <sub>7</sub>
50.	105	C <sub>6</sub> H <sub>5</sub> ĊO,Ċ <sub>8</sub> H <sub>9</sub>
51.	119	CF <sub>3</sub> – CF <sub>2</sub>
52.	122	C <sub>6</sub> H <sub>5</sub> COOH
53. 54.	127 128	HI

#### 27.9 GAS CHROMATOGRAPHY-MASS SPECTROMETRY

It is a tchnique in which gas chromatography is coupled with mass spectrometry. The procedure consist in injecting a very small sample of a mixture of compounds (using a syringe) into a heated port of the gas chromagraph, where the sample is vapourized and swept by a flow of a inert gas into a capillary column (a coiled tube usually 10 - 30 meters long and 0.1 to 0.5 mm in diameter). The inside of the capillary column is coated with a non-polar 'stationary phase' which is normally a non-polar silicon-based polymer. The various components of the mixture travel at different rates (depending on their b.p's and degree of adsorption on the stationary phase) in the column. The first component to pass through the column is low boiling and non polar material. This is followed by materials with higher boiling points or stronger adsorption on the stationary phase. Each component takes different time (retention time) to pass through the column. Thus, each component of the mixture passes into the mass spectrometer, where the molecules are bombarded by electrons and mass spectra of each component is separately recorded. The ability of GC/MS to separate mixtures and give mass spectra of each component is a indispensable tool in analysis of a mixture of organic compounds.

# 27.9.1 Applications of Gas Chromatography-Mass Spectrometry

- (i) Structure elucidation of components in a mixture of compounds. Analysis of a complex mixture can be achieved by using a combined gas chromatograph-mass spectrometer (GC–MS).
- (ii) Components of Coffee aroma. Coffee aroma is a mixture of several hundred simple compounds. Each of these compounds can be identified by GC–MS.
- (iii) Drug metabolism. Administration of drugs produces a number of metabolites. Their structures can be conveniently elucidated by GC – MS.
- (iv) Pesticides and pollutants. GC Mass spectroscopy is routenly used for the analysis of pesticides residues and other pollutants. This procedure has been used for determining the presence and estimation of pesticides in human tissue and poly chlorinated pesticides in food.

#### 27.10 NEGATIVE ION MASS SPECTROMETRY

It has been stated that in Mass Spectrometry the result is recorded as a spectrum of positive ion fragments. The positive ion fragments are separated on the basis of mass. However, in negative ion mass spectrometry, negative ions are formed and the result recordeds as in case of usual mass spectrometry.

Negative ion mass spectrometry is not accepted as a useful method, as many organic compounds do not form stable negative ions. Also the negative ions are

tradiationally generated by high energy electrons (10 - 70 eV), the ion currents are small.

It has been found that negative ions can be formed in plenty from simple compounds containing elements with high electron affinity. Such compounds include SO<sub>2</sub>, PCl<sub>3</sub>, IClO<sub>3</sub> or organic compounds containing electron withdrawing groups like F, NO<sub>2</sub>, CN etc. The information obtained from negative ion mass spectrometry is complementary to the positive ion spectra.

# 27.10.1 Negative Ion Formation

A simple molecule on interaction with an electron produces negative ion (or molecular anions) by resonance capture, dissociative resonance capture or ion-pair formations (equations 1, 2 and 3 respectively)

$$AB + e \rightarrow AB^{\Theta}$$
 ...(i)

$$AB + e \rightarrow \dot{A} + B$$
 ...(ii)

$$AB + e^{\Theta} \rightarrow A^{\Theta} + B^{\Theta} + e^{\Theta}$$
 ...(iii)

All the above processes [equations (i) to (iii)] occur in the ionization chamber of a mass spectrometer. However, the relative abundance of ions is dependent on pressure. It has been found (Von Ardenne and Z. Angew, Phys 1959, 11, 121) that use of low energy electrons (2 to 4 eV) from an argon discharge tube increases the formation of negative ions. On the basis of the spectra of large number of compounds, it was shown that ions with masses near the molecular ion such as  $M^-$  (equation (i)), M-1 (loss of hydrogen) and M+16 and (M-1)+16 predominated. The last two species are formed by the reaction with oxygen present in the ion source.

# 27.10.2 Reactions Observed during Negative ion Chemical Ionization

Some of the important reactions observed during negative ion chemical ionization are given below:

- 1. Interation of electron/molecules give negative ions by the three processes [equations (i), (ii) and (iii)] as already explained.
- 2. The formed negative ions can be lost on contact with a neutral species

$$X^- + N \rightarrow XN + e^-$$

where N is a neutral species

3. The charge association reaction

$$A^- + B \rightarrow B^- + A$$

is exothermic if the affinity of B is greater than the electron affinity of A.

4. Clustering reactions or third order association are commonly observed for negative ion (as in the case of positive ions). Thus, the anions form solvated species in presence of polar molecules like water.

# 27.10.3 Fragment Patterns of Negative Ions

Following are given the fragment patterns of negative ions derived from common organic compounds.

## 27.10.3.1 Hydrocarbons

Being weakly acidic, simple alkanes are not easily deprotonated even by strongest available base,  $-\mathrm{NH}_2^-$ . In case of simple alkyl radicals, the electron affinities are negative and so the anions are unstable compared to radicals. However, methyl anion and cyclopropyl anions are exceptions (electron affinities of Mė and cyclopropyl radical,  $\dot{C}_3H_5$ , are + 18 and + 7.9 K cal mol<sup>-1</sup> respectively). It is well known that these ions are easily produced by the decarboxylation of RCO $_2^-$  ions.

Olefins and benzyl derivatives are easily deprotonated.

The negative ion mass spectra of aromatic compounds has been recorded using a low voltage ion source (mass spectrometer equipped with a Duoplas–Matron ion source). In this procedure the molecular ions and other related peaks are found to be in abudant species.

Aromatic hydrocarbons like benzene, naphthalene and other polyclic aromatic hydrocarbons give spectra resulting from electron capture and expulsion of hydrogen (Table 27.8).

Compound	Formula	Base peak (m/z)	Other peaks (m/z)
Benzene	C <sub>6</sub> H <sub>6</sub>	77 (M – 1)	75
Naphthalene	C <sub>10</sub> H <sub>8</sub>	127 (M – 1)	126, 125
Anthracene	C <sub>14</sub> H <sub>10</sub>	178 (M)	177
Phenanthrene	C <sub>14</sub> H <sub>10</sub>	177 (M – 1)	178, 176, 175

Table 27.8 Negative ion mass spectra of some aromatic hydrocarbons

All the above compounds give a peak (M + 15). These ions are formed as given below:

$$M + O + e^{-}$$
 $M + O^{-}$ 
 $M + O^{-}$ 
 $M + O^{-}$ 
 $M + e^{-}$ 
 $M + e^{-}$ 

The negative ion mass spectra of anthracene and tetracene is given in Figs. 18.21 and 18.22.

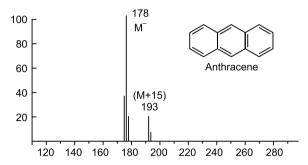


Fig. 27.21 Negative ion mass spectra of Anthracene

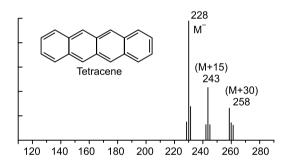


Fig. 27.22 Negative ion mass spectra of Tetracene

# 27.10.3.2 Alkyl Halides

Alkyl halides on reaction with a strong base in gas phase give either  $S_N^2$  or eliminations reactions. The reactions of ethyl fluoride with  $CD_3O^-$  is as shown.

$$CD_{3}O^{-} \xrightarrow{29\%} F^{-} + CD_{3}OH + C_{2}H_{4}$$

$$CD_{3}O^{-} \xrightarrow{29\%} [F^{-} \cdots HOCD_{3}] + C_{2}H_{4}$$

$$\downarrow CD_{3}O^{-} \xrightarrow{34\%} [C^{-}H_{2}CH_{2}F + CD_{3}OH]$$

$$\downarrow F^{-} + H_{2}C = CH_{2}$$

#### 27.10.3.3 Alcohols

The fragmentation of alkoxide ion takes place as shown below:

An interesting example of the fragmentation is given below:

$$\begin{array}{c} \text{Ph} \\ \text{Me} \end{array} \text{CH-O} \longrightarrow \begin{array}{c} (\text{Me}^-(\text{PhCHO})] \longrightarrow (\text{C}_6\text{H}_4)^-\text{CHO} + \text{CH}_4 \\ \\ [\text{Ph}^-(\text{MeCHO})] \longrightarrow (\text{C}_6\text{H}_5)^-\text{CHO} + \text{MeCHO} \\ \\ & \qquad \qquad (\text{CH}_2\text{CHO})^- + (\text{C}_6\text{H}_6) \end{array}$$

#### 27.10.3.4 PhenoIs

Virtually little work has been recorded for simple phenols. The phenoxide ion loses CO and CHO along with an ortho H. This is represented as given below:

$$C_6H_5O^ C_5H_5^- + CO$$
  $C_6H_4^- + CHO$ 

#### 27.10.3.5 Ethers

In presence of strong bases the simple dialkyl ether undergo elimination reaction in vapour phase; in this case deprotonated species are not formed

$${\rm HO^-} + {\rm C_2H_5OC_2H_5} \rightarrow {\rm C_2H_5O^-} + {\rm C_2H_4} + {\rm H_2O}$$

However, unsaturated ether, easily undergo deprotonation at either benzylic or allylic positions and the formed carbanion undergoes dissociations. An example is:

$$CH_2 = CH - \bar{C}H - (CH_2)_nOEt \rightarrow CH_2 = CH(CH_2)_{n+1}OCH_2CH_2^-$$
  
 $n = 1 - 7$  Eleb. intermediate

The reaction is more pronounced when n = 1. Thus,

The anion obtained from diallyl ether undergoes both 1, 2 and 1, 4-rearrangements, the products undergo oxy-cope rearrangements

Negative ion mass spectra of the anion PhCHOMe obtained by deprotonation of benzyl methyl ether is given in Fig. 27.23.

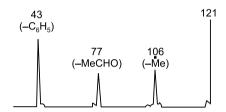


Fig. 27.23 Negation ion mass spectra of Ph CHOMe

#### 27.10.3.6 Ketones

The ion of the enolate of acetone eliminates a H atom to produce an ion radical and methane

$$(\mathsf{MeCOCH}_2)^- \begin{picture}(0,0) \put(0,0){\line(0,0){150}} \put(0,0)$$

Cyclohexanone enolate ion undergoes fragmentation as shown below:

# 27.10.3.7 Carboxylic Acids

Acetic acid on deprotonation yields MeCOO $^-$  along with the enolate ion (CH $_2$ COOH) $^-$ . Both the ions have characteristic fragmentations.

$$\begin{array}{c} \text{MeCOO}^- & \overset{\phantom{.}}{\longleftarrow} (\text{CH}_2\text{CO}_2)^{\scriptscriptstyle \mathsf{T}} + \overset{\phantom{.}}{\mathsf{H}} \\ \\ \text{[Me(CO}_2)] & \overset{\phantom{.}}{\longleftarrow} (\text{Me}^- + \text{CO}_2 \\ \\ \text{[O}^{\scriptscriptstyle \mathsf{T}}(\text{CO})] + \overset{\phantom{.}}{\mathsf{M}} e \\ \\ \text{(CH}_2\text{COOH)}^- & \overset{\phantom{.}}{\longleftarrow} (\text{CH}_2\text{COO})^{\scriptscriptstyle \mathsf{T}} + \overset{\phantom{.}}{\mathsf{H}} \\ \\ \text{[HO}^-(\text{CH}_2\text{CO})] & \overset{\phantom{.}}{\longleftarrow} \text{H}_2\text{CO}^- + \text{H}_2\text{O} \end{array}$$

Both the ions, MeCOO- and (CH<sub>2</sub>COOH)<sup>-</sup> are interconvertible.

#### 27.10.3.8 Amines

Isopropyl amine on deprotonation yields the anions Me<sub>2</sub>CH – NH and Me<sup>-</sup> (MeCH = NH). Both anions undergo fragmentation as shown below:

# 27.10.3.9 Nitro Compounds

Negative ion mass fragmentation of nitro benzene is given below:

# 27.10.4 Applications of Negative ion Mass Spectrometry

# 27.10.4.1 Qualitative and Quantitiative Analysis

Negative ion mass spectrometry is useful for both qualitative and quantitative analysis. For this a special ion source is incorporated in a special mass spectrometer. It has recently been suggested that in a MS ion source, attachment can be a dominant process for the formation of negative ion.

Bombardment of either the sample molecules or by an added gas (like nitrogen or orgen) generated slow electrons required for the production of a stable

carbanion. Following are given different types of stable carbanions in various types of empounds:

	Type of compound	Stable carbanion formed
1.	Ketones, aliphatic esters, aldehydes, alcohols, carboxylic acids, mercaptans and steriods	[M – H] <sup>–</sup>
2.	Condensed aromatics containing four or more condensed rigns or the same ring system in which a C – H group is replaced by N atom	[M] <sup>-</sup>
3.	Condensed aromatics with less than four condensed rings, phenanthridine, dinaphtho–[2, 1 : 1,' 2'] thiophene	[M – 2] <sup>–</sup>
4.	Compounds in which radical R is bound to a hetero atom	[M – R] <sup>–</sup>
5.	Nitrites, nitrogen heterocycles of lower mass, amines, amides.	CN⁻
6.	Nitro compounds	NO <sub>2</sub> -
7.	Compounds containing chlorine	Cl⁻, Cl⁻ <sub>2</sub>
8.	Sulphur compounds with low mass	S <sup>-</sup> , SH <sup>-</sup>

**Note:** Lower mass hydrocarbons, ethers, thiophenes etc. do not form stable carbanions. Their negative ion mass spectra exhibit a number of weak ion peaks and so these compounds are not suitable for negative ion spectroscopy.

# 27.10.4.2 Analysis of Bio-Organic Compounds

Under conventional 70 eV electron impact, most of the polar classes of bio-organic compounds are known to undergo extensive decomposition. However, using some other techniques like ionization, chemcial ionization and field desorption, the extensive fragments of such bio-organic compounds is minimised resulting in the formation of molecular ions.

Negative ion mass spectrometry is useful for the analysis of bio-active organic compounds. The formation of gas phase anionic species in molecules like amino acids or peptide derivatives is possible by using very low energy process—(2.9 eV). Using this technique, extensive decomposition is not observed and the formed anions correlated with the structure of the molecules. Following example illustrates dissociative resonance capture of 2.9 eV by the *p*-nitrobenzyl-N-acetyl amino acid derivatives (the *p*-nitrobenzyl group increases the electron affinity of the molecule, enhancing the initial electron capture to give the anion-radical intermediate).

#### 27.11 APPLICATIONS OF MASS SPECTROMETRY

# 27.11.1 Determination of Structure of Organic Compounds

On the basis of mass spectrometry only, it is normally not possible to determine the structure of the compound unless it is a simple molecule. For the determination of structure it is necessary to get information from other spectroscopic methods like IR, UV and NMR. The mass spectrum is useful to determine the exact molecular weight. This is particularly important in the case of unknown compounds which are availabe only in minute amounts. Prostaglandins and pheromones are some of the examples where mass spectrometry has provided valuable information for the determination of their structures.

Following steps are followed for the determination of structure of an organic compound by mass spectrometry:

- (i) Determine the molecular formula of the compound. From the mass spectrum find the all possible data from the molecular ion peak and the fragmentation peaks.
- (ii) The important ions are assigned formula.
- (iii) The neutral components lost are identified.
- (iv) Assign all possible structure which can give the various ions and neutral molecules as stated above.
- (v) On the basis of ions expected from all possible structures identify the simplest structures.
- (vi) The structure is finally confirmed by recording the mass spectrum of the compound with the known structure and comparing with the given spectrum.

# 27.11.2 Determination of Molecular Weight and Molecular Formula

Using 'High resolution' mass spectrometers it is possible to measure m/z values to 3 or 4 decimal places. This provides an accurate method for the determination of molecular weight.

Using the data given in Table 27.2, giving the exact masses the nuclides, the molecular weight and the molecular formula can be determined. As an example, the three molecules,  $O_2$ ,  $N_2H_4$  and  $CH_3OH$  all have atomic masses 32 using normal values for the atomic weights of O, N, H and C as 16, 14, 1 and 12 respectively. However, the actual atomic masses of the three molecules are different.

$$O_2 = 2(15.9949) = 31.9898$$
  
 $N_2H_4 = 2(14.0031) + 4(1.00783) = 32.0375$   
 $CH_4O = 12.00000 + 4(1.00783) + 15.9949 = 32.0262$ 

Thus, the high-resolution spectrometers can distinguish the three molecules (O<sub>2</sub>, N<sub>2</sub>H<sub>4</sub>,CH<sub>4</sub>O) and this is helpful for the determination of the molecular formula. The exact masses of nuclides are given in Table 27.3.

The molecular formula can also be determined from isotopic peaks. For detacts see section 27.5.4.1.

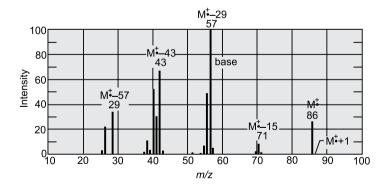
# 27.11.3 Miscellaneous Applications

Gas chromatography-mass spectrometry has been successfully used for detection of pesticides in the environment. A number of tetrachloro-p-dioxins and their derivatives are discharged into the environment as byproducts of the manufacture of chlorinated herbicides and pesticides and byproduct of municipal incineration can also be detected.

GC-MS has also been used for the analysis of drugs and their metabolites.

#### 27.12 SOLVED PROBLEMS

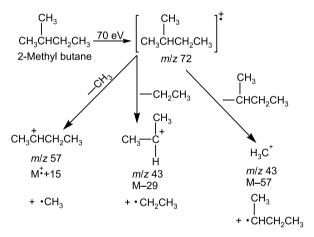
1. The mass spectrum of hexane is given below:



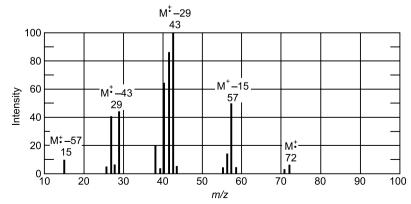
Assign the fragments to the various m/z peaks and explain how they occur. Following important fragments are expected:

The five fragments are clearly visible in the above mass spectrum.

2. What fragments are expected to be obtained in the mass spectrum of 2-methylbutane?



In view to the above, four peaks will be observed at m/z 72, 57, 43 and 15. The mass spectra of 2-methyl butane is given below:



3. What fragments are expected to be obtained in the mass spectrum of ethyl sec. butyl ether?

What fragments are expected to be obtained in the mass spectrum of ethyl sec. butyl ether?

$$CH_{3}-CH_{2}-CH-\ddot{\bigcirc}-CH_{2}-CH_{3}$$

$$CH_{3}-CH_{2}-CH-\ddot{\bigcirc}-CH_{2}-CH_{3}$$

$$CH_{3}-CH_{2}-CH-\ddot{\bigcirc}-CH_{2}-CH_{3}$$

$$CH_{3}-CH_{2}-CH-\ddot{\bigcirc}-CH_{2}-CH_{3}$$

$$CH_{3}-CH_{2}-CH-\ddot{\bigcirc}-CH_{2}-CH_{3}$$

$$CH_{3}-CH_{2}-CH-\ddot{\bigcirc}-CH_{2}-CH_{3}$$

$$CH_{3}-CH_{2}-CH-\ddot{\bigcirc}-CH_{2}-CH_{2}$$

$$CH_{3}-CH_{2}-CH-\ddot{\bigcirc}-CH_{2}-CH_{2}$$

$$CH_{3}-CH_{2}-CH-\ddot{\bigcirc}-CH_{2}-CH_{2}$$

$$CH_{3}-CH_{2}-CH-\ddot{\bigcirc}-CH_{2}-CH_{2}-CH_{2}$$

$$CH_{3}-CH_{2}-CH-\ddot{\bigcirc}-CH-\ddot{\bigcirc}-CH_{2}-CH-\ddot{\bigcirc}-CH-\ddot{\Box}-CH-\ddot$$

So, the peaks obtained are m/z 102, 87, 73 and 29.

What fragments are expected in case of ethyl methyl ketone?

$$H_{3}C$$

$$C = \overset{\bullet}{O}^{+} \xrightarrow{-CH_{3}^{+}} C_{2}H_{5}C = \overset{\bullet}{O} \xrightarrow{-C_{2}H_{5}} C_{2}H_{5}C = \overset{\bullet}{O}$$

$$m/z = 57$$

$$M^{+} m/z = 72$$

$$H_{3}C$$

$$C = \overset{\bullet}{O} \xrightarrow{-C_{2}H_{5}^{+}} H_{3}C C = \overset{\bullet}{O} \xrightarrow{-C_{2}H_{5}^{+}} H_{3}C C = \overset{\bullet}{O}$$

$$m/z = 43$$

The peaks obtained are at m/z 72, 57 and 43.

The base peak results in the loss of larger alkyl group. Thus, the base peak is at m/z 43.

5. What fragments are expected in the case of aliphatic and aromatic aldehydes?

The aliphatic aldehydes give M – 1 peak due to cleavage of C—H bond next to oxygen. They also give M–R peak (m/z 29, CHO<sup>+</sup>). The M–1 peak is a diagnostic peak even for long chain aldehydes. The C<sub>4</sub> and higher aldehydes undergo McLafferty cleavage of the  $\alpha$ ,  $\beta$  C—C bond to give a major peak at m/z 44, 58 or 72 depending on the  $\alpha$ -substituent. In straight chain aldehydes, other unique peaks are M–18 (loss of H<sub>2</sub>O), M–28 (loss of ethylene), M–43 (loss of CH<sub>2</sub>=CH— $\dot{\rm O}$ ) and M–44 (loss of CH<sub>2</sub>=CH— $\dot{\rm O}$ H).

Aromatic aldehydes are characterized by a large molecular ion peak  $\left(M-1,Ph-C\equiv \overset{+}{O}\right)$ .

The M-1 ion  $C_6H_5C \equiv \ddot{O}^+$  eliminates CO to give phenyl ion (m/z 77), which in turn eliminates CH  $\equiv$  CH to give  $C_4H_3^+$  ion (m/z 51).

**6.** Suggest a structure to a compound which gives the following mass spectral data:

As seen, there are 13 peaks. The most abundane ion peak at m/z 31 (100%) cannot be due to  $M^+$  ion as the (M+1) peak at m/z (66%) will have an high abundance for organic compound having low carbon content (or low molecular weigh). So, the molecular ion peak is m/z 32. Other peaks are identified as:

m/z	M-1	M	M + 1	M + 2
Relative	31	32	33	34
abundance as	(100)	(66)	(0.98)	(0.14)
% of M <sup>+</sup> abundance	_	100	1.48	0.21

The M + 1 peak which has 100% abundand suggests the presence of one carbon and the M + 2 abundance indicates one O atom, both of which together account for 12 + 16 = 28 amu.

The remaining mass is due to H and the molecular formula of the compound is CH<sub>4</sub>OH (methanol)

The structures of the ions are:

$$m/z$$
 32 33 15  $CH_3^- OH^+ CH_3^- O^+ CH_3^+$ 



### I. Multiple Choice Questions

- 1. Which of the following statement/statements is/are correct?
  - (a) In mass spectrometer, the molecules in the gaseous phase are bombarded with a beam of energetic electrons (70 eV).
  - (b) The molecular ions obtained as a resulted above (a) are radical cations.
  - (c) The mass spectrometer separates the various ions including the molecular ion on the basis of their mass-to-charge ratio.
  - (d) All are correct.
- 2. In the fragmentation of molecular ion derived from ammonia, the fragment(s) correspond to m/z.
  - (a) 16

(b) 15

(c) 14

- (d) All are correct
- 3. The presence of which of the following elements will give  $M^{+}$  + 2 peak
  - (a) C

(b) N

(c) S

- (d) O
- 4. In the mass spectra of 3,3-dimethyl hexane, the fragments are:

$$\begin{array}{c} \operatorname{CH_3} \\ (a) \ \operatorname{CH_3--CH_2--C} \\ \\ \operatorname{CH_3} \\ \end{array}$$

$$(b)\begin{bmatrix} \mathsf{CH_3} & \mathsf{CH_3} \\ \mathsf{CH_3} \mathsf{CH_2} - \mathsf{CH} - \mathsf{CH_2} - \mathsf{CH_3} \\ \mathsf{CH_3} \end{bmatrix}$$

- (c) 'CH2CH2-CH2-CH3
- (d) All the above
- **5.** A compound gives molecular ion peaks at m/z 86. This peak can arise from:
  - (a) n-Hexane

(b) 2-Methyl pentane

(c) Either (a) or (b)

(d) None of the above



- **1.** (d)
- **2.** (d)
- 3.(c),(d)
- **4**. (d)
- 5.(c)

# **II. Short and Long Answer Questions**

- 1. What are the basic principles of mass spectrometry?
- **2.** How is a mass spectrum represented?
- **3.** How will you determine the molecular formula using mass spectrometry?
- **4.** How will you find the presence of nitrogen using mass spectrometry?
- 5. How is the presence or absence of S, Cl and Br inferred from a mass sepectrum?

- **6.** How is correct molecular weight determined using mass spectrometry?
- 7. How is correct molecular ion peak recognized in a mass spectrum?
- **8.** In the mass spectrum of 2,2-dimethyl butane the most intense peak occurs at *m/z* 57. What carbocation this peak represent and how it arises from the molecular ion?

- 9. Using mass spectrometry, how will you establish the structure of an unknown compound?
- 10. What types of peaks to you expect in case of 3-heptanone?

- **11.** On the basis of mass spectra, how will you differentiate between the two isomeric alkanes *n*-octane and 2,2,4-trimethylpentane?
- **12.** On the basis of mass spectra, how will you differentiate between butyl isopropyl ether and butylpropyl ether?
  - **Sol.** Both will give molecular ion peak ( $M^+$ ) at m/z 116.

The molecular ion from butyl isopropyl ether will give an  $M^{\frac{1}{4}}-15$  peak at m/z 101.

However, the molecular ion from butylpropyl ether will give an  $M^{+} - C_2H_5$  peak at m/z 87.

$$CH_3 CH_2 CH_2 \stackrel{\leftarrow}{\text{CH}_2} \stackrel{\leftarrow}{\text{CH}_2} \stackrel{\leftarrow}{\text{CH}_3} \stackrel{\leftarrow}{\text{CH}_3} CH_2 \stackrel{\leftarrow}{\text{CH}_2} \stackrel{\leftarrow}{\text{CH}_2} CH_2 \stackrel{\leftarrow}{\text{CH}_2} CH_3$$

$$M^{\stackrel{\leftarrow}{\text{T}}} = 15$$

$$m/z 87$$

13.  $\beta$ -Myrcene on mass fragmentation gives peaks at m/69 and 67. Give structures to the fragments and explain their formation.

#### Hint:

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**14.** On the basis of mass spectrometry, how will you differentiate between primary, secondary and tertiary alcohol?

[Hint: In case of alcohols, cleavage of the C — C bond next to oxygen atom is of general occurence. So primary alcohols exhibit a prominent peak due to  $CH_2 = \ddot{Q}H$  (m/z 31). Secondary and tertiary alcohols cleave analogously to give a prominent peak due to  $\frac{R}{H}C = \ddot{Q}H$  [m/z 45 ( $R = CH_3$ ] and  $\frac{R}{R'}C = \ddot{C}OH$  [m/z = 59 ( $R = R^1 = CH_3$ )]. However, the largest substituent is most readily expelled.]







# **Polarimetry**

#### 28.1 INTRODUCTION

Polarimetry is one of the oldest technique. It deals with the measurement of the change of direction of the vibrations of plane of polarised light on interaction with optically active substances.

#### 28.2 PLANE POLARIZED LIGHT

A beam of ordinary light is known to consists of electromagnetic waves oscillating in many planes. If this ordinary ray of light is passed through a polarizer (*e.g.*, a Nicol prison, a special type of prism, obtained by combining two prisms of iceland spar suitably cut), the light emerging has its vibrations in one plane only. The emerging beam of light having oscillations in a single plane is said to be plane polarised (Fig. 28.1).

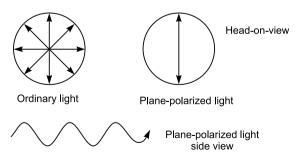


Fig. 28.1 Plane-Polarized light

When a plane-polarized light is passed through a solution of an optically active compound, the plane of polarized light is rotated (Fig. 28.2).

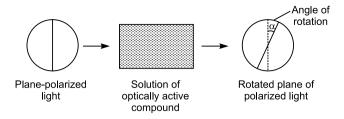


Fig. 28.2 Illustration of optical activity

A compound that rotates the plane-polarized light to the left (anticlockwise) is called **levorotatory** and a compound that rotates the plane-polarized light to the right (clockwise) is called **dextrorotatory**. (Fig. 28.3).

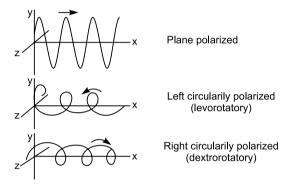


Fig. 28.3 Illustration of Levorotatory and Dextrorotatory

As a matter of convention, rotation to left is given a minus sign (–) and rotation to right is given a plus (+) sign.

#### 28.3 OPTICAL ACTIVITY

When plane polarized light is passed through certain organic liquids, solutions (like cane sugar in water) or quartz crystals, there is rotation of the plane of polarized light.

This property by virtue of which the plane of polarization of polarized light is rotated is called **optical activity**. The substance that possess this property is called **optically active substance**. As already stated, the substance that rotates the plane of polarized light towards right (clockwise) is called **dextro rotatory** (+) and the substance that rotate the plane of polarized light towards left (anti clockwise) is called laevo rotatory (-). A mixture of these two varties in equal proportion is optically inactive and is called a **racemic form**.

The rotation of plane-polarized light is an intrinsic property of optically active molecules. As already stated, when a polarized beam is passed through the solution of an optically active compound, the plane of the polarized light is rotated through

an angle  $\alpha$  (angle of rotation) (Fig. 28.2). This rotation depends on the number of optically active moleculs encountered. Thus,  $\alpha$  is proportional to both the concentration and the length of the sample solution.

The **specific rotation** is the characteristic property of an optically active substance and is expressed as

$$\left[\alpha\right] = \frac{\alpha}{l \times c} \tag{i}$$

where

 $[\alpha]$  = specific rotation in degrees

 $\alpha$  = observed angle of rotation in degrees.

l =length of the solution in decimeters (dm; 10 cm)

c =concentration of the sample solution in g/mL.

From the above equation, the specific rotation is defined as the observed angle of rotation at a concentration of 1g/mL and path length of 1 dm. As a matter of convention, specific rotation is reported as  $\left[\alpha\right]_D^T$ , when t stands for temperature and D for D-line of sodium used for determination.

#### 28.4 KINDS OF MOLECULES ANALYSED BY POLARIMETRY

It has already been stated that only a molecule which is optically active can be analysed by polarimetry. An optically active organic molecule is that which contains an asymmetric carbon. An asymmetric carbon is one which is attached to four different groups. As an example, in lactic acid CH<sub>3</sub> CH(OH) COOH, the asymmetric carbon (bold face) is attached to the groups CH<sub>3</sub>, H, OH, COOH. It has been shown that such molecules can be represented by two arrangements which are mirror images of each other (Fig. 28.4). Chemically, the two representations ore virtually identical.

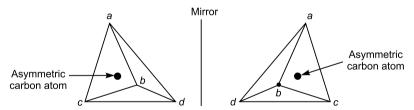


Fig. 28.4 Two possible arrangements four groups of molecule having an asymmetric carbon

#### 28.5 THEORETICAL CONSIDERATIONS

Optical activity was first observed by Biot (1815), who found that there are two types of quartz crystals, which rotate the plane of polarized light in opposite directions. It was subsequently found that even some compounds in solution exhibit the same property. The optical activity in solution is attributed to some molecular property. (*viz.*, arrangement of groups) which is retained in solution. It was, infact, Pasteur who

studied the salts of tartaric acid. He found that optically inactive sodium ammonium tartarate is a mixture of two different kinds of crystals, which were mirror image of each other and rotated the plane polarized light in opposite direction. Pasteur could separate the two kinds of crystals using a hand lens and pair of tweezers. Thus, ordinary sodium ammonium tartarate is a racemic mixture.

The cause of optical activity was first explained by Van Hoff and LeBell (1874), who postulated that the four valencies of carbon are directed in three dimensional space towards the four corners of a regular tetrahedron, the carbon atom being at the centre. If four groups (e.g., a, b, c, d) are attached to carbon, the molecule becomes asymmetric. Thus, for a compound *Cabcd*, there are two ways of arrangement of the four groups. These two structures are non-super imposible and are mirror images of each other. Thus, the presence of four different groups or atoms attached to a carbon atom is responsible for optical activity. This carbon is called asymmetric carbon atom (Fig. 28.5).

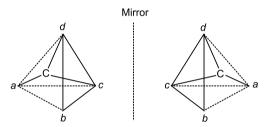


Fig. 28.5 Non-superimposible Mirror images

It has been noted that the compound  $C_{abcd}$  contains an  $sp^3$  hybrid carbon and is linked to four different monovalent atoms or groups. This carbon atom is known as asymmetric carbon atom.

Some other eamples of asymmetric objects include the letter P and a human hand. The letter A and a circle are examples of symmetric objects (Fig. 28.6).

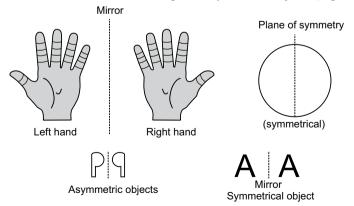


Fig. 28.6 Symmetrical and asymmetric objects

#### 28.6 POLARIMETER

It is used to measure the optical activity. The polimeter is made of two Nicol prisms, the polarizer and the analyzer. The polarizer is fixed and the analyzer can be rotated. The sample tube is placed between the polarizer and analyser (Fig. 28.7).

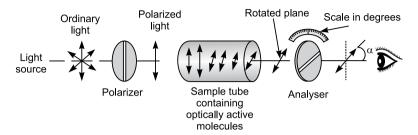


Fig. 28.7 Diagrammatic representation is a polarimeter

Initially, an optically inactive medium (solvent or air) is filled in the sample tube and the polarized sodium light from the polarizer passes through it. Subsequently, the analyzer is turned to get a dark field. This gives a zero reading on the circular scale (Fig. 28.7). Finally, the solution of the optically active compound is placed in the sample tube and the analyzer is turned to again get the dark field. The angle of rotation ( $\alpha$ ) is noted in digrees on the circular scale. The specific rotation is calculated from equation (i).

The visual polarimeters (discribe above are tiring on the eyes resulting in varying precision.

This problem has been overcome by the development a number photo-electric polarimeters.

#### 28.7 APPLICATIONS OF POLARIMETRY

If the concentration of an optically active sample is known, then by equation (i) the specific rotation can be calculated, which is helpful for the identification of the compound.

On the other hand, if specific rotation of a optically active sample is known, its concentration can be calculated by using the equation (i).

Polarimetry can also be used for the determination of an optically active substance in presence of other optically inactive substances. Since optical rotation is additive, so readings can also be made in the presence of known fixed amount of some other optically active species.

An important application of polarimetry is in sugar industry. Sucrose can be determined directly (in the absence of any other optically active substance) by measurement of the angle of rotation,  $\left[\alpha\right]_{D}^{\beta 0}$  is equal to + 66.5°. In case other

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optically active substances are present, it is necessary to measure the optical rotation before and after hydrolysis of sugar solution. It is known that sucrose undergoes hydrolysis in the presence of acid and the resulting mixture of fructose and glucose has optical rotation of  $-19.8^{\circ}$ .

The specific rotation changes from +66.5 (pure sucrose) to  $(-92.4^{\circ} + 52.7^{\circ})/2$  =  $-19.8^{\circ}$  on inversion.

The amount of sucrose present is calculated from the difference in rotation before and after inversion.

#### **Optical Isomersion**

Optically active comopounds are known to exist in two isomeric forms, which have different spatial arrangements of atoms. Such isomers are called optical isomers and the phenomenon is known as optical isomerism. The optical isomers are non-superimposable and are called enantiomers. These have same physical and chemical properties but have different crystalline shape, biological activities and rotate the pane polarized light equally but in opposite direction. Some of the physical properties

at 2-methyl-1-butanol 
$$CH_3$$
— $CH_2$ — $C$ — $CH_2$ OH are given Table 28.1.  $CH_3$ 

**Table 28.1** Some Physical properties of the enantioners of  $\alpha$ -methyl-1-butanol

Property	(+)-2-methyl-1-butanol	(–)-2-methyl-1-butanol
Optical rotation	+5.756°	–5.756°
B.P.	401.9K	401.9K
Density	0.8193	0.8193
Refractive Index	1.4173	1.4173

As seen, the physical properties of the two enantiomers of 2-methyl-1-butanol are exactly same.

As already stated, the enantiomers differ in their biological activities. A typical example of such compound is an biologically active anti-nausea drug used for expectant mothers in Europe and Great Britain (1959-1962). This drug called thalidomide was used as a sedative and subsequently withdrawn since it caused catastropic birth defects to children born to women who took thalidomide during pregnancy. It was subsequently found that this drug was sold as a mixture of its two enantiomers and

each of these has a different biological activity. Although one of the enantiomer had the desired therapeutic effect, the other isomer was responsible for the birth defects.

Fig. 28.7(a) Two enantiomers of thalidomide

It is possible to distinguish between the two enantiomers of a compound by their behaviour towards plane polarized light. As an example, the spatial arrangement of groups in case of lactic acid are represented as shown in Fig. 28.8.

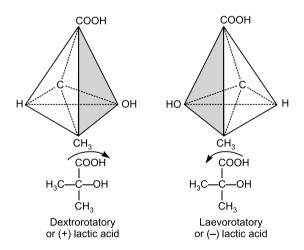


Fig. 28.8 Two enantiomers of Lactic Acid

The two models of lactic acid (Fig. 28.8) are not superimposible and are related to each other as an object and its mirror image. The isomer that rotates the plane of polarisation to right is dextro rotatory and its mirror image which rotates the plane of polarisation to the left is laevo rotatory. These can also be indicated as (+) and (-) lactic acid respectively.

## Configuration

It is the representation of a molecule in space and is best seen by molecular models. As an example, the configuration of the optical isomers of lactic acid are shown below (Fig. 28.9).

Fig. 28.9 Configuration of the optical isomers of lactic acid

According to Fisher projection, the molcule is so held that the asymmetric carbon atom is in the plane of paper. The groups at the top and bottom are inclined below the plane of the paper and the groups on the left and right are inclined above the plane. The Fisher projection formula of lactic acid are given below (Fig. 28.10).

Fig. 28.10 Fisher projection formula of lactic acid

In the enantiomers having a H attached to the asymmetric carbon atom, the projection formula showing the group other than H (OH group, in case of lactic acid) on the left hand side is assigned L-configuration and on the right hand side is assigned D-configuration. Thus, formula (A) is D-lactic acid and formula (B) is L-lactic acid. The direction of rotation of polarized light is also indicated along with the configuration D and L by writting positive sign (+) and negative sign (-) for dextro and leavo isomers respectively. Thus, the representation A is D(+) lactic acid and B is L(-) lactic acid.

## **Molecules having More than One Asymmetric Centres**

A typical example of a compound having two asymmetric carbons is tartaric acid. Each asymmetric carbon is attacted to the same four different groups, *i.e.*, –OH, –H, –COOH and –CH(OH)COOH. The spatial arrangement of various groups in tartaric acid are shown below (Fig. 28.11).

 Dextro-tartaric acid: Rotates the plane of polarization of light to the right. The rotation due to upper half is argumented by one due to the lower half.

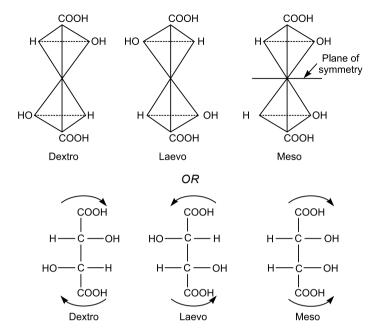


Fig. 28.11 Special arrangement of various groups in tartaric acid

- Laevo-tartaric Acid: Mirror image of the above. Rotates the plane
  of polarization of light to the left. The rotation due to upper half is
  argumented by the one due to the lower half.
- Racemic-tartaric Acid: A 1:1 mixture of the above two varities and so is inactive. However, it can be resolved into the dextro and laevo forms.
   In fact, it is inactive due to external compensation.
- Meso-tartaric Acid: An inactive variety. The rotation due to upper half is compensated by the rotation due to the lower half. Cannot be resolved into dextro and laevo forms. So it is inactive due to internal compensation. Unlike dextro and laevo forms, it has a plane of symmetry.

# EXERCISES

- 1. Explain the terms polarimetry, plane polarized light and optical activity.
- **2.** What is specific rotation. How it is measured?
- 3. What types of molecules can be analysed by polarimetry?
- **4.** Discuss the theory of polarimetry.
- **5.** Describe a polimeter showing its various components and their functions.
- **6.** Discuss the applications of polarimetry.
- 7. What are characteristics of an optically active substance?
- **8.** Explan the term optical isomerism.

# Part-VI

# **Instrumental Method of Analysis of Inorganic Compounds**

- Microwave Spectroscopy
- Nuclear Quadrupole Resonance Spectroscopy
- Raman Spectroscopy
- Mossbauer Spectroscopy
- Emission Spectrometry

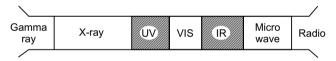


# Chapter 29

# **Microwave Spectroscopy**

#### 29.1 INTRODUCTION

Microwave spectroscopy is also known as **rotational spectroscopy**. The microwave region lies between far IR and radio frequency in the electromagnetic spectrum and extends from  $100 \ \mu m \ (3 \times 10^{13} \ Hz)$  to 1 centimeter  $(3 \times 10^{10} \ Hz)$ .



Electromagnetic Spectrum

Microwave spectroscopy is very useful for the determination of structures of molecules which do not give good results by using IR or Raman spectroscopy. The spectras are the result of transitions between the rotational energy levels of a molecule in gaseous state due to absorption of radiations striking in the mirowave region. Only molecules which possess a permanent dipole moment (e.g.,  $H_2O$ , HCl, CO, NO etc.) exhibit microwave spectra. Molecules not possessing permanent dipole moments (like  $H_2$ ,  $Cl_2$   $CO_2$  etc.) do not show microwave spectra.

When a molecule (having permanent dipole) rotates, an electric field is generated which interacts with the electric component of the microwave radiation. This result in the absorption of energy and thus the rotation of the molecule gives a spectrum. However, molecules not possessing permanent dipole moments do not interact and are said to be 'microwave inactive'. On the basis of theoretical considerations, it has been shown that microwave spectra is only about 1% of the optical absorption.

Using Microwave spectrum it is not possible to correlate certain parts of the spectrum to certain parts of the molecule. The microwave spectrum is due to the molecule as a whole.

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# 29.2 DIFFERENCES BETWEEN MICROWAVE SPECTROSCOPY AND IR SPECTROSCOPY

- (i) The microwave spectra is characteristic of molecules as a whole. However, IR spectra is characteristic only of the functional group present in the molecule.
- (ii) In the microwave spectra the resolution of lines is much greater compared to that obtained in the IR method.
- (iii) In microwave spectroscopy, the sample must be present in only the gaseous state. However, in IR spectroscopy the molecule can be used in solid, liquid or gaseous state. This is because rotational energy is not quantized in condensed phase like solid or liquid phase. These are quantized in the gaseous states of chemical system.
- (*iv*) The microwave spectras is of absorption type whereas IR spectra may be both absorption or emission type.

#### 29.3 THEORY OF MICROWAVE SPECTROSCOPY

It is known that rotational energy along with all other forms of molecular energy is quantised. This implies that in rotational energy there are certain permitted values of energy, commonly known as rotational energy levels which can be calculated for any molecule using the well known Schrodinger equation.

The mathematical calculations for simple molecule is fairly simple. However, for complex molecules the procedure involved is tedious and approximations are used. In the present section, the discussion on rotational energies of molecules in confined to linear diatomic molecules, rotational effects and rigid rotor approximation. It is, however, assumed that molecular dimensions are not affected by molecular vibration and molecular rotation.

#### 29.4 DIATOMIC MOLECULE AS A RIGID ROTATOR

A diatomic molecule is considered as rigid rotator having free axis of rotation; its nuclei are separated by a definite distance. Let us consider a diatomic molecule AB in which A and B are Joined by a ridig bar. Its length is given by

$$r = r_1 + r_2 \qquad \dots (i)$$

where  $r_1$  and  $r_2$  are the distances of the atoms A and B from the centre of gravity G of the molecule AB (Fig. 29.1). The moment of inertia I about G is given by the equation.

where  $m_1$  and  $m_2$  are the masses of the atoms A and B.

The system is balanced about its centre of gravity, G. In this case the following conditions are satisfied.

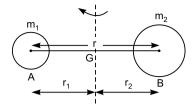


Fig. 29.1 A rotating diatomic molecule

$$m_1 r_1 = m_2 r_2 \qquad \dots (iii)$$

Substituting the equation (iii) in (ii), we get

$$I = m_2 r_2 r_1 = m_1 r_1 r_2$$
  
=  $r_1 r_2 (m_1 + m_2)$  ...(iv)

From equations (i) and (ii), we get

$$m_1 r_1 = m_2 r_2 = m_2 (r - r_1)$$
 ...(v)

On solving the equations (iv) and (v)

$$r_1 = \frac{m_2}{m_2 + m_1} r$$
 and  $r_2 = \frac{m_1}{m_2 + m_1} r$  ...(vi)

Substituting the values of  $r_1$  and  $r_2$  in equation (ii), we get

$$\begin{split} I &= \frac{m_1 m_2^2}{\left(m_1 + m_2\right)^2} r^2 + \frac{m_1^2 m_2}{\left(m_1 + m_2\right)} r^2 \\ &= \frac{m_1 m_2^2 + m_1^2 m_2}{\left(m_1 + m_2\right)^2} r^2 \\ &= \frac{m_1 m_2 \left(m_1 + m_2\right)}{\left(m_1 + m_2\right)^2} r^2 \\ &= \frac{m_1 m_2}{m_1 + m_2} r^2 \\ &= \mu r^2 & \dots (vii) \end{split}$$

where  $\mu$  is the reduced mass (of the diatomic molecule). The above equation (*vii*) gives the moment of inertia in terms of atomic mass and bond length.

It has already been stated that only a molecule having a permanent dipole generates an electric field which interacts with the electric component of the microwave region. Assuming that a diatomic molecule behaves like a ridig rotator, the rotational energy levels can be calculated by using Schrodinger's equation.

$$E_J = \frac{h^2}{8\pi^2 I} J(J+I)$$
 Joules, where J = 0, 1, 2; h is Planck's constant,

*I* is the moment of Inertia and *J* is rotational quantum number.

$$\frac{J(J+1)h^2}{4\pi^2} = \text{Square root of the rotational angular moment.}$$

The spectra are normally expressed in terms of wave numbers in the rotational region. It is thus useful to consider energies in these units.

$$\varepsilon_J = \frac{E_J}{hc} = \frac{h^2}{8\pi^2 Ihc} J(J+I) \text{ cm}^{-1} \qquad \dots (viii)$$

$$= \frac{h}{8\pi^2 Ic} (J+1) \text{ cm}^{-1} \qquad ...(ix)$$

where c is the velocity of light expressed in cm per second.

 $\frac{h}{8\pi^2 Ic}$  is commonly expressed as *B*.

So the equation (ix) can be written as

$$C_I = J(J+1) B \text{ cm}^{-1}$$
 ...(x)

Where B is the rotational constant and is expressed as Mc per second or cm<sup>-1</sup>, *i.e.*,

$$B = \frac{h}{8\pi^2 Ic} \text{cm}^{-1} \qquad \dots (xi)$$

using the equation (x) the energy levels can be diagrammatically shown as in Fig. 29.2.

when J = 0, equation (x) can be

$$\varepsilon_I = B(J+I) \text{ cm}^{-1} = B0 (0+1) = 0$$
 ...(xii)

From the above equation (xii) it is clear that the molecule is not at all rotating. However, when J = 1, equation (x) can be

$$\varepsilon_J = B \cdot 1 \ (1+1) = 2B \ \text{cm}^{-1}$$
 ...(xiii)

The above equation (*xiii*) shows that a rotating molecule has its lowest angular momentum.

In a similar way, value of  $\varepsilon_J$  can be calculated for J = 2, 3, 4... The allowed rotational energy levels of rigid diatomic molecules are shown in Fig. 29.2.

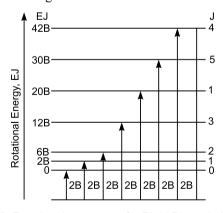


Fig. 29.2 Rotational spectrum of a Rigid Diatomic Molecule

Let us consider transition from rotational level of rotational quantum number J' to higher quantum number J. The difference in energy  $(\Delta E_1)$  between the two levels is given by

$$\begin{split} \Delta E_J &= E_J - E_{J'} \\ &= \frac{h^2}{8\pi^2 I} J \big( J + 1 \big) - \frac{h^2}{8\pi^2 I} J' \big( J' + 1 \big) \qquad \dots (xiv) \end{split}$$

The energy evolved ( $\Delta E_J$ ) according to quantum theory is given out in the form of spectral lines, whose frequency expressed in wave number is given by

$$\overline{v} = \frac{\Delta E_J}{hc} = \frac{h^2}{8\pi^2 Ihc} [J(J+1)] - J'(J'+1)$$

$$= \frac{h}{8\pi^2 Ic} [J(J+1) - J'(J'+1)]$$

$$= B[J(J+1) - J'(J'+1)] \qquad \dots (xv)$$

In case J = 1 and J' = 0, equation (xv) can be

$$\overline{v}_{J_0} \rightarrow {}_1 = [B(1+1) - 0(0+1)]$$

$$= 2B \text{ cm}^{-1} \qquad ...(xvi)$$

It follows from the above equation (xvi) that an absorption line appears at 2B cm<sup>-1</sup>.

In case the molecule is raised from J' = 1 to J = 2 level due to absorption of energy is microwave region, equation (xvi) becomes

$$\overline{v}_{J_0} \rightarrow {}_2 = B [2 (2+1) - 1 (1+1)]$$

$$B = (6-2) = 4B \text{ cm}^{-1} \qquad ...(xvii)$$

So an absorption line appears at 4B cm<sup>-1</sup>. In general, raising of a molecule from J to J + 1, equation (xvii) become

$$v_{J \to J+1} = B [(J+1) (J+2) - J (J+1)]$$
  
=  $B [J^2 + 3J + 2 - J^2 - J]$   
=  $2B (J+1) \text{ cm}^{-1}$  ...(xviii)

From the above equation (*xviii*), it is found that step wise raising of rotational energy gives an absorption spectrum considering of lines at 2*B*, 4*B*, 6*B*...cm<sup>-1</sup> However, a similar lowering gives similar emission spectrum (*see* Fig. 29.2).

#### 29.5 SELECTION RULES FOR ROTATIONAL SPECTRA

It has already been stated that a molecule must have a dipole moment in order to enable it to give rotational spectrum. All transitions are not permitted. The transitions are given by the selection rule.

$$\Delta J = \pm 1 \qquad \dots (xx)$$

Thus, only those transitions are permitted in which there is decrease or increase in the rotational quantum number by unity. This implies that  $J = 0 \rightarrow 1 \rightarrow J = 2 \rightarrow J = 4...$  transitions are not possible; these transitions are forbidden spectroscopically.

The theory of microwave spectroscopy as given in section 29.3 is confirmed by considering the example of carbon monoxide.

The moment of inertia and the bond length in case of carbon monoxide can be calculated from the observed spectrum.

The first line corresponding to J = 0 is observed in the rotational spectrum of carbon monoxide at  $3.84235 \text{ cm}^{-1}$ .

i.e., 
$$\overline{v}_J = 3.84235 \text{ cm}^{-1}$$
 ...(xxi)

Using the equation (xviii), we get

$$\overline{v}_J = 2B \left( J + 1 \right)$$

J = 0

Since

$$\overline{v}_I = 2B(0+1) = 2B$$

Using the equations (xx) and (xxi), we get

$$2B = 3.84235 \text{ cm}^{-1} \text{ or } B = 1.92118 \text{ cm}^{-1} \dots (xxii)$$

From equation (xi), we get

$$B = \frac{h}{8\pi^2 Ic} \text{ or } I = \frac{h}{8\pi^2 Bc}$$

$$B = \frac{6.624 \times 10^{-27}}{8\pi^2 \times 1.92118 \times 2.99976 \times 10^{10}}$$
$$= 1.45673 \times 10^{-39} g \text{ cm}^2 \qquad \dots(xxiii)$$

The bond length can be calculated by finding the value of reduced mass of CO as per equation (*viii*)

$$\mu = \frac{m_1 m_2}{m_1 + m_2} \times \frac{1}{N}$$

Here  $m_1$  is 12.0000 g (C) and  $m_2 = 15.9949 g$  (O)

$$\therefore \qquad \qquad \mu = \frac{12 \times 15.9949}{12 + 15.9949} \times \frac{1}{6.024 \times 10^{23}} g \qquad ...(xxiv)$$

As per equation (vii),

$$I = ur^2$$

Substituting equations (xxiii) and (xxiv) in equation (vii), we get

$$1.45673 \times 10^{-39} g \text{ cm}^2 = \frac{12 \times 15.9949}{12 + 15.9949} \times \frac{1}{6.024 \times 10^{23}} r^2$$

or 
$$r^2 = 1.28 \times 10^{-16} \text{ cm}^{-1}$$
  
or  $r = 1.131 \text{ Å}$ 

It was found that the rotational absorption of  $^{12}$ C  $^{16}$ O was at 3.84235 cm $^{-1}$  (C = 12 and O = 16) and that of  $^{13}$ C  $^{16}$ O was at 3.7637 cm $^{-1}$ . From these figures, the values of B and B' comes at.

Since 
$$B = 1.92118 \text{ cm}^{-1} \text{ and } B' = 1.83669 \text{ cm}^{-1}$$

$$B = \frac{h}{8\pi^2 I_c} \text{ and } B' = \frac{h}{8\pi^2 I'C}$$

$$\frac{B}{B'} = \frac{h}{8\pi^2 I_c} \times \frac{8\pi^2 I'C}{h} \times \frac{I'}{I} = \frac{\mu'r^2}{\mu r^2} = \frac{\mu'}{\mu}$$

$$\frac{\mu'}{\mu} = \frac{1.92118}{1.83669} = 1.046$$

On the basis of the mass of O to be 15.9994 g and that of C as 12.0000 g, we get

$$\frac{\mu'}{\mu} = 1.046 = \frac{15.9994 \times m'}{15.9994 + m'} \times \frac{12 + 15.9994}{12 \times 15.9994}$$
$$m' = 13.0007$$

We, thus, find that the atomic weight of <sup>13</sup>C comes out as 13.0007. This value is in agreement with the value obtained by other methods.

#### 29.6 INSTRUMENT FOR MICROWAVE SPECTROSCOPY

The microvave spectrometer consist of following six components.

٠.

- 1. The Source of Microwave: The main source of microwave radiations is an electronic device known as **klystron**. Since this device emits microwave radiation of a very narrow frequency range, it acts as its own monochromater. The applied voltage to the klystron determines the frequency of the emitted radiation.
- 2. Wave Guide: It is a rectangular tube made of copper or silver conducts the radiations. The wave gluides are bent or tapered in order to maintain the direction of the emitted beam from the klystron. The wave guides are evacuated as the air if present causes considerable absorption of radiation.
- **3. Window for Introduction of Sample:** The gaseous sample is introduced through a window situated in the wave guide tube; which is closed by thin mica at both ends. The pressure of the gas is so adjusted in order to make sharp absorption lines.
- **4. Detector:** The detector is a crystal of quartz situated at the far end of the wave guide tube. The detector is suitably tuned to appropriate frequency.
- **5. Amplifier:** The vibrations from the quartz crystal produce an electrical signal which is amplified.

**6. Oscilloscope:** The amplified signal from the amplifier are displayed as a pattern on an oscilloscope screen or recorded on a chart by pen-ink recorder.

Various parts of the microwave spectrometer are represented by a simple diagram (Fig. 29.3)

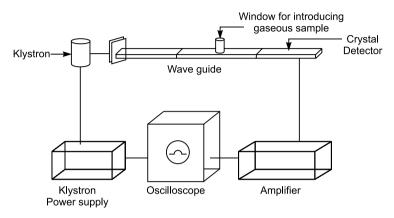


Fig. 29.3 Diagramatic representation of various parts of a microwave spectrometer

The radiations (monochromatic) of various wavelengths in the microwave region from klyston pass through the gaseous sample of the sample under investigation. The radiations pass along the rectangular wave guide tube to the quartz crystal detector, situated at the end of the wave tube.

The quartz crystal detector produce electrical signal, which is amplified by the amplifier and subsequently are diplaced either on an oscilloscopic screen or recoded on a chart. From the chart, it is possible to determine the frequency of the detected microwave radiation. From the data obtained it is possible to determine the intensity of transmitted beam, moment of inertia and intranuclear distances fairly accurately. Some data obtained by microwave spectroscopy are given in Tables 29.1 and 29.2.

Molecule	Bond	r(Å)	Bond	r(Å)
HCN	С—Н	1.06317	C—N	1.55535
CICN	C—CI	1.629	C—N	1.163
Br CN	C—Br	1.790	C—N	1.159
N NO	N—N	1.126	N—O	1.191
ocs	с—о	1.164	C—S	1.559
CH <sub>3</sub> CI	C—H	1.0959	C—CI	1.178
CH <sub>3</sub> F	С—Н	1.109	C—F	1.385
SiH <sub>3</sub> Br	Si—H	1.570	Si—H	2.209

Table 29.1 Molecular data obtained by Microwave spectroscopy

Molecule	Bond angle	Band angle
CHCI <sub>3</sub>	CI—C—CI	110°24′
CH₃CI	H—C—H	108° 0′
CH <sub>3</sub> F	H—C—H	110° 0′
SiH <sub>3</sub> Br	H—Si—H	110° 20′

**Table 29.2** Bond angles for some molecules obtained by microwave spectroscopy

### 29.7 APLICATIONS

Microwave spectroscopy is used for determination of structures of a number of molecules. Some typical examples are given below.

## 1. Xenon Oxyfluoride

On the basis of microwave spectrum, xenon oxyfluoride has a structure consistant with  $C_{4\nu}$  symmetry.

$$F = 0$$

$$F = 0$$

#### 2. Carbon Oxysulphide (OCS)

Carbon oxysulphide molecule is linear having two different interatomic distances. It is difficult to calculate two unknown in one equation. This problem has been overcome by using isotopic technique.

Let us first consider the isotopic species  $^{16}$ C  $^{12}$ O  $^{32}$ S. Assuming  $r_1, r_2, r_3$  are the distances of C, O and S from the centre of gravity, it follows that.

$$m_{C} r_{1} - m_{O} r_{2} = m_{3} r_{3} \qquad \dots (1)$$

$$O \qquad C \qquad G \qquad S$$

$$r_{2} \qquad r_{3} \qquad S$$

<sup>16</sup>O <sup>12</sup>C <sup>32</sup>S molecule

For this molecule, the moment of inertia is given by

$$I = m_C r_1^2 + m_O r_2^2 + m_3 r_3^2 \qquad ...(2)$$

It follows from equation (2) that

$$r_2 = r_{\rm CO} + r_1$$
 ...(3)

$$r_3 = r_{\rm cs} - r_1$$
 ...(4)

Where  $r_{\rm CO}$  and  $v_{\rm CS}$  are interatomic distances. Substituting equations (3) and (4) in equation (1), we get

$$(m_C + m_O + m_S)r_1 = m_S r_{CS} - m_O v_{CO}$$

or 
$$Mr_1 = m_S r_{CS} - m_O v_{CO}$$
 ...(5)

or 
$$r_1 = \frac{m_S r_{CS} - m_O r_{CO}}{M}$$
 ...(6)

Where

$$M = m_C + m_O + m_S$$

Substituting equations (3) and (4) in equation (2), we get

$$I = m_O (v_{CO} + r_1)^2 + m_C r_1^2 + m_S (r_{CS} - r_1)^2 \qquad ...(7)$$

Substituting the value of  $r_1$  from equation (5) in equation (7), we get

$$I = \frac{m_S r_{CS}^2 - m_O r_{CO}^2 + 2m_O m_S r_{CO} r_{CS} - m_S r_{CS}^2}{M} \qquad ...(8)$$

The values of  $r_{CO}$  and  $r_{CS}$  (in equation 8) are not known. For determining their values an expression (as in 8) for the moment of inertia (I) for another isotope  $^{16}O$   $^{12}C$   $^{34}S$  is derived.

$$I' = \frac{m'_{S}r_{CS}^{2} - m_{O}r_{CO}^{2} + 2m_{O}m'_{S}r_{CO}r_{CS} - m'_{S}r_{CS}^{2}}{M} \qquad ...(9)$$

It is assumed (in equation 9) that the isotopic substitution does not change the interatomic dirances,  $r_{CO}$  and  $r_{CS}$ .

The value of  $r_{CO}$  and  $r_{CS}$  are determined on solving equations (8) and (9) provided the values of I and I' are known, which are determined from the microwave spectra of  $^{16}O$   $^{12}C$   $^{32}S$  and  $^{16}O$   $^{12}C$   $^{34}S$ .

The interationic distances that are obtained from various isotopic species are given in Table 29.3.

Pairs of Isotopic molecules	r <sub>co</sub> (Å)	r <sub>cs</sub> (Å)
<sup>16</sup> O <sup>12</sup> C <sup>32</sup> S and <sup>16</sup> O <sup>12</sup> C <sup>34</sup> S	1.1647	1.5576
<sup>16</sup> O <sup>12</sup> C <sup>32</sup> S and <sup>16</sup> O <sup>13</sup> C <sup>32</sup> S	1.1629	1.5591
<sup>16</sup> O <sup>12</sup> C <sup>34</sup> S and <sup>16</sup> O <sup>13</sup> C <sup>34</sup> S	1.1625	1.5594
<sup>16</sup> O <sup>12</sup> C <sup>32</sup> S and <sup>18</sup> O <sup>12</sup> C <sup>32</sup> S	1.1552	1.5653

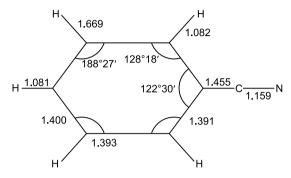
Table 29.3 Interatomic distances in OCS

It has been found (Table 3.13) that in isotopes interatomic distances differ depending on the changes in isotopes. This is attributed to zero point vibrations

which are persent at all temperatures. Since the amplitude of zero point vibrations are dependent on the mass of atoms, so bond lengths are also expected to change with change in isotopic substitution.

# 3. Benzonitrile (C<sub>6</sub>H<sub>5</sub>CN)

On the basis of microwave spectra benzonitrile has a deformed benzene ring and not to regular hexagonal structure.



## 4. Ammonia (NH<sub>3</sub>)

Bleaney and Townes found that in the minowave spectra of ammonia molecule each line is split into a doublet. This is attributed to the inversion of the molecule.

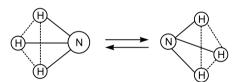


Fig. 29.4 Inversion of ammonia molecule

The microwave spectrum of ammonia is the result of motion of N atom between the centre of three H atoms (Fig. 29.4). The spectrum obtained (by plotting a graph between potential energy and position of N atom) is of double minimum type shown with a central barrier of 2070 cm<sup>-1</sup> (6 Kcal). Each minimum is observed as a curvature near 1000 cm<sup>-1</sup>, which is due to NH bending fundamental. There is a splitting of the lower rotational levels (Fig. 29.5), which is due to quantum mechanical tunnel effect due to finite barrier height and low mass of H atoms.

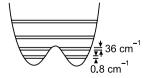


Fig. 29.5 Energy diagram showing inversion coordinate of ammonia

In fact, the inversion of ammonia is a vibrational motion. As this inversion is hindered, the interation results in energy difference in microwave region causing the splitting in rotational lines.

Unlike ammonia, phosphine  $(PH_3)$  and arsine  $(A_5H_3)$  do not show doublings (as in the case ammonia). This is because the inversion frequencies in these systems are slow due to increase in barrier height.

### 5. Barrier Heights of Some Molecules

With the help of microwave spectroscopy, it is possible to measure the barrier heights of certain molecules. In case a part of the molecule can rotate about a single bond, the internal potential energy will be dependent on the orientiation of this part with the rest of the molecule. Barrier height of certain molecules are given in Table 29.4.

Molecule	Barrier to rotation about single bond	
CH <sub>3</sub> OH (C—O)	1.07	
CH <sub>3</sub> OCH <sub>3</sub> (C—O)	2.72	
C <sub>6</sub> H <sub>5</sub> CH <sub>3</sub> (C—C)	13.94	
CH <sub>3</sub> NO <sub>2</sub> (C—N)	0.006	

**Table 29.4** Barrier Heights of certain molecule (Kcal Mol<sup>-1</sup>)

**6. Ozone:** Using microwave spectroscopy, it is possible to establish the triangular structure of ozone.



- **1.** What is microwave spectroscopy? Explain the difference between microwave spectroscopy and infrared spectroscopy?
- **2.** Explain the theory of microwave spectroscopy.
- **3.** Diatomic molecule is a rigid rotator. Explain.
- **4.** Discuss the selection rules for rotational spectra.
- **5.** Describe the various components of a microwave spectrometer and explain their functions.
- **6.** How many rotational lines are observed in HCl gas?
  - **Hint :** The number of rotational lives are determined by the selection rules.
- 7. In polyatomic molecules, assumption is made that bond length is not changed by isotopic substitution. How will you explain that the assumption is correct?

8. In case carbon monoxide the microwave spectrum consists of the following lines:

$$J = 0 \rightarrow J = 1 : 1.15 \times 10^5 \text{ Mz}$$
  
 $J = 1 \rightarrow J = 2 : 2.30 \times 10^5 \text{ Mz}$ 

What will be the bond distance of carbon monoxide?

[**Ans.** 
$$r = 0.1134 \text{ nm}$$
]

- 9. What do you understand by 'Inversion spectrum of ammonia'? Explain.
- 10. Write notes on:
  - (a) Barrier Height
  - (b) Structure of ozone
  - (c) Structure of benzonitrile.





# Chapter 30

# Nuclear Quadrupole Resonance (NQR) Spectroscopy

#### 30.1 INTRODUCTION

Nuclear quadrupole is a resonance phenomenon and is related to NMR. The unsymmetrical distribution of electric charge of a spinning nuclei is referred to as Nuclear quadrupole moment. All nuclei having a spin number  $1 \geq 1$  possess an electric quadrupole moment (eQ) in addition to the magnetic moment. The electric quadrupole moment (eQ) is measure of the deviation of the distribution of the positive charge in the nucleus from spherical symmetry. In the radio frequency region of the electromagnetic spectrum, transition between the two levels are observed. These transitions are commonly known as nuclear quadrupole resonance transitions. An appartus called nuclear quadrupole resonance spectrometer is used for the direct measurement of the frequencies in solid state. The nuclear quadrupole resonance spectrometer resembles nuclear magnetic resonance spectrometer though a magnet is not required.

Using this technique, isotopes of many elements including chlorine, bromine, iodine, nitrogen, arsenic, antimony, tin can be studied. However, as in the case of NMR, <sup>12</sup>C and <sup>16</sup>O cannot be studied.

#### 30.2 THEORY

The nuclei which have integral and half integral nuclear spins  $\left(I=1,\,\frac{3}{2},\,2,\,\frac{5}{2}\,\right)$  etc. have nuclear electric quadrupole moments. These interact which the electric field gradients which are created at the nuclei by the asymmetric distribution of charge which arises from the extranuclear electrons or the bonding electrons in the

molecule, of which the nuclei is a part. The NQR energy levels for the integral and half integral nuclear spins (*i.e.*,  $I \ge 1$ ) are given by the expression.

$$E_m = \frac{eQ.q}{4I(2I-1)} \left[ 3m_1^2 - I(I+1) \right] \qquad ...(i)$$

$$= A \Big[ 3m_1^2 - I(I+1) \Big] \qquad ...(ii)$$

where,

$$A = \frac{eQ.q}{4I(2I-1)}$$
 and  $m_1 = -I, -I+1, ...+I$ .

In the equation (i), Q (or e Q) represents the nuclear quadrupole, q is the electric field gradient (EFG) and I is the nuclear spin. The product (eQ.q) of the two quantities (eQ and q) is known as nuclear quadrupole coupling constant.

Another factor which determines the extent of splitting of the quadrupole energy level is the field gradient q. So the splitting is related to eQq. By suitable selection of the axes and defining the co-ordinates system, the components of the field gradient (q) along the x, y and z axes are labelled as  $q_{zz}$ ,  $q_{yy}$  and  $q_{xx}$ .

As a matter of convention  $q_{zz}$  is the largest field gradient and  $q_{xx}$  is the lowest field gradient, i.e.,  $q_{zz} > q_{yy} > q_{xx}$ . In case the three gradients  $(q_{zz}, q_{yy}, q_{xx})$  are equal, the field gradient is spherical and the quadrupole levels degenerate indicating that all orientations of the nucleus interact equally with the electronic charge. However, when  $q_{zz} \neq q_{xx} = q_{yy}$ , there is axial symmetry around the z-axis. In case of such molecules, the NQR spectra can be easily interpreted. In such cases  $q_{zz}$  is represented as q and the maximum field gradient is along the z-axis. As an example, the chlorine atom in  $Cl_2$  and in  $CH_3Cl$  are axially symmetrical and so the bond axis in  $Cl_2$  and C—Cl bond axis in  $CH_3Cl$  are the z-axis. In such molecules the energy of different quadrupole state are given by equation (i). This can be explained as follows:

For a nuclear spin  $I = \frac{3}{2}$ ,  $m_1$  can have values  $\frac{3}{2}$ ,  $\frac{1}{2}$ ,  $-\frac{1}{2}$ ,  $-\frac{3}{2}$ .

For  $m_1 = \frac{3}{2}$  the equation (i) gives

$$\frac{E_3}{2} = + \frac{eQ.q}{4}$$

Since in the above equation  $m_1$  is square term, so,  $m_1 = -\frac{3}{2}$  will also have identical energy.

We, thus have a doubly degenerate set of quadrupole energy.

In a similar way  $m = \pm \frac{1}{2}$  will also have a doubly degenerate set (Fig. 30.1).

The transition energy,  $\Delta E$  will correspond to

$$\frac{eQ.q}{4} - \frac{\left(-eQq\right)}{4} = \frac{eQ.q}{2}$$

Only a single transition is expected in an axially symmetric field. *eQq* expressed in energy units is directly calculated from the frequency of absorption.

$$eQq = 2\Delta E = 2 hv.$$

eQq is expressed as a frequency (mc sec<sup>-1</sup>).

In the above case, eQq will be twice the frequency of the NQR transition.

The above discussion can be extended and transitions for other nuclei can be calculated.

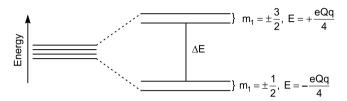


Fig. 30.1 Quadrupole energy levels

The mechanism by which radio frequency field results in transitions between the quadrupole energy level is similar to that in NMR. The major difference between NMR and NQR is that in NMR an external magnetic field is used to establish spacing of the Zeeman levels. However, in NQR the spacing is due to molecular or crystalline electric field. It is due to this reason that in NQR spectrometers magnet is not needed.

It has, however, been found that in NMR spectra of crystalline solids, quadrupole splitting may be observed in those cases, in which coupling constant in not too large, *i.e.*, when  $eQq_{zz}$  is approximately 1. In case the coupling constant has a value of 3 MHz to 500 MHz, accurate measurements are best made by direct NQR spectroscopy.

#### **30.3 NQR INSTRUMENT**

A good NQR spectrometer should have a high sensitivity and it should be possible to tune a wide range of frequencies. A Schematic representation of an NQR spectrometer is given in Fig. 30.2.

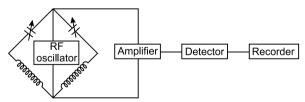


Fig. 30.2 Schematic representation of a NQR spectrometer

In the NQR spectrometer, RF oscillator supplies power to a balanced bridge circuit. On absorption of energy by the sample (in the inductive coil), an imbalance in caused in the circuit. The resultant voltage is recorded after amplification. The

spectras thus obtaind are called pure quadrupole spectras. These are obtained only for solids. In case of liquids and gases, the collisions between the molecules at normal pressure results in continuous change in the axis due to electric field gradient at the nucleus which is averaged to zero. Due to this limitation, NQR has less range of applications compared to NMR.

For obtaining a good NQR spectra, the crystalling solid must be cooled to 77K. The amount of the sample depends on the strength of resonance. Normally 50 mg of p-dichlorobenzene produces an observable <sup>35</sup>Cl resonance lines. However, for obtaining detectable <sup>14</sup>N line resonance lines need several grams of hexamethylene tetramine.

#### 30.4 APPLICATIONS OF NOR

(a) Interpretation of eQq Data: Normally it is difficult to interpret eQq values in terms of ionic character,  $\pi$  bonding and s and d hybridization. This can be seen clearly in compounds in which there are large differences in ionic character (Table 30.1).

Molecule	eQq <sub>mol</sub>	Molecule	eQq <sub>mol</sub>
Br <u>Cl</u>	-103.6	<u>Br</u> Cl	876.8
I <u>CI</u>	-82.5	Li <u>Br</u>	37.2
Ti <u>Cl</u>	-15.8	Na <u>Br</u>	58.0
K <u>Cl</u>	0.004	D <u>Br</u>	533.0
Rb <u>Cl</u>	0.774	F <u>Br</u>	1089.0
CsCl	3.0	D <u>I</u>	-1827.0
F <u>Cl</u>	-146.0	Na <u>l</u>	-259.87

**Table 30.1** Values of eQq for some halides

It is noted from the data presented in Table 30.1 that smaller  $eQq_{\mathrm{mol}}$  values are obtained from ionic compounds. The eQq values of a number of compounds are explained assuming that d hybridization is unimportant. Ionic character and s hybridization can be estimated.

It is found that electron releasing substitution give negative  $\sigma$  values and smaller eQq values. This is attributed to increased ionic character in the C—Cl bond making chlorine more negatively charged.

(b) Nature of Chemical Bond: Using NQR the electronic structure of molecules can be derived. Even the extent of hybridization and ionic character can be known by the comparison of the quadrupole coupling constant in molecular and atomic state of the same molecule. However, the exact nature of bonding is not possible to deduce from the NQR data due to involvement of a number of unknowns. For example, in case of  $H_2S$  [electronic configuration of sulphur is [Ne]  $3S^2$   $3p^4$ ; the HSH angle is nearly  $90^{\circ}$ ], it is assumed that pure *p*-orbitals are involved in the bonding. However, the results obtained from NQR yield a large symmetry parameter of n = 0.60 for sulphur. The electric field gradient is calculated from the following equations:

$$\begin{split} eQq_{xx} &= -\left(\frac{N_y + N_z}{2} - N_x\right) eQq_{\text{atom}} \\ eQq_{yy} &= -\left(\frac{n_x + N_z}{2} - N_y\right) eQq_{\text{atom}} \\ eQq_{zz} &= -\left(\frac{N_x + N_y}{2} - N_z\right) eQq_{\text{atom}} \end{split}$$

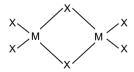
where,  $N_x$ ,  $N_y$  and  $N_z$  are the effective electron populations of  $p_x$ ,  $p_y$  and  $p_z$  orbitals of the atom.

η can be written as

$$\eta = \frac{q_{xx} - q_{yy}}{q_{zz}} = \frac{3(N_y - N_x)}{N_x + N_y - 2N_z} = 0.60$$

Supposing there is a lone pair in the  $p_z$  orbital, than  $N_z = 2$  and 1.33  $N_y = N_x + 1$ . But  $N_x$  cannot be equal to  $N_y$ . So there is no involvement of  $\pi$  orbital in the bonding. The large value of asymmetry parameter indicate the involvement of orbitals having approximately 15% d-character.

(c) Structures of Group III Holides: In group III halides of the type  $MX_3$  (e.g., AlBr<sub>3</sub>), the basic halogen spectrum gives three resonance lines, two being closely spaced and the third is far below. This shows that the three halogen atoms are not chemically equivalent. This has also been supported by x-ray data indicating that the halides exist as dimers having two types of halogen atoms, which correspond to the bridge end positions.



The small difference of one set of resonance frequencies is attributed to slight difference in the crystalline field around the two chemically equivalent set of atoms.

(d) Study of charge-transfer Compounds: Charge-transfer compounds are formed most probably by mixing equimolar amounts of CBr<sub>4</sub> and p-xylene. Similar compounds are also formed by CCl<sub>4</sub> and p-xylene, bromine and benzene. On the basis of studies on frozen solution of these mixtures, Hooper found that the

resource frequencies of the halogens are not different from those in pure molecular state. On this basis, it was concluded that these charge transfer compounds are not formed in the ground state.

(e) Study of Chloromethanes and Chloroacetyl Chlorides: In the above compounds, there is increase in the values of coupling constants for Cl<sup>35</sup> per chlorine atom to the extent of 3.5 MHz. The larger shift is attributed to an effective increase in electronegativity of the entity to which a chlorine atom in bonded due to in inductive effect.

**Table 30.2** Coupling constants in Chloromethanes and Chloromethyl chlorides [e/g. Q/h (MHz)]

Molecule	R = H	R=COCI
RCH <sub>2</sub> CI	68.0	60.8
RCH <sub>2</sub> CI RCHCl <sub>2</sub>	72.0	65.0
RCCI <sub>3</sub>	76.6	67.4
CCI <sub>4</sub>	81.2	_

As seen, in chloroacetyl chloride there is increase in coupling constant with the effective electronegativity of the group bonded to the carbonyl chloride. In non-carbonyl chlorides (R = H), the coupling constant have higher value of ( $\sim 10 \text{ MHz}$  higher) than that for carbonyl chlorides.

# EXERCISES 1

- 1. Discuss the principle underlying NQR spectroscopy.
- 2. What are the limitations of NQR?
- 3. Is it possible to use NQR for studying gases and liquids. Give reasons?

- 4. Describe the instrument for NOR.
- 5. How is NQR used for studying
  - (a) the nature of chemical bond
  - (b) group III halides
  - (c) charge transfer compounds.
- 7. What is electric field gradient? How does it arise in NQR?



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# Chapter 31

# **Raman Spectroscopy**

#### 31.1 INTRODUCTION

Most of the spectroscopy techniques involve absorption of light of a particular wavelength. However, Raman spectroscopy deals with scattering of light. Molecules like H<sub>2</sub>, N<sub>2</sub>, O<sub>2</sub> etc., do not possess permanent dipole moment and so do not show IR spectra. However, such molcules do show Raman spectra. This is because of the consequence of change in *polarisability* which causes change in the induced dipole moment at the vibrational frequency (*see* also section 31.4). In fact, using Raman spectroscopy, it is possible to examine the vibrational spectra of molecules which do not respond to IR absorption spectroscopy. Raman spectroscopy gives additional information related to molecular structure. This is because in Raman scattering, each line has a characteristic polarisation and it is this characteristic polarisation which provides information related to the molecular structure. The vibrations that are active in IR are inactive in Raman spectra and *vice versa*. In fact *Raman spectroscopy complements IR spectroscopy*.

Another advantage in Raman spectroscopy is that it uses ultraviolet or visible radiation rather than infrared radiation in IR spectroscopy. Raman spectroscopy also finds application for the investigation of polypeptides and proteins in aqueous medium.

#### 31.2 PRINCIPLE OF RAMAN SPECTROSCOPY

It was Rayleigh who observed that on irradiation of a substance with monochromatic light, the scattered light is in a direction perpendicular to the incident light (Fig. 31.1). This phenomenon is known as Rayleigh scattering.

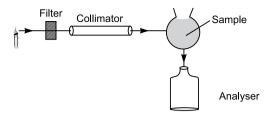


Fig. 31.1 Raleigh scattering

On the basis of theoretical considerations, Sametel (1923) predicted that a substance (solid, liquid or gaseous state) on irradiation with monochromatic light gives scattered light, containing frequencies different from that of the incident light. The Smekel's predictions were confirmed by Sir C.V. Raman (1928). Raman found that when a beam of monochromatic light is passed through a substance (solid, liquid or gaseous state), the scattered light was found to contain additional frequencies over and above that of incident frequency. This is known as **Raman effect**.

The wavelengths of lines in scattered light were modified in Raman effect. These lines were called **Raman lines** (*see* also section 31.3) These are of two types: stokes lines (having wavelengths greater than that of incident light) and Anti-stokes lines (have wavelengths shorter than that of incident light).

For his work on the scattering of light, and for the discovery of the effect named Raman effect, Sir C.V. Raman was awarded 1930 Nobel Prize in Physics.

The frequencies of the lines in the scattered light are called **Raman shifts**, designated as  $\Delta v$ . Thus, if  $v_i$  is the frequency of incident radiation and  $V_s$  in the frequency of scattered radiation, then the Raman shift,  $\Delta v$  in calculated as:

$$\Delta v = v_i - v_s$$

The Raman shifts are independent of the frequency of the incident light. Raman shift  $(\Delta v)$  is positive for stoke's lines and negative for Anti-stoke's lines [Fig. 31.2 (a) and (b)].

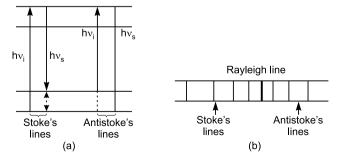


Fig. 31.2 Stokes and Anti-stoke's lines

Raman spectroscopy was not used for analytical work for about 30 years (1930-1960). Subsequently, with the discovery of 'Laser' (Light Amplification by Stimulated Emission of Radiation) by Town *et al.* (1950s), Raman spectroscopy became an important tool for analytical work.

The laser radiation is a radiation of high intensity in a narrow beam having well defined frequency and is coherent and monochromatic. It can be focussed easily and collimated. Most of the Raman spectrometers have built in laser source. The spectrum can be quickly recorded by using a Laser Raman spectrometer. The scientest, Charles H. Townes was awarded 1964 Nobel Prize in Physics for his work on Laser. The prize was shared with Nikolai Basov and Alexandor Prokhorov.

#### 31.3 CHARACTERISTICS OF RAMAN LINES

Some of the important characteristics of Raman Lines are given below:

- (i) The stoke's lines are invariably greater than the corresponding Antistoke's line. Due to this it is difficult to observe anti-stoke's line in conventional Raman spectroscopy.
- (ii) Raman Lines are displaced symmetrically about the parent lines. With rise in temperature, the separations from parent lines decreases.
- (iii) The frequency of the absorption band of a material is the difference in frequency between the modified and parent line.
- (iv) Raman shift, Δv, normally lies between the far and near infrared regions. This implies that the Raman frequencies are mostly identical with the infrared vibrational frequencies. It is, however, not necessary that all Raman lines will have the corresponding IR bands and vice-versa.

# 31.4 DIFFERENCES BETWEEN RAMAN SPECTRA AND INFRARED SPECTRA

It has already been stated that Raman spectroscopy complements IR spectroscopy (section 31.1). However, Raman spectra are quite different from IR spectra as given below:

Raman Spectra		IR spectra	
1.	Scrattering of light by vibrating molecules	Absorption of light is by vibrating molecules.	
2.	Ram spectra will be observed by the polarizability of the molecule	The criterion of IR spectra is the presence of permanent dipole in a molecule.	
3.	Spectra can be run in aqueous medium.	IR spectra cannot be run in aquous medium since $\rm H_2O$ is opaque is IR radiations.	

(Contd...)

Raman Spectra	IR spectra
The method is accurate but not very sensitive.	The method is both accurate and sensitive
The optical systems are of glass or quartz.	Optical systems are made of special crystals like CaF <sub>2</sub> Na Br etc.
In Raman effect, vibrational frequencies of large molecules can be measured.	Vibrational frequecies of very large molecules cannot be measured.
7. The source of incident radiation is mercury lamp or more recently laser.	The source of incident radiation is Nernst Glower (electrically heated filament of ${\rm ThO_2}$ and ${\rm CeO_2}$ ).

#### 31.5 POLARIZABILITY

It has already been stated (section 31.1) that diatomic molecules (like  $O_2$ ,  $N_2$ ,  $H_2$ ) show Raman spectra (and not IR spectra). The Raman spectra in diatomic molecules is the consequence of change in polarisability which causes change in the induced dipole moment at the vibrational frequency. In fact, it is the polarizability of a molecule which is responsible for Raman effect. A molecule, on subjecting to electromagnetic radiation will be polarized as the electric component of radiation force field will subject the protons and electrons to forces in opposite directions. The induced dipole moment,  $\mu$  is proportional to the strength of the electric field,  $\epsilon$ .

or 
$$\mu \propto \varepsilon$$
$$\mu = \alpha \varepsilon$$
$$\therefore \qquad \alpha = \frac{\mu}{c}.$$

The constant of this proportionality is called polarizability,  $\alpha$ . Polarizability, is, in fact, a measure of elastic deformation of the vibrational configuration of a molecule. In Raman effect, polarizability is a measure of the electron cloud by the electric field. Only when the polarizability of the molecule changes with the vibrational mode, the molecule will be Raman active.

#### 31.6 EXPLANATION OF MECHANISM OF RAMAN EFFECT

The mechanism of Raman effect is more complex than the IR absorption. Two theories have been proposed to explain the mechanism. These are Classical theory and the Quantum theory.

## 1. Classical Theory

This theory explains the existance of Raman effect. According to this theory, an application of an electric field to a molecule displaces its electrons and nuclei,

thereby producing an induced dipole in the molecule and the molecule is said to be polarized. If E is the strength of the field and  $\mu$  is the magnitude of the induced dipole moment then we say that:

$$\mu = aE \qquad ...(i)$$

where a is the polarizability of the molecule (see also section 31.5).

Subjecting a simple molecule (which can be polarized) to a beam of radiation of frequency v, the electric field experienced by each molecule changes according to the following equation:

$$E = E_0 \sin 2\pi vt \qquad ...(ii)$$

Thus, the induced dipole undergoes oscillation of frequency v. From equations (*i*) and (*ii*) we can say.

$$\mu = aE = aE_0 \sin 2\pi vt \qquad ...(iii)$$

An oscillating dipole emits radiation of frequency, v. The above expression (iii) explains Raleigh scattering. For deriving expression (iii), vibration and rotation of molecules have not been taken in account.

It has been found that the Raman shift is equal to twice the frequency of rotation of molecule.

#### 2. Quantum Theory

Quantum theory is useful to predict correctly the selection rules and intensities in Raman spectra. According to quantum theory, Raman effect is regarded as the result of collisions between the light photons and molecules of the substance.

Supposing that a molecule of mass m in the energy state  $E_p$  moving with a velocity V and colliding with a light photon hv undergoes a change in its energy state and its velocity. After suffering a collision, let the new energy state is  $E_q$  and velocity is V'. Applying the principle of conversion of energy, we write

$$E_p + \frac{1}{2}mv^2 + hv = E_q + \frac{1}{2}mv'^2 + hv'$$

It has been proved that the change in velocity of the molecule is negligible. So, the above equation can be written as

$$E_p + hv = E_q + hv' \qquad \dots(i)$$

or 
$$V' = V + \frac{E_p - E_q}{h} \qquad ...(ii)$$
 or 
$$V' = V + \Delta V$$

Consideration of the equation (ii), there may be three situations.

(a) If  $E_p=E_q$ , the difference in frequency (Raman shift),  $\Delta V$  is zero  $\left[i.e., \frac{E_p-E_q}{h}=\text{zero}\right]$ . It implies than V'=V. This refers to unmodified line and so the molecule only deflects the photon. This collision is elastic and is similar to **Rayleigh Scattering**.

- (b) It  $E_p < E_q$ , then V' < V and this corresponds to Stoke's lines. In this case, the molecule has absorbed some energy from the photon and so the scattered photon will have lower energy.
- (c) If  $E_p > E_q$  then V' > V and this corresponds to anti-stoke's lines. In this case, the molecule was earlier in the excited state and it gave some of its intrisnic energy to the incident photon. So the scattered photon will have greater energy.

The change in intrinsic energy of a molecule is dependent on the quantum rules.

$$E_p - E_q = \pm nhv_c$$

where  $v_c$  is the characteristic frequency of the molecule and n = 1, 2, 3... etc.

The above equation, when n = 1 (in simplist molecule) is written as

$$V' = V \pm V_c$$

It follows from the above equation that difference in frequency (V - V') between the incident and scattered photon in the Raman effect will correspond to the characteristic frequency  $v_c$  of the molecule.

The Raman scattering and Rayleigh scrattering are depicted in Fig. 31.3.

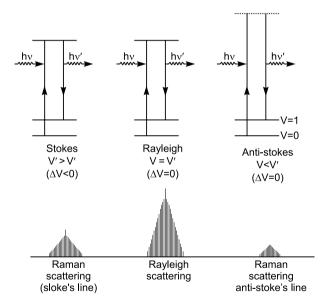


Fig. 31.3 Raman and Rayleigh scattering

Equation (i) can be written as

$$E_n - E_a = h(v - v') = h\Delta v_{\text{Raman}} = h_c \Delta \bar{v}$$

The shifts in frequency (V - V') are called **Raman shifts**. These fall in the range  $100 - 4000 \text{ cm}^{-1}$  for vibrational energy changes.

Since 
$$\overline{v} = \frac{1}{\lambda}$$
,  

$$\therefore \qquad \Delta \overline{v} \text{ (in cm}^{-1}\text{)} = \frac{10^8}{\lambda (\text{in Å})}$$

So Raman shift (in cm<sup>-1</sup>) is given by

$$\Delta \overline{\nu} = \frac{10^8}{\lambda_{ex}(\text{in Å})} - \frac{10^8}{\lambda_{Raman}(\text{in Å})}$$

where  $\lambda_{ex}$  is the wavelength corresponding to the exciting (incident) frequency.

#### 31.7 RAMAN SPECTROMETER

In Raman Spectrometer a sample is irradiated with monochromatic light. The scattered light is observed at right angles to the incident radiation. The instrument consists of the following components:

- (i) Light Source: Since the Raman effect is weak, it is necessary to use a light source of high intensity. Most of the instruments use a mercury arc as a light source. Using suitable filters, light of single wavelength, 4358 Å is obtained and used in Raman spectroscopy. For using such a light source, the sample must be colourless and concentrated solution must be used. These disadvantages could be overcome by using a laser source (helium-neon). The modern Raman spectrometers have inbuilt laser source. No filters are necessary when lasers are used since the energy output is in the form of monochromatic narrow parallel beam. Also the resolution is much better than that of mercury excited line. Also coloured solutions can be examined without causing electronic transition. The laser tube contains 7:1 mixture of helium and neon gas for optimum output of 6328 Å laser line.
- (ii) Filters: Filters are necessary if mercury are is used as a light source. The filters are made of nickel oxide glass or quartz glass. At times a coloured solution like an aqueous of iodine in carbon tetrachloride or ferricyanide may be used as a monochromator. It has already been mentioned that in laser fitted Raman spectrometers, no filters are necessary.
- (iii) Sample Holder: The type of sample holder to be used depends on the intensity of source. For gaseous samples, the sample holder should be bigger in size than for liquids. Solids are dissolved in a suitable solvent. Any solvent which is suitable for UV spectra can be used. A good solvent is water. It is particularly used for the study of inorganic compounds.
- (*iv*) **Detectors:** Ordinary Raman spectrometer is used to photograph the Raman spectrum. Raman lines can be detected by two different ways.

An easy way consist in gathering the scattered light emerging through a glass window at the end of the sample tube and passed through a grating or prism and then focussed on to a photographic plate. Subsequently the plate is developed and both the frequencies and intensities of lines can be measured using external equipment. The other method is photographic method of recording the Raman spectrum. Raman spectrometers are now have photomultiplier tubes for direct measurement and automatic scanning the spectrum. The spectrum from the monochrometer is passed through a slit which permits a narrow wavelength region to pass through and focussed on to a photomultiplier type detector. The photomultipier type detector uses an amplifier and a recorder to record directly the Raman spectrum.

A diagramatic representation is a typical Raman spectrometer is shown in Fig. 31.4.

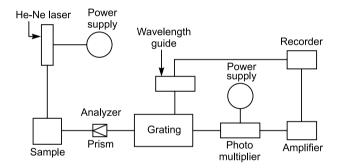


Fig. 31.4 Diagrammatic Representation on a typical Raman spectrometer

### 31.8 INTENSITY OF RAMAN PEAKS

The intensity of Raman peaks is dependent on the polarizability of the molecule, intensity of the source and concentration the active group.

#### 31.9 APPLICATIONS OF RAMAN SPECTROSCOPY

Raman spectras can be conveniently obtained from milligrams or micrograms samples by using laser as the light source in Raman spectrometers. Spectras of solids (powders, crystals, polymers and coloured substances), liquids or gases can be recorded. Even aquous solutions of corrosive and air-sensitive substances can be used. The only requirement is that the substance must be pure. Liquids can be purified by vacuum distillation or gas chromatography. Solvents must be pure (spectrographic grade).

Raman spectroscopy has been used in a Large number of cases for the identification of compounds, structure elucidation and study of complexes and ionic equilibria in solution. This technique finds application in inorganic, organic and physical chemistry. In fact, it is also used for quantitative analysis and polymer chemistry. Some of the important applications are given below:

## (a) Application in Inorganic Chemistry

(i) Structure of CO<sub>2</sub>: Carbon dioxide is believed to be a symmetrical linear molecule O—C—O. This finds support in the fact that two fundamental lines (V<sub>2</sub> and V<sub>3</sub>) in infrared and one (V<sub>1</sub>) in Raman spectra. This has been confirmed experimentally.

IR spectra 667 cm<sup>-1</sup> ( $V_2$ ) and 2349 cm<sup>-1</sup> ( $V_3$ ) Raman spectra 1340 cm<sup>-1</sup> ( $V_1$ ).

In this case both IR and Raman spectra (which are complementary to each other) are used.

- (ii) Structure of N<sub>2</sub>O: In case N<sub>2</sub>O a symmetrical molecules i.e., N—O—N (like CO<sub>2</sub>) it should show a behaviour similar to CO<sub>2</sub>. However if N<sub>2</sub>O is unsymmetrical, N—N—O, V<sub>1</sub> should be active both in IR and Raman spectra. This additional fundamental line has been observed both in IR and Raman spectra. Thus, N<sub>2</sub>O has been found to be unsymmetrical, i.e., its structure is N—N—O.
- (iii) Structure of  $SO_2$ : Sulphur dioxide shows three vibrations (at 1150, 520 and 1300 cm<sup>-1</sup>) corresponding to  $V_1$ ,  $V_2$  and  $V_3$  respectively in both IR and Raman spectra. This implies that the molecule lacks symmetry. Thus,  $SO_2$  has a bent structure.
- (iv) Structure of H<sub>2</sub>O: Like SO<sub>2</sub>, water is a non-linear triatomic molecule. The three fundamental vibrations are active both in IR and Raman. The H<sub>2</sub>O molecule also has a bent structure like SO<sub>2</sub>.
- (v) Chloro Complexes of Mercury: Raman spectra of a solution of mercuric chloride and ammonium chloride (in molar ratio 1:2) shows a strong line at 269 cm<sup>-1</sup>, which compares with the strong line at 273 cm<sup>-1</sup> in the Raman spectra of solid ammonium tetrachloro mercurate (II). This indicates the formation of HgCl<sub>4</sub><sup>2-</sup> in solution.
  In a similar way Raman spectra of 1:1 and 1:2 reaction products of mercuric chloride and potassium chloride shows the formation of complexes HgCl<sub>4</sub> and HgCl<sub>4</sub><sup>2-</sup> respectively.
- (vi) Nature of Bonding: In the IR spectra of tetrahedral complexes ML<sub>4</sub> (e.g., ZnCl<sub>4</sub><sup>2-</sup>, CdCl<sub>4</sub><sup>2-</sup> and (HgCl<sub>4</sub><sup>2-</sup>) and octahydral complexes ML<sub>6</sub> (e.g.,) SiF<sub>6</sub><sup>2-</sup> and SF<sub>6</sub>) no bands are observed. This is because during vibration only the bond lengths change. However, with IR spectras of

the above complexes ( $ML_4$  and  $ML_6$ ), intense lines are obtained. It is possible to get useful information about the strength of metal-ligand bond from the M–L bond stretching force constant. The ions like  $PO_4^{3-}$ ,  $SO_4^{2-}$  and  $ClO_4^-$  have been found to have large force constants. This proves that there is a  $d\pi - p\pi$  bonding between the central atom and oxygen atom in addition to  $\sigma$ -bonding.

(vii) Hydrogen Cyanide: On the basis of Raman Spectra of hydrogen cyanide (two bonds at 2062 cm<sup>-1</sup> and 2094 cm<sup>-1</sup>) it has been shown that the two isomers are in a dynamic equilibrium

$$HC \equiv N \longleftrightarrow \overline{C} \equiv \overset{+}{N} - H$$

(viii) Carbon Disulfide: For carbon disulphide all the vibration are both IR and Raman active. On the basis of Raman spectra it is concluded that CS<sub>2</sub> has a centre of symmetry.

## (b) Applications in Physical Chemistry

(i) Ionic Equilibria in Solution

The dissociation of nitric acid is represented as

$$HNO_3 + H_2O \Longrightarrow H_3O^+ + NO_3^-$$

The Raman spectra of anhydrous HNO<sub>3</sub> shows a number of lines (at about 1400 cm<sup>-1</sup>), which are characteristic of —NO<sub>2</sub> group. However, on dilution this band fades away gradually and a new band near 1046 cm<sup>-1</sup> (characteristic NO<sub>3</sub> ions) appears. This change on dilution is due to ionization. It is possible to obtain dissociation constant of HNO<sub>3</sub> by monitering the intensity of NO<sub>3</sub> and nitric acid in the Raman spectrum.

The dissociation of H<sub>2</sub>SO<sub>4</sub> is represented as

$$H_2SO_4 + H_2O \Longrightarrow H_3O^+ + HSO_4^-$$
  
 $HSO_4^- + H_2O \Longrightarrow H_3O^+ + SO_4^{-2}$ 

In Raman spectra of  $\rm H_2SO_4$  (more than 25% concentration) Raman line appear at 1050 cm<sup>-1</sup>, attributed to the presence of  $\rm HSO_4^-$ . However, on dilution this line gradually fades and a Raman line characteristic of  $\rm SO_4^{2-}$  appear. Thus, the ionization  $\rm H_2SO_4$  in two steps (as shown above) is confirmed. The concentration is  $\rm SO_4^{2-}$  is small except in very dilute solution of  $\rm H_2SO_4$ . The dissociation constant of  $\rm H_2SO_4$  can be calculated from the intensity of  $\rm HSO_4^-$ ,  $\rm SO_2^{2-}$  ions and  $\rm H_2SO_4$ .

- (ii) Study of Single Crystals: Raman spetroscopy is valuable in the study of single crystals. It is well known that *x*—ray diffration has been used for structure analysis. However, Raman procedure is preferred when sufficient qualitative information is available, since it is faster and comparatively much cheaper.
- (iii) Study of Degree of hydrolysis of a Salt: Using Raman spectroscopy, the degree of hydrolysis can be calculated by measuring the relative intensities of lines characteristic of the base, the acid or the salt.

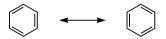
### (c) Applications in Organic Chemistry

Raman spectra is helpful to determine the molecular structure of organic compounds. It has been found that each group has its own characteristic frequency. The Raman lines generally occur in the region 500–3500 cm<sup>-1</sup> Raman frequencies of some functional groups are given in the following Table (31.1)

Group	Wave number cm⁻¹	Group	Wave number cm <sup>-1</sup>
C—H (aromatic)	3050	S—H	2570
C—H (aliphatic)	2920-2970	О—Н	3400
C=O (anhydrides)	1570	C—CI	650–710
C=O (ketones)	1610	C—Br C—I	600–570 500–530
C=O	1720	C≡N	2150
(aldehydes)		с—с	1000
C=O (acids)	1650	C≕C	1900-2400

Table 31.1 Raman frequencies of some functional groups

(i) Benzene: Raman spectra of benzene give two strong Raman lines at 995 cm<sup>-1</sup> (C—C) and 1050 cm<sup>-1</sup> (C—C). This gives support to the Kekula formula.



(ii) Differentiation between o-, m- and p-disubstituted Benzene Derivatives: m-Substituted benzenes give an strongly polarised line at 995 cm<sup>-1</sup>, which is not present in the ortho and para isomers.
 o- and p-Compounds can be distinguished by the presence of a line at

about 625 cm<sup>-1</sup> in the para isomer, which is absent in the ortho isomer.

- (iii) Differentiation between Cis and Trans Isomers: As an example dichloroethylene, C<sub>2</sub>H<sub>2</sub>Cl<sub>2</sub> is present in cis and trans configurations in equilibrium. The trans compound has a centre of symmetry. Coincident frequencies observed in IR and Raman spectra is assigned to cis configuration. Thus the analysis of the mixture of cis and trans-isomers can be done.
- (iv) Olefinic Functional Group: The Raman spectra gives much more useful information about the olefinic functional group than the IR spectra. Thus, as an example C=C stratching vibrations for Olefins is weak and undetected in IR absorption in many cases. However, Raman band is very intensive (1600 cm<sup>-1</sup>). Its position is dependent on the nature of the substitutents and their geometry.
- (v) Ring Size in Cycloalkanes: Raman spectroscopy is useful in the determination of ring size in paraffins. As an example, cycloalkanes give a characteristic Raman peak in the region 700–1200 cm<sup>-1</sup>. The position of the peak decreases continuously from 1190 cm<sup>-1</sup> for cyclopropane to 700 cm<sup>-1</sup> for cyclo-octane. The IR vibrations are weak or do not exist in cycloalkanes.
- (d) Study of Polymers: Raman spectroscopy is helpful in the characterisation of polymers. Physical properties such as arrangement of molecules with respect to each other, crystallinity, tacticity and amorphous character can be determined. These are useful for determining the strength and durability of polymers. The usual IR spectroscopy is not helpful in this respect, since the polymer sample has to be dissolved in a suitable solvent in which the original character of the polymer is changed.

The Raman spectra of polymers exhibit simultaneously properties of crystals and liquids. This is due to the existance of both amorphous and crystalline regions in polymers. Ram spectroscopy permits a number of conclusions to be reached about the structural units of polymers *i.e.*, C = C bonds ( $1660 \text{ cm}^{-1}$ ), benzene nuclei ( $1000, 1600, 3060 \text{ cm}^{-1}$ ) symmetric Si-O-Si vibrations ( $490 \text{ cm}^{-1}$ ) in silicons and C—S and S—S vibrations ( $650 \text{ and } 510 \text{ cm}^{-1}$ ) in polysulfides. All these peaks are intense in Raman spectra and are weak or absent in IR spectra.

(e) Quantitative Analysis: It is difficult to determine the absolute intensities of Raman lines. However, the intensity of the peaks is measured by reference to an reference line, which is arbitarily chosen (line of CCl<sub>4</sub> at 459 cm<sup>-1</sup>). The reference line is scanned before and after the spectra of the sample. Peak heights or scattering intensities are converted into scattering coefficients as follows:

 $\label{eq:Scattering coefficient} \begin{aligned} \text{Scattering coefficient} &= \frac{\text{Height of the sample peak}}{\text{Average height of the dual trace of}} \\ &\quad \text{CCl}_4 \text{ peak (before and after the spectra of the samples)} \end{aligned}$ 

For recording the height of the sample peak and average height of the dual trace of CCl<sub>4</sub> peak are recorded in cells of same dimensions.

For quantitative analysis the scattering coefficient is directly proportional to the intensity of Rman lines. For a mixture containing components of the same molecular type, the scattering coefficient is proportional to the volume fraction of the compound present. However, if a mixture has components of disimilar type, Raman shifts vary among various compounds resulting is a broad band. In such cases, the area under the peak is used as a measure of scattering intensity.

Liner relationship of the intensity vs. concentration in Raman procedure makes the identification of the major components of a mixture easier.



- 1. Explain why molecules like H<sub>2</sub>, N<sub>2</sub>, O<sub>2</sub> etc. show Raman spectra but these are IR inactive.
- 2. Raman spectroscopy complements IR spectroscopy. Comment.
- 3. What principles are involved in Raman spectroscopy?
- **4.** What are Raman lines and Raman shifts? Discuss the characteristics of Raman lines.
- **5.** What are the differences between Raman spectra and IR spectra?
- **6.** Explain the mechanism of Raman effect.
- 7. Describe the various components in a Raman spectrometer giving their functions.
- **8.** Give some important applications of Raman spectroscopy.
- **9.** How will you show that
  - (i) Carbon dioxide is a symmetrical linear molecule
  - (ii) N<sub>2</sub>O molecule is unsymmetrical
  - (iii) Water and SO<sub>2</sub> have bent structures
  - (iv) In hydrogen cyanide, the two isomers are in dynamic equilibrium.

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- 10. How will you determine the size of the ring in cycloalkane?
- **11.** What are the advantages of using laser as a source of light in Raman spectrometer?
- 12. What is the requirement for a compound to be Raman active?
- **13.** Explain why Raman stoke's lines are more intense than the Anti-stoke's lines.
- 14. What is Raman effect? Explain.





# Chapter 32

# **Mossbauer Spectroscopy**

## 32.1 INTRODUCTION

Mossbauer spectroscopy also known as nuclear gamma-ray resonance spectroscopy involves nuclear transition resulting from the absorption of  $\gamma$ -rays by the sample. This technique finds applications in a number of diverse fields like solid state physics, metallurgy, chemistry and biochemistry. Using this procedure, it is possible to estimate iron or tin content in ores, alloys etc. in as low as 0.03 per cent in a much shorter time (about 10 minutes). This technique has also been used to detect percentage of charged states of the same atom, *e.g.*, Fe<sup>2+</sup> and Fe<sup>3+</sup> in a material. This information is difficult to obtain from any other technique.

Mossbauer spectroscopy is based on Mossbauer effect.

## 32.2 MOSSBAUER EFFECT

It is a phenomenon relating to gamma ray spectrum which has important applications. Emission of a gamma ray photon by an atom causes the atom to recoil. This reduces the energy of the emitted gamma ray by an amount equal to recoil energy. It was Mossbauer who discovered in 1957 that if the emitting atom is strongly embedded in solid lattice, recoil cannot occur and the recoil energy is distributed in the lattice. Thus, the energy of the emitted gamma ray photon is not reduced. This makes it possible to produce gamma rays with predetermined wavelength and carry out precise measurements of many types. This effect has been used to study molecular structure and many other problems in nuclear and solid state physics. For his work on resonance absorption of gamma radiation and the discovery of Mossbauer effect, Mossbauer, won 1961 Nobel Prize in Physics, which was shared by Hofstadter.

On the basis of the above, it can be said that Mossbauer effect is based on recoil-free gamm ray resonance absorption phenomena. The main feature is the production of monochromatic radiation, which can be used in resolving minute energy perturbations. In fact, the application of Mossbauer-effect is due to its ability to detect small variations in the energy of interaction of nucleus with extra-nuclear electrons, which was not possible to observe before the discovery of Mossbauer effect.

# 32.3 MOSSBAUER SPECTROMETER

A Mossbauer spectrometer consists of a radioactive source and an absorber, both having atoms of the same element. The source emits recoil free gamma rays, which are absorbed by atoms of the same element in the absorber. In case the atoms in source and absorber are different, the emission and absorption spectra is slightly shifted. For getting resonance fluorescence, the source is mounted on a suitable linear velocity drive. If it moves away from the absorber, the frequency will undergo Doppler shift. When the Doppler shift becomes equal to the shift due to chemical environment, resonance fluorence absorption takes place. A gamma ray counter measures the intensity of the gamma rays emitted by the absorber (Fig. 32.1).

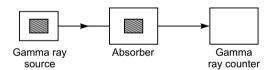


Fig. 32.1 Diagrammatic representation of a Mossbauer spectrometer showing various components

A plot of the count rate as function of the velocity of the source is shown in Fig. 32.2.

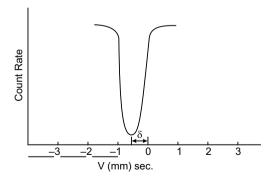


Fig. 32.2 Isomer shift of the Mossbauer line

The isomer shift is the shift of the resonance curve from zero velocity and is denoted by  $\delta$ . This shift provides important information about the nature of the bonding in metals.

In some cases, as in the case of  $^{57}$ Fe, two lines are obtained in place of a single line (Fig. 32.3). This is attributed to an effect called **nuclear quadrupole interaction.** In this case the separation between the two peaks is denoted by  $\Delta E_Q$  and is called **quadrupole spitting**. This effect is attributed to the departure of the nuclear environment from cubic symmetry and so gives valuable information about the local crystal structure.

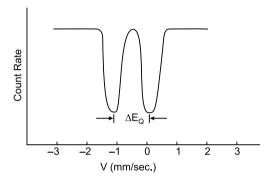


Fig. 32.3 Quadrupole splitting

It is possible that the source is paramagnetic and the absorber is in magnetically ordered phase. Thus, as an example <sup>57</sup>Fe in stainless steel is paramagnetic whereas in carbon steel it is in a ferromagnetic state. A spectrum of six fingers (Fig. 32.4) is obtained if the source is stainless steel and the absorber of carbon steel.

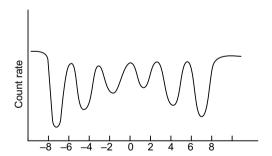


Fig. 32.4 Spectrum of six fingers using source of stainless steel and observer of carbon steel

The spectrum of six fingres of <sup>57</sup>Fe (Fig. 32.4) is due to Zeeman splitting. It provides valuable information about the magnetic state of the atom.

# 32.4 NUCLIDES AND THEIR CHARACTERISTICS

In Mossbauer spectroscopy the commonly used isotopes are <sup>57</sup>Fe, <sup>119</sup>Sn, <sup>151</sup>Lu, <sup>119</sup>Sb and <sup>119</sup>In. A nucleide must have the following characteristics:

- (i) The  $\gamma$ -radiation should have energy of 10–200 KeV.
- (ii) The nuclide should have a resonable half life (about one year) so that constant activity is available for a series of experiments.

Table 32.1 gives the important Nuclides used is Mossbauer spectroscopy.

Nuclei (Stable)	E <sub>r</sub> K <sub>e</sub> V	T <sub>½</sub> sec	E <sub>R</sub> for free nucleus
<sup>57</sup> Fe	14.4	1.4 × 10 <sup>-7</sup>	0.19 × 10 <sup>-2</sup> eV
<sup>67</sup> Zn	93.0	9.4 × 10 <sup>-6</sup>	6.90 × 10 <sup>-2</sup> eV
<sup>61</sup> Ni	71.0	5.2 × 10 <sup>-9</sup>	4.30 × 10 <sup>-2</sup> eV
<sup>119</sup> Sn	24.0	19 × 10 <sup>−8</sup>	0.26 × 10 <sup>-2</sup> eV

Table 32.1 Some important Nuclides and their Characteristics

Schematic diagram of a typical Mossbauer spectrometer is shown in Fig. 32.5.

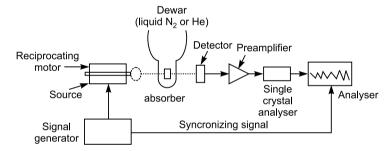


Fig. 32.5 Diagrammatic representation of a typical Mossbauer spectrometer

A number of isotopes, which can be used for the study of Mossbauer effect are produced artificatilly. These include <sup>57</sup>Fe, <sup>67</sup>Zn and <sup>119</sup>Sn.

# 32.5 APPLICATIONS MOSSBAUER SPECTROSCOPY

As already stated (Section 32.1) Mossbauer spectroscopy has been used to determine the presence of iron and tin in ores, alloys and industrial wastes. The presence of small amounts of iron in iron rust and absestos. Also the concentration of different charged states of the same atom, e.g.,  $Fe^{2+}$  and  $Fe^{3+}$  in materials like  $Fe_3O_4$  can be estimated, since these states have difference in isomer shift, quadrupole splitting and nuclear zeeman splitting.

The inhomogeneity in alloys can also be studied. The mechanism of corrosion, thermal decomposition processes etc. can also be studied. Information about the

electronic state and covalent bonding can be obtained from the isomer shift and quadrupole splitting data.

Some other interesting applications are given below:

### 1. Nature of Chemical bond

The Mossbauer spectra of iron ( $^{57}$ Fe) complexes like Na<sub>4</sub> [Fe (CN)<sub>6</sub>] and Na<sub>2</sub>[Fe(CN)<sub>5</sub>).NO]. 2H<sub>2</sub>O has been studied. It has been found that in complex, Na<sub>4</sub>[Fe(CN)<sub>6</sub>], iron is present in octahedral symmetry giving single line spectrum (Fig. 32.6). However, is the complex, Na<sub>2</sub>[Fe(CN)<sub>5</sub>.NO].2H<sub>2</sub>O, the distorted configuration splits the line to a well developed doublet, which is attributed to extensive  $\pi$  bonding in Fe—N—O linkage (Fig. 32.7).

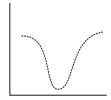


Fig. 32.6 Mossbauer spectra of Na<sub>4</sub>[Fe(CN)<sub>6</sub>]

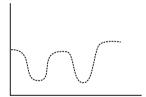


Fig. 32.7 Mossbauer spectra of Na<sub>2</sub> [Fe(CN)<sub>5</sub>.NO]2H<sub>2</sub>O

On the basis of Mossbauer spectra, the  $\pi$ -bonding capacities of some ligands are found to decrease in the order:

$$\mathrm{NO^{+}\! >\! CO \! >\! CN^{-}\! >\! SO_{3}^{2-}\! >\! PPh_{3}\! >\! NO_{2}^{-}\! >\! NH_{3}}$$

The Mossbauer spectra of  $Fe(CN)_6^{4-}$  ion and of  $[Fe(CN)_5 NO_2]^{2-}$  ion (showing quadrupole splitting) is given in Figs. 32.8 and 32.9 respectively.

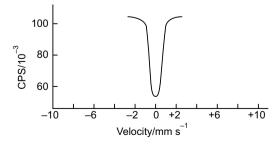


Fig. 32.8 Mossbauer spectra of  $Fe(CN)_6^{4-}$  ion



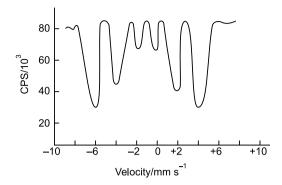


Fig. 32.9 Mossbauer spectra of [Fe(CN)<sub>5</sub>NO]<sup>2-</sup> ion showing quadrupole splitting

## 2. Chemical Shifts

The electron density at the nucleus affects the magnitude of the chemical shift. It is known the electron density at the nucleus of a atom is zero in case of *p* and *d*- orbitals. So, it can be said that *s*-orbital density is important.

The finding that the chemical shifts is helpful for the measurement of the relative *s*-electron density, which gives the bond character of an atom or ion attached to the Mossbauer nucleus. The chemical shifts in case of tin compounds using Mossbauer isotope <sup>119</sup>Sn are given Table 32.2.

Valence State Electronic Configuration		Chemical Shift (mm s <sup>-1</sup> )
Sn <sup>4+</sup>	5s° 5 p°	0
Sn(4-covalent)	5sp°	2.1
Sn <sup>2+</sup>	5s <sup>2</sup> 5p°	3.7

Table 32.2 Chemical shifts of some <sup>119</sup>Sn compound

The outermost electronic configuration of Sn is  $5s^2$   $5p^2$ . This implies that the 4+ state has no outer s-elections. However, in the +2 state, the higher  $5p^2$  electrons are removal leaving only two s. electrons. And in the case of 4-covalent state there is only s-electron. The chemical shifts (Table 32.2) support the linear structures. It is thus easier to find the valence state of an unknown tin compound from its Mossbauer spectra.

# 3. Quadrupole Effects

It has been found that majority of the Mossbauer nuclei have half integral spins and that the spin of the excited state is different from that in the ground state. On this basis, it can be said that both the nuclear states have spin more than 1/2. This implies that one or both are expected to have a quadrupole moment, which will interact with the electric field gradients in the vicinity. Examples of excited

state nucleus to have I = 1/2 and ground state I = 1/2 include <sup>57</sup>Fe, <sup>119</sup>Sn and <sup>129</sup>Xe. In such cases, the Mossbauer spectra gives useful data on the magnitude of crystal fields.

## 4. Determination of Structure

On the basis of Mossbauer spectra, certain structures can be easily ruled out. As as example, consider the Mossbauer spectra of iron dedecacarbonyl,  $\text{Fe}_3(\text{CO})_{12}$  (Fig. 32.10).

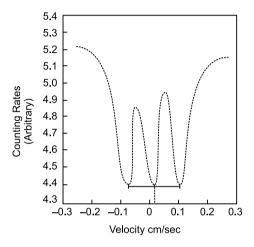


Fig. 32.10 Mossbauer spectra of  $Fe_3$  (CO)<sub>12</sub>

As seen, the Mossbauer spectra (Fig. 32.10) consists of three lines. There are three possible structures of the compounds (structures a, b and c in Fig. 32.11).

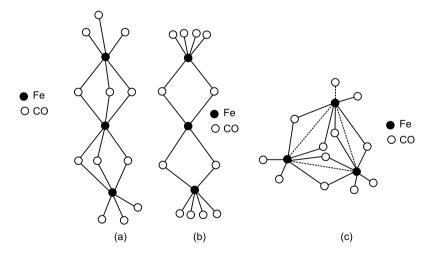


Fig. 32.11 Structure of Fe<sub>3</sub>(CO)<sub>4</sub>

The structure(C) is not possible as three equivalent atoms can give a maximum of two line spectrum (in the absence of magnetic splitting). In structures (a) and (b) only two of the three iron atoms are equivalent. Therefore, it is expected that the spectrum should be a single line with a quadrupole splitting into two. So the most probable structure for  $\operatorname{Fe_3(CO)_{12}}$  is (a) since a almost octahedral arrangement is expected to give a single line spectrum. On the other hand structure (b) will give a four line spectrum due to quadrupole splitting at two different iron atoms.

An interesting Mossbauer spectra of six sample of dust and rocks from moon are given in Fig. 32.12. It is found that varying proportion of pyroxene (an iron bearing silicate mineral) and FeTiO<sub>3</sub>.

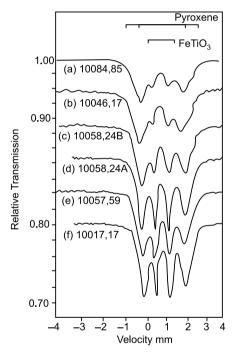


Fig. 32.12 Mossbauer spectra of six samples of dust and rocks from moon showing varying proportions of pyroxene (an iron bearing slicate mineral) and FeTiO<sub>3</sub>



- 1. Explain Mossbauer spectroscopy.
- 2. What is Mossbauer effect?
- **3.** On what conditions Mossbauer effect is likely to occur?
- **4.** What is recoil emission?
- **5.** Discuss the important applications Mossbauer spectroscopy.

- **6.** What are Mossbauer nucleides? Discuss the essential characteristics which a nuclide should have in order to exhibit Mossbauer effect.
- **7.** Give an account of some important nuclides which are used in Mossbauer spectroscopy.
- **8.** What do you understand by chemical shift?
- 9. What is isomer shift giving example of iron and tin compounds?
- **10.** What do you understand by quadrupole splitting?





# Chapter 33

# **Emission Spectroscopy**

## 33.1 INTRODUCTION

Most of the spectroscopic techniques deal with molecules. However, emission spectroscopy deals with atoms. The emission spectroscopy consists in the excitation of a sample using thermal or electric energy and the emitted radiation by the excited sample. This procedure is useful for metal analysis. Using this technique all metals and metalloids (like arsenic, silicon, selenium) can be identified and determined in low concentration (1 ppm). The element must be present in solid state. The liquid solution are evaporated to give solids and then analysed.

Emission spectroscopy is a very specific procedure, since each element gives wavelength pattern specific for each element. The sample can be analysed directly without any preliminary treatment. Also a metal can be analysed in short time (about one minute). However, the sample cannot be recovered back and the method is useful for the analysis of only metals (including metalloids).

When a sample is excited (thermally or electrically), its electrons, especially the valence electrons move from lowest to heighest energy levels. The excited electrons in turn rapidly emit a photon of energy and returns to the ground state. The emission of radiation from an exicted atom forms the basis of emission spectroscopy.

## 33.2 TYPES OF SPECTRA

In emission spectroscopy, we are concerned only with the emission spectra. These are of three types.

(a) Continuous Spectra: This type of spectra result when matter in the bulk is heated. As an example, incandescent solids like iron or carbon on heating until

they glow, emit continuous spectra. Such spectra do not show sharp lines or discrete bands. Continuous spectra depend on the temperature and do not depend on the chemical composition. Such spectras are not useful for spectrochemical analysis; these are generally avoided by volatisation of the sample before excitation.

- (b) Band Spectra: This type of spectra is given by excited molecules, which emit bands characteristic of the concerned molecule. So it is also called molecular spectrum. Ordinarly, a band spectrum consists of groups of lines which are very close together and so appear as continuous bands. Such spectras are of no use in emission spectroscopy. These can, however, be avoided by giving sufficient energy to the molecules so that they are degraded into individual atoms.
- (c) Line Spectra: This type spectra consist of discrete irregular lines. Such a spectrum is obtained only when the light emitting substance is in atomic state. So it is also called atomic spectrum. In atoms there are only electronic energy levels compared to vibrational and rotational energy levels (which are superimposed on the electronic energy levels) in molecules. Thus, line spectra is of predominate type in emission spectroscopy. Each element gives lines of different wave length, which depend on the energy of the excitation source. As an example sodium emits two characteristic lines which are due to the interaction between spin magnetic moment (due to p-electrons) and the magnetic field which results from the orbital motions of all electrons. The spin-magnetic moment may either oppose or reinforce with the orbital field and gives two energy states.

Characteristic doublets are obtained in all single electron systems (Na, Mg<sup>+</sup>, Al<sup>++</sup>, K, Rb, Cs). In these the wave length decreases with increase in atomic number of the atom, since larger is the nuclear charge greater is the energy differences between the quantum levels of the atoms.

A singlet and triplet series of lines are obtained in the emission spectra of magnesium atom. In fact, this is characteristic of all alkaline earth metals, Al<sup>+</sup>, Be<sup>+</sup> etc.

# 33.3 COMPARISON OF EMISSION SPECTROSCOPY WITH FLAME PHOTOMETRY

In flame photometry, the energy source is low and is useful mostly for Groups I and II metals and not for metalloids and transition metals.

(a) Flames: Flames are in general useful for those molecules which do not require very high temperature for excitation and dissociation into atoms. The temperature of the flame depends on the ratio of the type of fuel and oxidant, It also depends on the type of burner employed and the region in the flame that is focussed on the entrance slit (Table 33.1).

Fuel	Oxidant	Temperature (°C) of the flame
Natural gas	air	1700
Natural gas	oxygen	2700
Acetylene	air	2200
Acetylene	oxygen	3200
Hydrogen	oxygen	2800

Table 33.1 Commonly used fuels and oxidants and the temperature the flame

As seen the fuels is natural gas, acetylene or hydrogen and the oxidant is air or oxygen.

Flames in general are stable using rather cheaper materials. However, the composition of flames have an influence on inferences causing erroneous readings.

(b) Direct Current (DC) arc: A versatile excitation device, DC arc is used both for quantitative and spectrochemical analysis. A regulated power supply that furnishes 110-230 V at 3 to 30 A is the source of direct current. The current flows across an arc gap in series with a variable register R (10 to 40  $\Omega$ ) and a induction coil (L). On the other hand, metals are excited by an electric discharge and the temperature of the order 4000—6000 K is reached. In accordance with Boltzmann distribution, this is an effective atomic excitation system and most of the metals and non-metals can be excited using this procedure.

In flame photometry, commonly used emission lines arise from the transition between that first excited state and ground state. However, in emission spectroscopy these lines are useful only when high sensitivity is required. At higher temperature (as in emission spectroscopy) these lines became self-absorbed and reverse in intensity. These are called reversible lines. In fact non-reversible lines are preferred since line reversed makes quantitative analysis difficult to interpret.

Thus, emission spectroscopy is more sensitive and preferred over flame photometry.

# **33.4 INSTRUMENTATION**

In emission spectroscopy, the sample is excited by using a suitable excitation source (thermally a electrically). The excited atom emits radiation, which is passed through a monochromator and finally to the defector, which measures the radiation. The emission spectrometer consists of following components.

### 1. Source for Exciation

The function of the source is to vapourise the sample and dissociate into atoms and excite the electrons in the atoms to higher energy levels. A number of sources can be used for excitation. Some of such sources are given below (Fig. 33.1). The

sample (solid or liquid) is kept on the lower electrode in the arc gap. The arc is started after closing momentarily the gap and applying 100 to 220 V DC. When the current starts flowing, the temperature (in the gap) rapidly rises. At this stage the electrodes are separated by 20 mm to 1 cm. An electric arc establishes across the gap. The temperature of the arc 4000 to 8000°K is obtained.

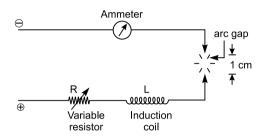


Fig. 33.1 DC arc

The current is adjusted by the variable resistance in the circuit R and the flow of the current stablises by the induction coil.

DC current arc generates high temperature which is very sensitive excitation source. Since the excitation energy obtained in the arc is thermal (and not electrical), it can excite all the metal elements.

(c) Alternating Current (AC) arc: The components of the circuit for an AC current arc is shown in Fig. 33.2. Using a transformer, high voltage of the order 2000 to 5000 V is maintained. This helps the arc to Jump the gap. In the primary circuit, a current of 1 to 5 V is maintained by the adjustment of a variable inductance L in this circuit. The current in the circuit is alternating at a frequency at 60 Hz. This extinguishes the arc 115-120 times per second. The arc picks a new surface area after each cycle and so whole of the sample gets arched and excited.

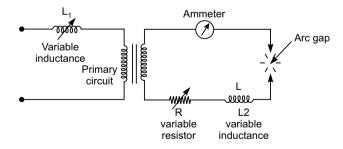


Fig. 33.2 Components of Circuit for an AC current

(d) Alternating Current (AC) Spark: The circuit for AC spark sources is schemetically represented in Fig. 33.3. A voltage of 110 to 220 is maintained in the primary circuit. This is stepped to 10,000 to 40,000V by a step-up transformer. The secondary circuit has a capacitor (C), an induction coil(L), a resistor  $(R_2)$ , a spark gap for the sample and an auxiliary spark gap, commonly known as synchronous spark gap.

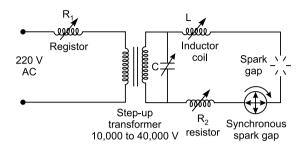


Fig. 33.3 Schematic representation of AC spark source

AC spark source is an excellent source for quantitative analysis since it is reproducible and stable. However, using AC spark source only minute amounts of the sample is volatilised, since the excitation of the sample is electrical, *i.e.*, by the bombardment of electrons.

(e) Laser Beam: Excitation of atoms or ions of a sample can be carried out using laser beam. The process consist in focussing the laser beam only on a small area of the specimen. Small amounts (about 50 microns) of solid specimen can be easily ionized. This process is called non-destructive as very small amount of the sample is vapourised.

# 2. Sample Holder

With the help of sample holder, the sample is introduced into electrical discharge. Different types of sample holders are used for solids and liquids.

# Sample Holder for Solids

A typical sample holder for solids is given in Fig. 33.4.

Different types of samples holders are used for different types of solids like metals, alloys, wood, plastic, paint, polymers, soil etc. A convenient procedure is to dissolve the sample in a known solvent like  $Al_2O_3$ ,  $SiO_2$  or C and drying to get a powder. This pre-treatment of the sample is necessary. The powder (obtained as given above) is loaded into a carbon sample holder as shown in Fig. 33.4. After loading, the sample holder is placed in position as one of the electrodes used in the discharge. From top surface of the electrode electrical discharge takes place, the sample is vapourised into plasma of the discharge resulting in spectrometric emission.

Using this procedure animal tissues, body fluids, plant material, soil etc. can be analysed for their metal components.



Fig. 33.4 Sample Holder for Solids (Side View)

Metals, metallic sheets or roads of alloys can be directly used as one of the electrodes of the electrical discharge. However, the sample has to be cleaned and shaped properly to fit into the electrode holder.

# Sample Holder for Liquids

A number of methods are available for the analysis of liquid samples. Two methods, *viz.*, a porous cup container and rotating disk electrode are commonly used.

The porous cup container (Fig. 33.5a) allows the liquid sample to percolate through it into the electrical discharge at a slow rate. A problem that arises in this method is that dissolved solids (like NaCl) in the sample form a crust at the base of the holder and so it gives erratic results. Reproducible results can be obtained if dissolved solids are less than one per cent of the sample. In case the solid contents are more, the sample be dried and ashed and run as solid samples.

In the rotating disk electrode (Fig. 33.5b), a rotating disk rotates through the liquid sample and surface of the disk becomes wet. Thus, the rotating disk carries the sample into the discharge at a steady rate. The emission intensity increases with the first two revolutions and then the signal becomes steady and can be measured. However, calibration data should be obtained under identical conditions.

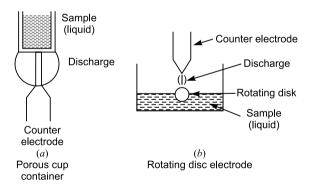


Fig. 33.5 Sample holders for Liquids

#### 3. Monochromators

The monochromator separates various lines of the emission spectra. Two types of monochromators are commonly used. These are prism monochromator and grating mochromator.

## **Prism Monochromators**

These are made of quartz or fused silica so that they are transparent to UV radiation. On passing light of one wavelength (emission line), it emerges as two lines [Fig. 33.6(a)]. This can be ractified by using two half prisms. The first half splits the light into two beams and the second half recombines the two beams to a single beam [Fig. 33.6(b)]. Such a combination of two half prisms is known as cornu type prism.

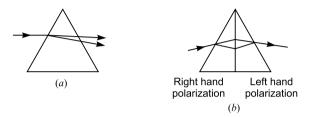


Fig. 33.6 (a) Single prism, (b) Two half prisms

In prism monochromators, emission lines of unknown wavelengths are difficult to identify by simple measuring their dispersion and so standards with emission lines from the spectrum of a unknown sample should be used.

# **Gratting Monochromators**

In gratting monochromators, the gratting is a series of parallel straight lines in a plane surface. For use in UV region the grating should bear 15000-30000 such gaps per square inch. These monochromators give better resolution and are used in sophisticated equipments.

## 4. Slits

In emission spectrographs, two slits are provided, one at the entrance and the other placed after the monochromator. The entrance slit checks the entry of stray light and permits only the light from the sample to enter the optical path. The exit slit blocks all but the desired wavelength to reach the detector. The slits are normally made of two parallel metal strips and are usually 1 cm high and 0.1 mm wide. However, the exact size depends on the experimental conditions.

#### 5. Detectors

Two types of detectors, *viz.*, photomultipliers and photographic plate are widely used. Photomultipliers are used for quantitative analysis and photographic plates are used for qualitative analysis.

**Photographic Plates** are used to record all emissions lines from the sample. The wavelength of radiation lines are determined from the photograph of the emission spectrum. The emitting elements can be identified from these lines.

Using photographic plates, a large number of spectral lines can be simultaneously recorded.

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**Photomultipliers:** In these photomultiplier tubes are used and such emission spectrographs give direct reading of the wavelength of the emitted lines. Such instruments require a large number of photomultiplier tubes for the simultaneous detection of different emission line. Due to this, direct reading instruments are costly. However, such instruments are very fast and more precise and so these are preferred.

The optical diagram of an emission spectrograph is shown in Fig. 33.7.

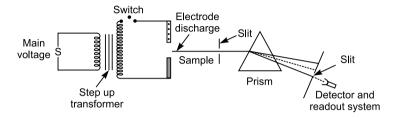


Fig. 33.7 Optical diagram of an emission spectrograph

## 33.5 APPLICATIONS OF EMISSION SPECTROSCOPY

Emission spectroscopy is useful in qualitative and quantitative determination of elements. This technique provides virtually no information for the molecular form of the sample. It has already been stated that the sample cannot be recovered since it is destroyed by electrical discharge.

# 1. Qualitative Analysis

The emission lines on a photographic plate are identified by comparison with previously recorded spectra of known elements. Trace amounts can also be determined by identification of *raies ultima* or RU lines as given below:

On excitation a metal emits complex spectra as a mixture of some strong and weak lines. If the concentration of the metal is reduced, the weaker lines disappear. On further dilution some more waker lines disappear and finally few lines remain. These are known as RU lines and are visible only when the concentration of the metal is considerably reduced. It is found that trace amounts of metals can be confirmed by the observation of at least three lines. In case all the three RU lines are not detected, the element is not present in detectable amount. Table 33.2 lists some of the elements which can be determined by this procedure.

Element	RU lines wave length (Å)	Element	RV lines wavelength Å
Ag	3280	Со	3453
	5209		3529

Table 33.2 Element detected by RU lines

(Contd...)

Element	+RU lines wave length (Å)	Element	RU lines wavelength Å
Au	2421 2802	Cr	4254 5206
Al	3961 3944	Cu	3247 3274
As	2288 2370	Fe	3581 3791
Ca	4426 4434	Hg	2536 4458
Cd	2288 3403	К	7664 7698

For determination of **metals**, photograph of the emission spectrum of an compound to be identified is taken along with the spectrum of iron. Comparison with iron spectra, the wavelength the lines from the sample are determined and by comparing with spectra of other elements, the elements in the sample are identified. In case the comparison instrument is unavailable, the identification of the line becomes tedious. In such cases, the wavelength of iron lines are first determined by refering to compilation published earlier. Finally, the wavelength of a line of interest is done by measuring its distance from one or more of known iron lines which are located nearby. Using a grating spectrograph, linear interpolation is used to calculate the wavelength. However, if prism spectrograph is used wavelength is obtained by using Hartmann formula

$$1 = \lambda_0 \frac{C}{d - d_0}$$

where  $\lambda$  is the wavelength of the line, d is its distance from some fixed point on the plate.  $\lambda_0$ , c and do are constants used which are evaluated from the measurements of d for three known wavelengths is the reference iron spectra.

# 2. Quantitative Analysis

For quantitative analysis, the intensity of one line of each element has to be determined. In case the compound is in small amount, most intense line is chosen. However, for larger concentration a waker line could be measured. Intensity measurement is generally carried out using photomultiplier detectors. Following two methods are employed in quantitative spectrographic analyses.

(a) Comparison Sample Method: In this method, the spectrum of the unknown sample and various standards are photographed alternalety on the same plate under same conditions. The concentration of the desired constituent is determined by comparison of the blackening of the lines of this constituent with the same lines on the standards. Both photometric or visual comprison of the blackening can be used.

The method is accurate provided it is possible to maintain constant excitation and exposure conditions. However, this is difficult.

(b) Internal Standard Method: This method is generally used in quantitative analysis. The method consist in the addition of a known amount of a standard (which should not be present in the sample to be analysed) to the sample. Excitation of the sample having internal standard in the electric discharge give spectral lines of the sample and internal standard. The lines are photographed on a photographic plate. We have to measure the intensity of the sample emission line and intensity of the internal standard emission line.

Knowing the concentration of the internal standard and the ratio of the intensity of the two lines, it is possible to calculate the concentration of the element which is to be determined.

(c) Miscellaneous Applications: Emission spectrscopy is useful in determination of the impurities of nickel, chromium, silicon, manganese, molybdenum, copper, aluminium etc. in iron and steel in metallurgical processes. Alloys of aluminium, magnesium, zinc, copper, lead and tin can be analysed.

In oil industry, lubricating oils are analysed for iron, nickel, chromium, manganese, copper, aluminium etc. The source of these metals in oil is because of the motion of piston and bearings.

In petroleum industry, oil feed stoks can be analysed for iron, nickel and vanadium. The presence of these make the fuel poor.

Animal tissues can be analysed for many elements like sodium, potassium, zinc, copper, calcium, magnesium, iron and nickel. The concentration of these trace elements are correlated to the ageing process.

# EXERCISES

- 1. What is the basis of Emission spectroscopy? Explain.
- 2. Discuss the various types of emission spectra.
- **3.** How does Emission spectroscopy differ from flame photometry?
- **4.** Describe the various components of a Emission spectrometer and what are their functions?
- **5.** Discuss the applications of Emission spectrscopy.
- **6.** What are the merits and demerits of prism and grating in emission spectroscopy.
- What type of spectra are obtained by the excited atoms and excited molecules.
- **8.** What are the different excitation source that can be used in emission spectrometer? Which one is the best?
- **9.** Explain what is ment by reversal.



# Part-VII)

# **Miscellaneous Instrumental Methods**

- Atomic Absorption Spectroscopy
- Flame Photometry
- Fluorimetry and Phosphorimetry
- Nephelometric and Turbidimeric Techniques
- Retractory and Interferometry
- X-ray Methods



# Chapter 34

# **Atomic Absorption Spectroscopy (AAS)**

# 34.1 INTRODUCTION

Atomic absorption spectroscopy (AAS) is used for detection and estimation of metals in solutions. It was only in 1955 that the use of AAS for detection and estimation of metallic elements was reported by Dr. A. Walsh of Australia. This discovery became a revolutionary development in the analysis of trace metals. This technique has been successfully used for the determation of as many as sixty five elements. The procedure involves converting the sample in atomic vapour followed by measurement of absorption of atomic vapour at a selected wavelength, which is characteristic of each individual element. The absorbance thus measured is proportional to the concentration of the element. In AAS technique, the element to be determined is first reduced to the elemented state and then vapourised and imposed in the beam from the source. This technique is specific as the atoms of only a particular elements can absorb radiation of their characteristic wavelength. Thus, a metal can be estimated in presence of other metals. The AAS method is extremely sensitive and trace elements can be determined in as low as 1 ppm.

The AAS method, however, cannot be used for elements like Ti, W, Mo, V etc., since oxides of these elements are formed in the flame. Another disadvantage is that certain elements like alkali metals, ionize at higher temperature. Since ions do not absorb atomic absorption wavelengths, the net result is a loss of signal. So such elements which form ions can not be correctly estimated.

When light of a particular wavelength (specific to the element under examination) passes through an atom (in the flame), part of the energy is absorbed. The atom changes from low energy state to higher energy state.

The total amount of light absorbed by atoms is given by the expression

$$v = \frac{\pi e^2}{mc}$$
. Nf

where, e is the electronic charge, m is the mass of electron, c is the velocity of light, N is the total number of atoms that absorb at frequency v in the light and fis the oscillator strength or ability of each atom to absorb at frequency v. Since  $\pi$ , e, m and c are constants, the above equation can be expressed by the following simplified form.

Total amount of energy absorbed = constant  $\times N \times f$ .

From the above, it is seen that absorption by atom is independent of the wavelength of absorption and temperature of the atom. Due to these features, the AAS has distinct advantage over **flame emission spectroscopy**.

# 34.2 THE INSTRUMENT AND PROCEDURE OF ESTIMATION

The most important component of AAS consists of a glass tube containing an inert gas 'Argon' at several mm pressure, an anode and a hallow cathode, the inside of which is coated with the metal to be analysed (Fig. 34.1).

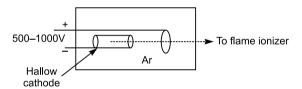


Fig. 34.1 Hallow cathode lamp

On application of high potential across the electrodes, an electric current of several milliamperes flows through the lamp. The Ar<sup>+</sup> ions produced inside the tube impringe on the cathode with very high energy, leading to sputtering of metal atoms from the surface of the cathode. These energised metal atoms emit radiations with a very narrow wavelength, characteristic of the metal. The radiations from the hallow cathode lamp passes through a flame automizer. (Fig. 34.2).

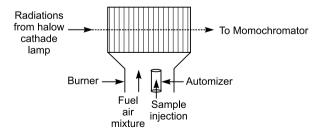


Fig. 34.2 Flame automizer

In the flame automizer, the metal (in the form of solution) sample to be analysed is aspirated. Here the solution gets converted into the metal, which in turn is decomposed in the flame and the metal gets reduced to the elemental state, forming a cloy of atoms, which absorb a fraction of the radiation in the flame. When a flame automizer is used, emission by the metal in the flame is at the same wavelength as the absorption wavelength of the metal since same electronic transition is involved. The decrease in radiant energy increases with the concentration of the metal in the sample.

The resultant energy (light beam) passes through a monochromator in order to eliminate any extraneous light resulting from the flame and finally to the detector and recorder. A diagrammatic representation of an atomic absorption spectrometer is shown in Fig. 34.3.

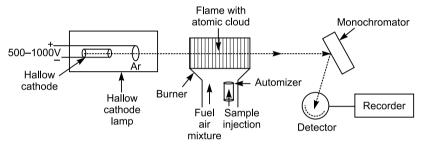


Fig. 34.3 Diagrammatic representation of AAS showing various components

The hallow cathode lamps are available for all elements that can be analysed by atomic absorption spectrometer.

Common fuels used in the burner are propane, hydrogen and acetylene and common oxidants are air, oxygen and nitrous oxide. Depending on the element (metal) to be estimated the temperature in the owen is adjusted accordingly. Following Table 34.1 gives the temperature for different fuels and oxidants.

Fuel	Oxidant	Temperature (°C)
Propane	air	1900
Acetylene	air	2200
Propane	Oxygen	2800
Acetylene	Nitrous oxide or oxygen	3000

**Table 34.1** Temperature for different flames

Elements such as copper, lead, zinc and cadmium are easily converted into atomic state and so low temperature flame is useful (*e.g.* propane-air or acetyleneair). Alkaline earth metals form refractory oxides and so require higher temperature for their decomposition. Some metals like aluminium, beryllium and rare earths

form stable oxide and they require still higher temperature which are achieved by acetylene oxygen or acetylene-nitrous oxide. The ratio of fuel to oxidant also determine the formation of atom in a flame.

Most common monochromotors used in AAS instrument are prisms or gratings. For some metals (like alkali metals) filter can be used in place of prism. The monochromator is invariably located between the burner and the detector.

The detectors which are commonly used in AAS instrument are film and photomultiplier; the latter type are used more commonly and these convert the radiant energy into an electrical signal. Normally direct reading calibrated meters are used. The meter is calibrated by adjusting to 100 per cent transmission (or zero absorbance) when a blank solution is sprayed into the flame.

It has been found that much better results are obtained by replacing flame automizer and the sample holder by a graphite furnace. The furnace has a hollow graphite cylinder placed in such a way that the beam of light from hollow cathode lamp can pass through it. A small amount of solution containing metal is injected in the tube through a hole at the top. On passing electric current through the tube, the metal sample is obtained in dry state and the absorption of the metal is recorded as a signal (peak). The Fig. 34.4 shows a graphite furnace with other components.

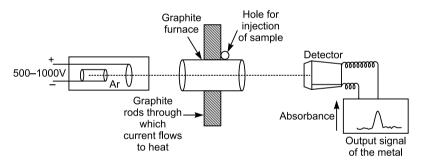


Fig. 34.4 Atomic absorption spectrometer using graphite furnace

The graphite furnace is 1000 times better than the flame detector. Table 34.2 below gives the detection limits for some elements.

<b>Table 34.2</b> Comparative detection limits of some elements by Flame AAS
and graphite furnace AAS (L. Herbert, S.B. Khan and R.G. Smith Scheicher,
American Laboratory, 65-79, Aug., 1979)

Element	Detection limits (PPM)		
	Flame AAS	Graphite furnace AAS	
Al	20	0.004	
As	100	0.06	

Element	Detection limits (PPM)			
	Flame AAS	Graphite furnace AAS		
В	1000	_		
Cd	1	0.008		
Co	5	0.03		
Cr	3	0.005		
Cu	2	0.008		
Fe	5	0.003		
Mn	3	0.004		
Мо	10	0.06		
Ni	8	0.02		
Р	10	3.0		
Pb	10	0.03		
Pt	50	0.45		
Se	100	0.10		
Si	60	0.10		
Ti	50	0.30		
U	7000	_		
V	20	0.15		
Zn	0.6	0.0007		

# 34.3 DETERMINATION OF THE CONCENTRATION OF ELEMENT IN ppm

The concentration of element in ppm is determined by referring to a calibration curve, which is drawn by spraying standard solutions of the element and plotting absorbance against concentration (ppm). A calibration curve for copper at 3428 Å is shown in Fig. 34.5.

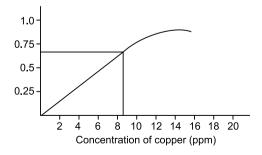


Fig. 34.5 Calibration curve for copper at 3428 Å

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Using the above procedure, elements like Cd, Cu, Co, Cr, Ca, Fe, Pb, Mn, Ni, Ag and Zn can be done using acetylene-air flame.

Low concentration of Cd, Cr and Pb can be determined by chelation with ammonium pyrrolidine dithiocarbamate, followed by extraction of the complex with methyl isobutyl ketone and aspiration into acetylene-air flame.

Element like Al, Ba, Be, Si and V can be estimated by direct aspiration of their solutions into acetylene-nitrous oxide flame.

Low concentration of Al and Be can be estimated by chelation with 8-oxyquinolinol, extraction with methylisobutyl ketone and aspiration into acetyle-nitrous oxide flame.

Some elements like As and Se are estimated by conversion to their hydrides followed by aspiration into Hydrogen-argon flame.

# Interference from light emitted by the flame

The radiations emitted by the flame interfere with the results. This problem can be overcome by interposing a rotating wheel (known as chopper) between the hollow cathode lamp and the flame. The chopper helps to break the steady light from the lamp into pulsating or intermittent light. The result in that the photocell (detector) receives only the pulsating current. Thus, only the pulsating current is amplified and recorded. In this way the absorption of light is measured without inteference from the light which is emitted by the flame.

### 34.4 DOUBLEBEAMATOMICABSORPTIONSPECTROMETER

The atomic absorption spectrometer which has been so far used is a single beam spectrometer. In this spectrometer, the light source (hollow cathode lamp) is placed before the burner and mechanical chopper is placed between the light source and the flame. In the absence of any sample in the flame, the detector and amplifier units are so arranged to give full deflection. After the aspiration of sample solution in the flame, the output of deflection is recorded. A disadvantage of the single beam atomic absorption spectrometer is low stability. However, increased stability can be obtained by using a double beam spectrometer. In this instrument, the chopped beam from the hollow cathode lamp is divided into two part, one part passes through the flame and the other part does not. The two beams are subsequently recombined and passed through a monochromator and detector-meter system.

# 34.5 ATOMIC ABSORPTION SPECTROSCOPY VERSUS FLAME EMISSION SPRECTROSCOPY

- 1. The absorbance in AAS is linearly related to the concentration. This is not so in flame emission spectroscopy.
- 2. The atoms in flame emission spectroscopy on putting in a flame get excited. The excited atoms being unstable, emit a photon of light and

return evantually to the unexcited state. It is the measurement of the emitted radiation which form the basis of flame emission spectroscopy. The emitted radiation is the sum total of all energies which are emitted as excited atoms attain ground state.

On the other hand, in AAS, the signal obtained is from the difference between the intensity of the source (in the absence of metallic element) and decreased intensity due to metallic elements present is the optical path.

3. In flame emission spectroscopy, the intensity of emission is dependent on the temperature of the flame. However, in AAS it is not so.

## 34.6 INTERFERENCE

In AAS technique there may be two types of interference. These are chemical interference and solvent interference.

# 34.6.1 Chemical Interference

This arises due to reactions occurred in the flame and is attributed to different anions present with the metal in a sample. The stability of the metal compounds formed during atomization depends on the anion present. The production of metal in the flame depends on the anion present. As an illustration, calcium chloride easily gives calcium as compared to calcium phosphate; the latter is comparatively more difficult to break down than calcium chloride Thus, two solutions of equal concentration of calcium in calcium chloride and calcium phosphate will absorb different amounts of radiation. However, such interference can be overcome by plotting calibration curves with standard which has the same anion present as in the sample to be analyzed.

## 34.6.2 Solvent Interference

The absorbance depends on the solvent used. Aqueous solutions are shown to show comparative low absorbance value compared to the same concentration of metal in organic solvent. Obviously, the metal is atomized more easily from organic solvent than from aqueous solution. So in this case also, the standards must be in the same solvent as the metal solution.

# 34.7 ADVANTAGESOFATOMICABSORPTIONSPECTROSCOPY

(i) The AAS procedure is specific of a particular element since the element can absorb radiation of its own characteristic. This, as we known is achieved conveniently since the hollow cathode lamp is coated with the element, which is to be analysed.

The concentration of the element is determined by referring to calibration curve which is plotted as explained in Fig. 34.5.

This technique is particularly useful for the determination and estimation of metallic pollutants in air, water and soil. A large number of metals can be estimated by this technique (*see* Table 34.2).

A small disadvantage in AAS technique is that only the element which is used in the hollow cathode lamp can be estimated.

However, using a multi element source (which is now available). Mitchell (1973) has been able to determine a number of elements simultaneously. A special detection system (*e.g.*, Videocon detection system) is used.

(ii) AAS is independent of the temperature of the flame.

# 34.8 APPLICATIONS OF ATOMIC ABSORPTION SPECTROSCOPY

It is now well-known that the sample for AAS must be in the form of solution. In case of solid samples (like alloys, soils, animal tissues, fertilizers, ores, cement, bone ash etc.), these must be dissolved in a suitable solvent and then aspirated into AAS.

Liquid samples (like urine, blood, wine, solutions used in electroplating, petroleum and pollutions in water) are directly analysed.

Metallic pollutants in air (or gases) are first collected by filtering through a fine filter (as shown in Fig. 34.6). By this procedure all types of particulate matter are collected.

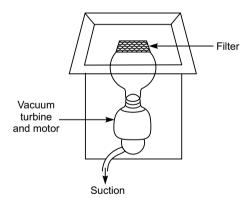


Fig. 34.6 A typical filtering assembly for collection of all particulate matter in air

The particulate matter is collected are heated in a mufftle furnace at  $500^{\circ}$ C for about 60 min to burn organic matter. The residue is extracted with  $2 \times 40$  mL 1:1 redistilled HNO<sub>3</sub> at about  $100^{\circ}$ C (heating for about 40 min for each extraction). The combined HNO<sub>3</sub> extract is filtered and evaporated to a small volume (3-4 mL) and then diluted with distilled water. The resulting solution is aspirated in AAS for determination/estimation of metals.

Following are given some of the applications of atomic absorption spectroscopy:

### 1. Detection and estimation of elements

As already stated a number of elements can be detected and estimated using AAS.

# 2. Determination of metals in Biological Systems

The procedure involved consist in decomposition of organic matter with 50 per cent  $H_2O_2$  or with conc.  $H_2SO_4$  and  $HNO_3$ . There is no standard procedure possible, since the quantities of  $H_2O_2$  and acids differ from material to material.

This method is used for determination of metals in liver and meat products. It should be noted that H<sub>2</sub>SO<sub>4</sub> cannot be used for determination lead due to formation of insoluble PbSO<sub>4</sub>.

# 3. Determination of Metallic Elements in Blood Serum

For diagonises of a number of pathological condition, it is necessary to determine various metallic elements (like calcium, magnesium sodium and potasium) in blood serum. The AAS technique is the best for determination of various elements in blood serum. The method consist in diluting the serum sample 10, 20 or 40 times in presence of LiCl. The determination in presence of LiCl is useful to overcome underestimation of calcium due to supression by phosphate. The solution so obtained is aspirated in AAS instrument and absorbance measured and compared with those for aqueous standard solutions. Calcium and magnesium can be estimated in presence of LiCl. Both sodium and potassium can be determined in serum by dilutions of 50:1 or 100:1.

## 4. Determination of Lead in Petrol

Lead gets deposited in bones and is a considered to be a cumulative poison. It is a general toxicant and gets accumulated in the skeleton. Infants, children up to six years of age and pregnant women are most suspectable to the adverse health effects.

Lead can be estimated in petrol by the spectrometric method or by Atomic Absorption spectrometric method.

(a) Spectrometric Method: It is based on the reaction of lead with dithizone to form a coloured complex. The coloured intensity (measured at 490 nm) gives the concentration of lead.

Following steps are involved:

(i) A sample of petrol (known volume) is treated with dilute nitric acid. The mixture is warmed to remove petrol. Lead gets extracted into dilute HNO<sub>3</sub>. The solution is neutralised with conc. NH<sub>4</sub>OH and treated with KCN (1.5g) and Na<sub>2</sub>SO<sub>3</sub> (5g). The solution is diluted to 500 mL and 10 mL dithiazone solution (68 mg in 250 mL CCl<sub>4</sub>) added. The solution

(using a separatory funnel).

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is shaken for about 60 seconds and red coloured CCl<sub>4</sub> layer separated

(ii) The absorbance of red coloured solution (obtained above) is measured at 515 nm against reagent blank.

The concentration of lead in ppm is determined by referring to a standard curve using lead solution of 0 to 20 ppm under identical conditions and politting against absorbance.

# (b) Atomic Absorption Spectrometric Method

In this method, petrol (known volume) is treated with dilute nitric acid and petrol removed by warming. To the residue is added pyrrolidine dithiocarbamate followed by extraction with methyl isobutyl ketone and aspiration of the extract into flame of AAS.

The concentration of lead is finally determined by referring to standard curve by using standard lead solutions of 0 to 20 ppm and treated under identical condition and plotting concentration against absorbance.

# 5. Simultaneous Determination of Metallic Elements

The AAS technique (as described) is useful for the determination of only one element at a time. Now multielement hollow cathode source is available with a detection system having a sepetral range of 2320 to 3281 Å. Using this technique, Mitchell has been able to determine simultaneously eight element (Zn, Cd, Ni, Co, Fe, Mn, Cu and Ag).

## 6. Estimation of Mercury in Water

Mercury in a known toxic metal. Its intake is responsible for neurological disorder and even death. It is estimated by flameless atomic absorption method (see section 34.9). Following steps are involved.

(i) Water sample (100 mL) is taken in a 500 mL distillation flask. To this is added 5-10 mL 5 per cent KMnO<sub>4</sub> solution followed by refluxing for 4 hr. using ice cold water for circulation in reflux condenser. To the cooled solution is added few mL of 30 per cent H<sub>2</sub>O<sub>2</sub> and solution boiled to remove excess of H<sub>2</sub>O<sub>2</sub>.

- (ii) The volume is made up to 250 mL.
- (iii) In the mean time, add 20 per cent SnCl<sub>2</sub> in 10 mL conc. HCl and 8 mL 10 per cent HNO<sub>3</sub> to reaction vessel (B). The solution is stirred (magnetic stirrer). Hg vapours in the reagent are pumped through 3 mL 20 per cent NaOH, and 50 per cent H<sub>2</sub>SO<sub>4</sub>.
- (*iv*) Finally introduce 1-2 mL of digested sample [obtained in step (*i*)] into the reaction vessel (*B*) containing 2 mL to Hg-free SnCl<sub>2</sub> and 8 mL 10 pecent HNO<sub>3</sub>. The mixture is stirred vigorously (magnetic stirrer).
- (v) Air is bubbled through vessel A and thus pumps Hg vapour through 20% NaOH (vessel C), 50% H<sub>2</sub>SO<sub>4</sub> (vessel D) and then to absorption cell.
- (vi) The absorbance is measured at 253.7 nm using a mercury hollow cathode lamp as the light source.
- (vii) The amount of Hg in water sample is measured by using a calibration curve which is drawn by taking known concentration of Hg<sup>2+</sup> solution and dilution to get 2, 4, 10, 12, 14, 16 ppm and determine the absorption by following the procedure as described above and plotting absorbance against concentration.
- (viii) The resulting Hg vapour are absorbed in 1% KMnO<sub>4</sub> 10% H<sub>2</sub>SO<sub>4</sub> in vessel E.

All the above steps are shown in Fig. 34.7.

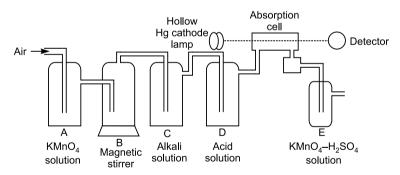


Fig. 34.7 Estimation of Mercury

As seen, the first trap (A) contains  $KMnO_7$  solution in order to purify air. The second cotainer (B) contains reagents and water sample (step IV) to generate Hg vapour. The third container (C) contains 22% NaOH solution to absorb acid vapours and the fourth container (D) contains 50%  $H_2SO_4$  (to absorb moisture). The container (E) contain  $KMnO_4 - H_2SO_4$  solution to Absorb Hg vapours so that they do not leak in the atmosphere.

## 34.9 FLAMELESS ATOMIC ABSORPTION METHOD

This is a special technique for estimation of mercury in water. It involves reduction of  $\mathrm{Hg^{2^+}}$  to elemental  $\mathrm{Hg^0}$  by  $\mathrm{SnCl_2}$  solution at room temperature. The resulting  $\mathrm{Hg}$  vapour is swept by air into an absorption cell, which is subject to beam of radiant energy from an  $\mathrm{Hg}$  vapour lamp. Measurement of atomic absorption at 253.7 nm gives the amount of mercury in ppm. Using this technique it is possible to estimate  $10^{-9}\mathrm{g}$  (nano gram) quantities of mercury. For more details *see* section 34.8.

# **EXERCISES**

- 1. Describe the sailent features of Atomic Absorption spectrometric instrument.
- **2.** What is the principle involved in the estimation of metallic elements in AAS procedure?
- **3.** Explain, why in AAS technique, the results are independent of the wavelength and temperature?
- **4.** What is the function of monochromator and chopper in AA Spectrometer?
- **5.** Explain what type of interference your come across in AAS method. How will you eliminate these interference?
- **6.** Describe double beam spectrometric technique. Why it is considered better than sigle beam AA Spectrometric technique.
- 7. How will you determine low concentration it Cr, Cd and Pb using AAS technique.
- 8. Compare AA Spectroscopy with Flame Emission spectroscopy
- 9. How are Al, Be and rare earths estimated?

[Hint. These elements form stable oxides and so higher temperature is required, which is achieved by using acetylene-nitrous oxide flame (temperature 3000°C).





# Chapter 35

# Flame Photometry

# 35.1 INTRODUCTION

Also known as **flame emission spectroscopy**, flame photometry is based on the emission of radiation when a metal is aspirated into flame. The radiation emitted is characteristic of the metal and the intensity of radiation is helpful to know the concentration of the metal aspirated into the flame.

In flame photometry, the atoms on putting in a flame get excited. The excited atoms being unstable, emit radiation and return evantually to the unexcited state. It is the measurement of the emitted radiation which form the basis of flame photometry. The emitted radiation is the sum total of all energies which are emitted as excited atoms attain ground state. The intensity of emission is depended on the temperature of the flame. In this way flame photometry is different from atomic absorption spectroscopy (*see* also section 36.5).

# 35.2 PRINCIPLE OF FLAME PHOTOMETRY

The metallic salt solution is aspirated into the flame. Following are given the subsequent steps involved:

- (i) The solvent in the sample gets evaporated resulting in the formation of a residue.
- (ii) The residue gets decomposed into neutral and/or radicals.
- (iii) The neutral atoms get excited to form excited atoms.
- (iv) The excited atoms, being unstable emit radiation and revert back to the ground state.
- (v) The wavelength and the intensity of radiation are measured.

The radiation emitted are given by the expression

$$E_2 - E_1 = hv \qquad \dots (i)$$

where  $E_2$  is the energy of higher energy level and  $E_1$  is the energy of lower energy level.

h is the Plancks constant and v the frequency emitted light.

$$E_2 - E_1 = h.\frac{c}{\lambda}$$
 and 
$$\lambda = \frac{h.c}{E_2 - E_1}$$

Thus it is possible to determine the intensity of radiation  $(\lambda)$ , which is characteristic of the element and the intensity of the radiation is used to determine the concentration of element in the flame.

As already stated (step *ii*), the neutral atoms or radicals, which are formed in the flame may also combine to give new products. The formation of the new products result in decrease in concentration of netural atoms. In fact, this is **chemical interference in flame photometry**.

The fraction of number of free atoms that get excited in the flame is given by Boltzmann distribution:

$$\frac{N^*}{N_0} = Ae^{-E/RT}$$

where

 $N^*$  is the number of excited atoms

 $N_0$  is the number of atoms remaining in the ground state

A is a constant for a particular element

E is activation energy

R is a gas constant

T is the temperature of the flame.

On the basis of the above equation it is inferred that the fraction of atom that get excited depends on the temperature of the flame. The temperature of the flame depends on the type of fuel and oxidant used. Temperatures of some flames are given in Table 35.1.

Table 35.1 Flame temperatures attained by fuel and oxidant

Fuel	Temperature (°C)			
	Air	Oxygen		
Hydrogen	2100	2780		
Methane	2000	2700		
Propane	1900	2800		
Butane	1900	2900		
Acetylene	2200	3000		
Cyanogen	_	4575		

In flame photometry, the flames used have comparately less amount of energy compared to other sources such as emission spectrograps or plasma emission torches. In view of this, flame photometry is useful for the determination of metals which get easily excited. Examples include alkali metals and alkaline earth metals. In fact flame photometry is not useful for determination of transition metals and other metals, which require much more energy to get excited. Such metals are estimated by emission spectroscopy or plasma emission where which higher energy is available; the latter procedures (viz. emission spectrography or plasma emission) cannot be used for the estimation of alkali metals and alkaline earth metals (in which much more energy is avalible), more ionization of the concerned metals leading to erraneous results.

At higher temperatures, the alkali metals and alkaline earth metals get ionised as is seen by consideration of the calculated ionisation potential of these metals at 2200°C, 3000°C and 3200°C; the latter temperature is achieved by using acetylene-nitrous oxide flame.

Elements	lonisation potential (eV)	2200°C acetylene- air)	3000°C (acetylene- oxygen	3200°C acetylene- nitrous oxide
Lithium	5.39	0.01	16	_
Sodium	5.14	1.1	26	_
Potassium	4.34	9.0	82	_
Rubidium	4.18	14.0	89	_
Cesium	3.48	29.0	96	_
Magnesium	7.64	_	0.01	6
Calcium	6.11	0.01	7	43
Strontium	5.69	0.01	17	84
Barium	5.21	2	42	88

Table 35.2 Per cent ionisation in at various temperatures

On the basis of the above data, it is clear that for the estimation of alkali metals and alkaline earth metals, it is best to use acetylene-air flame (2200°C). Under these conditions there is minimum ionisation.

## 35.3 COMPONENTS OF A FLAME PHOTOMETER

A flame photometer consists of the following components:

1. Burner: The function of a burner is to produce a flame, which as has already been stated has the following functions:

- evaporation of the solvent from the sample of the element to form a residuce.
- decomposition of the residue to form neutral atoms.
- excitment of the neutral atoms to form excited atoms, which in turn emit radiant energy.

The temperature of the flame depends in the type of burner used and the nature of the fuel (*see* table 35.1) and oxidant and their ratio.

In flame photometry different types of burners have been used. Some of these are given below:

- (a) Mecker Burner: This is a usual type of burner used in laboratories and uses a mixture of CNG and oxygen. This can be used for alkali metals only, since it produces low temperatures. A disadvantage of this burner is that the flame is not homogeneous and have different regions in the flame (like an oxidising region and a reducing region) which are responsible for giving different concentrations of excited atoms. As such, these are not used.
- **(b)** Total Consumption Burner: In this type of burner (Fig. 35.1) all the sample that enters the capillary tube finds its way into the flame and so it is called total consumption burner. The oxidant in this burner is hydrogen—oxygen mixture or acetylene-oxygen mixture. The fuel (H<sub>2</sub> or acetylene) and oxidant enter the burner from side tubes and burn to give a flame. The liquid sample is aspirated into the flame through a capillary tube with the help of oxygen. Subsequent steps of getting converted into solid residue, atomisation and excitation follow. The temperature of the fuel is adjusted from acetylene oxygen ratio. This type of burner was used earlier for a number of years but was subsequently replaced by other type of burners.

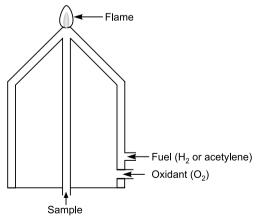


Fig. 35.1 Diagrammatic representation of a total consumption burner

A disadvantage associated with total consumption burner is turbulence as the gases are mixed and burnt in the same area.

(c) Premixed or Laminar–Flow Burner: In this type of burner, the fuel, oxidant and the aspirated sample are adequately mixed in a chamber fitted with baffles (or spoilers). The mixture subsequently enters the burner head and then into the flame. In this type of burner, only about 5 per cent of the sample reaches the flame. However, this results in effecient atomization of the sample in the flame. Rest of the sample is drained out. The flame produced in Laminar-flow burner is stable, non-turbulent and noiseless. A diagrammatic representation of premixed or Laminar-flow burner is given in Fig. 35.2.

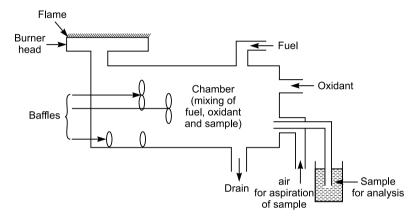


Fig. 35.2 Diagrammatic representation of a Pre-mixed or Laminar-Flow burner

The sample to be analysed must be in a single solvent. In case, a mixture of two solvents is used, the more volatile solvent will get evoporated in the chamber-the sample is left in the form is undissociated atoms in less volatile solvent. Due to this, only small number of atoms would reach the flame giving rise to reduction of emission intensity giving erraneous results.

(d) Lundergarph Burner: In this type of burner, the sample is aspirated (with help of oxygen) in a separate chamber from which it gets mixed with the fuel resulting in the formation of flame and subsequent steps of automisation and excitation. An diagrammatic representation Lundergraph burner is given in Fig. 35.3.

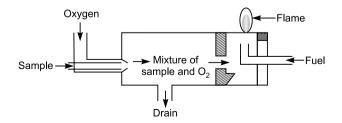


Fig. 35.3 Diagrammatic representation of Lundergraph Burner

#### 2. Mirrors

A concave mirror is located behind the burner in such a way that there is maximum reflection of the radiation to the entrance slit. This arrangement prevents loss of radiation from the flame (which is normally emitted in all directions). The instrument must be protected from corrosive atmosphere (which may tarnish the surface of the concave mirror.

#### 3. Slits

Most of the flame photometry instrument have two slits, the entrance and exit slits. The entrance slit, as already stated is placed after the mirror. This slit prevents interferring light so and only the light of desired wavelength passes on to the monochromator. The second slit is positioned after the monochromator. This slit prevents the light with undesired wavelength from reaching the detector (Fig. 35.4).

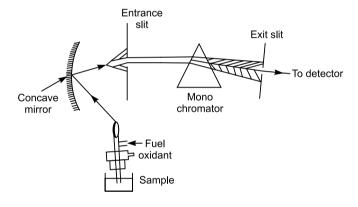


Fig. 35.4 Placement of Slits

#### 4. Monochromator

In most of the instruments monochromator is a prism (Fig. 35.4), which is made of quartz. Though dispersive power of glass is more than quartz, but quartz is transparent over the entire region. In some instruments in place of prism monochromator, greating type of monochromator (which is a series of straight line cut into plane surface) is also used.

#### 5. Filters

Some elements give emission spectrum which contains only few lines. In such a case, optical filters are used in place of slit and the monochromator system. The filters are made from a appropriate material which is transparent over a narrow spectral range. This filter is placed between the flame and the detector so that radiation of desired wavelength from the flame directly enters the detector and measured. The filter absorbs the undesired wavelength.

The optical filters which are used in place of slit and prism monochromator are called filter monochromators. Such instruments can, however, be used for only few elements.

#### 6. Detectors

The detector measures the intensity of radiation falling on it. In some instruments photomultiplier detectors are used. This produce an electrical signal (from the radiation falling on them) which is measured with a suitable meter.

# 35.4 SELECTION OF APPROPRIATE SOLVENT FOR DISSOLVING THE SALT IN FLAME PHOTOMETRY

As already stated, the sample to be analysed must be in a single solvent. The solvents which are normally used are water or any of the pure organic solvent. In case water is used as a solvent more energy is required to evaporate it. In such a case an inorganic salt is left, which needs more energy to decompose it. This is turn slows down the atomization process. However, if an organic solvent is used, it burns in the flame and the atomization process is increased, and there is enhancement of the signal. In case organic solvent is used, it should be ascertained that it is pure and no residue is left in the flame.

#### **35.5 INSTRUMENT**

Different varieties of flame photometers have been used. These are of two types:

- Simple flame photometer
- Internal standard flame photometer.

## 35.5.1 Simple Flame Photometer

A diagrammatic representation of a simple flame photometer is shown in Fig. 35.5.

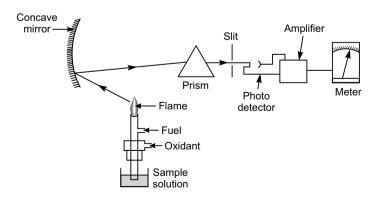


Fig. 35.5 Diagrammatic representation of a simple flame photometer.

The solution of sample to be analysed is sucked by an atomiser, which is operated by fuel or oxidant and aspirated into the flame. The emitted radiation is reflected *via*. a conave mirror on to a prism and a slit. The radiation of an appropriately selected wavelength is made to strike photodetector, producing an electrical signal which is measured by a suitable meter.

The temperature of the flame is adjusted by the flow rates of the fuel to oxidatant using pressure regulators (not shown in the figure).

#### 35.5.2 Internal Standard Flame Photometer

In this flame photometer, an internal standard (lithium) is used. The sample solution containing internal standard (lithium is equal concentration) is sucked by an atomiser, which is operated by the fuel or the oxidant and aspirated into the flame. The radiation emitted passes through a suitable filter and strikes a mirror. The mirror divides the emitted radiation into two parts. One part is due to internal standard (lithium) and the second part arises due to the presence of the element to be measured. Both the radiations fall on separate amplifiers and then on to a common detecting system (a calibrated potentiometer), which records the ratio of the intensity of the element to be determined to that of internal standard. A diagramatic representation of an internal standard flame photometer is given in Fig. 35.6.

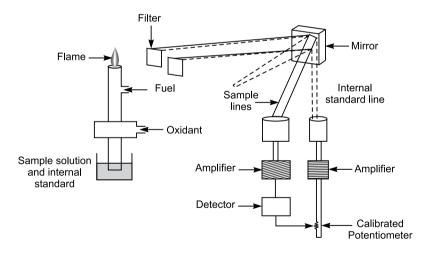


Fig. 35.6 Diagrammatic representation of as internal standard flame photometer

The internal standard flame photometer is much better than the simple flame photometer (section 35.5.1). This is because in internal standard flame photometer the errors arising due to difference in viscosity and surface tension are considerably reduced.

#### 35.6 TECHNIQUES OF ANALYSIS

A number of techniques have been used for flame photometric analysis. Some of these are discussed below.

## 35.6.1 Analysis Involving Calibration Curves

This technique involves the plotting a calibration curve. The method consists in preparing a number of standard solution of known concentration of the element which is to be estimated. The preparation of standard solutions forms the subject matter of a subsequent section (see section 35.7).

As a first step distilled water is aspirated into the flame and the instrument should read zero. The next step consist in aspirating standard solutions (containing 10, 5, 2.5 and 1 ppm of the element (sodium ion) one by one and the intensities of emissions measured in each case. Finally, a calibration curve is plotted using emission vs concentration. A straight line should be obtained.

Finally the unknown sample of the element (sodium ion) is aspirated into the flame. From the measurement of emission of intensity and refering to calibration curve the concentration of the element (sodium ions) is determined.

This procedure is used in case simple flame photometer has been used (see section 35.5.1).

#### 35.6.2 Analysis Involving Internal Standard

In this technique, lithium is chosen as an internal standard. Various steps involved are:

- (i) Standard solutions of lithium are prepared and each solution aspirated into the flame and emitted intensity is recorded.
- (ii) The emitted intensity of the unknown solution is measured as in step (*i*).
- (iii) The ratio of the two intensities is calculated and plotted vs. the concentration of the element to be determined to get a calibration curve.
- (iv) The unknown concentration is determined by referring to the curve.

## 35.6.3 Analysis Involving Addition of Standard

The technique involves determination of emission obtained for an aliquot of the unknown and for as identical aliquot containing a known quantity the element being determined. The concentration of the element in both the solutions is determined from the calibration curve. (also see section 35.5.2).

#### 35.7 PREPARATION OF STANDARD SOLUTIONS

As already stated, standard solutions are used in all the techniques described. Following are given procedures for making standard solutions of lithium, sodium, potasium and calcium.

- (*i*) Lithium: Pure lithium carbonate (2.6617g) is dissolved in dilute HCl (minimum amount) and the solution is diluted to 500 mL with distilled water in a calibrated flask. This solution contains 1000 mg of lithium per mL. It is suitably diluted to obtain solutions containing 20, 10, 5 and 2 ppm of lithium ions.
- (ii) Sodium: Sodium chloride (AR grade) (1.2709g) is dissolved in distilled water is a 500 mL standard flask. This solution is used for the preparation of four solutions containing 10, 5, 2.5 and 1 ppm of sodium ions.
- (iii) Potassium: Potassium chloride (AR grade) (0.9045 g) is dissolved in 500 mL standard flask in distilled water. This solution contains 1000 mg K per mL. The solution is used to prepare solutions containing 10, 5 and 2 ppm of potassium ions.
- (*iv*) Calcium: Pure calcium carbonate (1.7486 g) is dissolved is minimum amount of dilute HCl (AR grade). The solution is diluted with distilled water to 500 mL. This solution contains 1.400 mg calcium per mL and to used for preparing solutions of 100,50,25 and 10 ppm of calcium ions.

#### 35.8 INTERFERENCES IN FLAME PHOTOMETRY

Following are given some of the commonly encountered interferences in flame photometry.

(a) Ionisation Interference: Some metal atoms ionise at high temperature of the flame. For example, sodium ionises as follows

$$Na \Longrightarrow Na^+ + e^-$$

The ionisation results in the decrease in the concentration of neutral atoms, which in turn decreases the emission of radiation resulting in erraneous results. Such interference can be over come by addition of larger quantity of a potassium salt to the solution of the unknown as standard. This prevents the ionisation of sodium, but it itself undergoes ionisation. In this way the emission of sodium atom is enhanced. Such a interference is restricted to alkali metals (*see* also section 35.2).

(b) Spectral Interference: In case two elements are present these may exhibit different spectra characteristic of each element, but their spectras may overlap and both emit at some particular wavelength. The detector cannot differentiate between the sources of two radiations and will record total signal giving erronous results. As an example, iron line at 3247.28 Å overlaps the copper line at 3247.54 Å and iron line at 2852.13 Å overlaps the magnesium line at 2852.12 Å. Such type

of interference can be overcome by removing the interferring element. Alternatively, calibration curves are prepared from solutions having similar quantities of the interferring element.

Some times, it may so happen, that the spectral lines of two or more elements are close but their spectra do not overalp. Such a interference cannot be entirely eliminated since both the spectral line pass through the slit. In this case also, calibration curves can be prepared from solutions having similar quantities of both the elements.

- (c) Cation-Anion Interference: The presence of certain anions like oxalate, sulphate, phosphate and aluminate can affect the radiation intensity emitted by an element. As an example, calcium in presence of phosphate forms a stable salt resulting in depression of calcium signal (since calcium phosphate being stable does not easily decompose). Similary, barium in presence of sulphate forms stable BaSO<sub>4</sub> and low emission intensity is obtained. This type of interference can be eliminated either by removal of the anion or by using calibration curves with same anion at the same concentration in the standard solution as that in the sample.
- (d) Cation-Cation Interference: At times there is mutual interference of two cations. For example, there is mutual interference aluminium with calcium and magnesium. Also sodium-potassium have such interference on each other. The explanation of such interference is not well understood.
- (e) Interference due to Formation of Oxides: Some elements from stable oxides in presence of oxygen (which is used as an oxidant). This results in removal of major fraction of the metal-atoms resulting in low emission intensity. All alkaline earth elements are known to form stable oxides. Such interference can be removed by using much higher temperatures of flames. In this way the oxides get dissociated to form free atoms.

### 35.9 FACTORS WHICH AFFECT INTENSITY OF EMITTED RADIATION

The intensity of radiation emitted in flame photometry is affected by the following:

- (i) Viscosity: Addition of substances that cause increase in viscosity lower the intensity of emitted light. This results in reduced efficiency of atomisation.
- (ii) Presence of acids: The intensity of emitted radiation is decreased in presence of acids. This is attributed due to disturbance of the initial dissociation equilibrium.

(iii) Presence of Other Metals: The intensity of emitted radiation is altered due to presence of other metals. This can be overcome by using special type of filters which will selectively absorb radiations from other metals

#### 35.10 LIMITATIONS OF FLAME PHOTOMETRY

- (i) Only liquid samples are used. At times it is tedious to prepare liquid samples.
- (ii) This technique cannot be used for direct determination of all metals.
- (iii) Inert gas cannot be determined by this procedure.
- (*iv*) No information is obtained about the molecular form of the metal. Only total metal content present in a sample can be determined.

#### 35.11 APPLICATIONS OF FLAME PHOTOMETRY

- (i) Qualitative Analysis: Flame photometry is mostly used for the detection of alkali metals (Na, K, Li) and alkaline earth metals (Mg, Ca, Sr, Ba). Though some of these metals (like Na, K, Ca, Ba, Sr) can be detected by flame colour tests, but this method is not very reliable. The best method is to use flame photometry. Use of an appropriate filter or monochromator separates the wavelength characteristic of different metals. The method is simple, fast and reliable.
- (ii) Quantative Analysis: Besides alkali and alkaline metals, other metals can also be estimated by using high opitcal resolution unit. Some of the other metals which can be estimated include Al, Sb, As, Ba, Bi, Ca, Cu, Fe, Pb, Hg, Ni and Pd.
- (iii) Multielement Analysis: A number of methods have been developed for carrying out similtaneous multielement analysis. Mitchell *et al* (1975) used a Vidicon detection system with 0.25 m monochromator. By this procedure spectral lines as close as 20 Å could be readily resolved. As many as 10 elements could be simultaneously determined.
  - Bush *et al* (1974) used a silicon diode vidicon detector and an 0.5 m monochromator and could resolve lines as close as 1.4 Å, Busch *et al* could determine simultaneously seven elements (Mo, Fe, Cu, Al, Ti, Mn and K).
- (iv) Miscellaneous Applications: Flame phtometry has been used for
  - analysis of biological fluids and tissues for detecting the presence of Na, K, Al, Ca, Co and Fe.

- analysis of soil for determining the presence of Na, K, Al, Ca, Co and Fe.
- analysis of natural and industrial waters.
- analysis of glass, cement and metallurgical products.

For comparison of flame photometry with atomic absorption spectroscopy. See section 36.5.

# EXERCISES

- 1. Discuss the principle of flame photometer.
- 2. What type of interferences you come across in flame photometry?
- **3.** What are the limitations of flame photometery?
- 4. What factors affect the intensity of emitted radiation in flame photometry?
- 5. Write a note on simultaneous multielement analysis by flame photometry.
- **6.** Give the applications of flame photometry.
- 7. Using higher temperaturer of the order of 3200°C using acetylene nitrous oxide why alkali metals cannot be estimated.

**Hint:** At higher temperature the alkali metals are easily ionised giving erraneous results.

8. In flame photometry, the monochromator (prism) is made from quartz and not glass. Explain.

**Hint:** The dispersive power of quartz is less than that of glass but quartz is transparent over the entire region.





# Chapter 36

# Fluorimetry and Phosphorimetry

#### **36.1 INTRODUCTION**

There are a number of substances which absorb light energy (ultraviolet or visible). These molecules *via*. collusions with neighbouring atoms or molecules lose extra energy as heat. However, in some cases, there are substances which lose only a part of the excess energy as heat and emit the remaining energy as electromagnetic radiation and return back to the ground state. Such a process of emitting radiation is known as **luminescence**.

Luminescence is of two types:

- (i) Fluorescence: If a beam of light is incident on some substances, they emit radiations, such a phenomenon is known as *fluorescence*. This phenomenon is instaneous and starts after the absorption of light and stops as and when the incident light is cut off. The substances showing the phenomenon of flueorescence are called *flueorescent substances*.
  - Thus, fluorescence is the property of many substances of absorbing light of one wavelength and in its place emitting light of another wavelength.
- (ii) Phosphorescene: It is a kind of luminescence in which a substance after light radiation falling on it, emit light continuously even after the incident light is cut off. It is a type of delayed fluorescence and is called phosphorescence. Such substances are called phosphorescent substances.

The life time of phosphorescene ( $10^{-4}$  to 20 seconds) is much longer that of fluorescence ( $10^{-16}$  to  $10^{-4}$  second).

#### 36.2 FLUORESCENCE AND ABSORPTION METHOD

Ultraviolet and visible spectrometers and fluorimeters are complementary instruments. Since only few compounds exhibit fluorescence, a valuable tool for analytical process is available. Thus, fluorescent compounds can be determined in presence of non-fluorescent compounds without subjecting the sample to any preliminary treatments in order to remove interference due to absorption.

The fluorescence methods are versatile compared to absorption methods. In fluorescence both absorption and emission wavelengths can be used. Also by using wavelengths at other than the maxima, interference can be avoided. However, in spectrometric absorption methods errors arise due to either the rapidly rising or falling portions of a spectrum resulting in loss of sensitivity. In fluorescence method no such restrictions are there.

Fluorescence methods are about 100 time more accruate than the absorbance methods. Using fluorescence methods it is possible to determine two components (which emit same fluorescence colours) simultaneously, provoded their respective excitation wavelengths are sufficiently far.

A limitation associated with fluorescence method is that the fluorescent intensity is dependent on pH, so careful buffering is necessary.

#### **36.3 FLUOREMETRY AND PHOSPHORIMETRY**

On the basis of experimental considerations, fluoremetry is much simpler than phosphorimetry. The two techniques are, however complementary. Being more selective, phosphorimetry is used when interferring substances are present in the sample.

In phosphorimetry there is no scattering problems, which are associated in fluoremetric analysis. Also in phosphorescence, the quantum efficiences are greater. This is because the measurements are carried out at –196°C (liquid air or liquid nitrogen temperature). On the other hand fluoremetric analysis is performed at room temperature. Also the lower temperature used in phosphorimetry do not permit quenching to become serious.

In case fluorometry and phosphorimetry are equally sensitive, the former (fluoremetry) is preferred.

#### 36.4 THEORY

The irradiation of molecules with light of appropriate frequency take about  $10^{-15}$  seconds for absorption of light. This results in the molecules to move from ground state to the first excited singlet electronic state. The excited singlet state may encounter one of the following three phenomena:

 The excited singlet state being comparatively unstable may return back to the ground state by collisional deactivation without emitting radiation.

- The molecule in the singlet excited state may emit light photon in an ultraviolet or visible state. This phenomenon is called *fluorescence*.
- The molecule with comparatively stable excited state may give rise to a metastable triplet state and subsequently return to the ground state by emission of an ultraviolet visible light photon. This phenomeon is called *phosphorescence emission*.

The transition from triplet to ground state is forbidden by the spin symmetry and is a slow process. So the life-span of phosphorescence is longer than that of fluorescence.

The mechanism of phosphorescence (as stated above) involving singlet-triplet decay finds support by the magnetic susceptibility and ESR measurements.

At this stage it is appropriate to understand singlet and triplet state of molecules. Both these terms (arise from the consideration of atomic spectroscopy) define the number of unpaired electrons when magnetic field is absent. In case there are n number of electrons, it implies that (n + 1) fold degneracy of equal energy states are associated with the electron spin. However, if there are no unpaired electrons (n = 0), there will be only n + 1 or 0 + 1 or 1 spin state. Such a state is known as *singlet state*. In a similar way, systems having 1, 2, 3, ...unpaired electrons are in double, triplet, quartet etc. states respectively.

A number of factors affect fluorescence and phosphorescence. Some of these factors are given below:

(i) Nature of Molecules: Only the molecules which can absorb ultraviolet or visible radiation show the phenomena of fluorescence and phosphorescence. Thus, molecules having conjugated double bonds exibit these phenomena. In other words, aliphatic and saturated cyclic organic compounds do not respond to fluorescence and phosphorescence.

The presence of electron donating groups (in the molecule) like  $-\mathrm{NH}_2$ , —OH etc have a marked effect on fluorescence and phosphorescence. On the other hand, the presence of electron withdrawing groups like —COOH, — $\mathrm{NO}_2$ , — $\mathrm{N}=\mathrm{N}$ — decrease or even destroy fluorescence. It is found that the introduction of a high-atomic number atom into a  $\pi$ -electron system results in the enhancement of phosphorescence and decrease in fluorescence.

(ii) Concentration: A very dilute solution (10 to 100 times weaker than employed in spectrophotometry) is required. A problem, may however, arise due to adsorption of the fluorescent substance on the walls of the container. So a strong stock solution is kept and diluted as and when required.

- (iii) Light: For the excitation in fluorescence in quantitative work, monochromatic light is necessary. This is necessary, since the intensity varies with wavelength. However, a possible source of error is the Raman emission from the solvent used. In order to remove interference from Raman scatter, it is best to use radiation of a wavelength far removed from the fluorescence peak.
- (iv) Oxygen: In presence of oxygen, the fluorescent substance is oxidised to non-fluorecent product. It is, therefore, necessary to use de-aerated solution.
- (v) pH: Change in pH has a significant effect on fluorescence. As an example, phenols are fluorescent in both undissociated and dissociated forms. It implies that the fluoresence from a solution of a phenol will show two peaks, one of which is due to the ionic forms. The peak due to ionic forms can be suppressed in strongly acidic solutions.
- (vi) Temperature: An increase in temperature decreases fluorescence due to deactivation of the excited molecules by collision. In a similar way some substances which are not fluorescent at room temperature emit light when excited at low temperature.
- (vii) Viscosity: A decrease in viscosity causes decreased fluorescence.

### 36.4.1 Relation between Fluorescence Intensity and Concentration

It has been found that fluorescent intensity is proportional to the concentration of the fluoresent substances.

#### **36.5 TYPES OF TRANSITIONS IN FLUORESCENCE**

In general, fluorescence is not observed with ultraviolet radiation having wave length less than 250 nm. This is because the high energy of radiation causes deactivation. So flurescence due to  $\pi^* \to \sigma$  transition is generally not observed. The emission however, remains confined to less energetic transitions of the type  $\pi^* \rightarrow \pi$  and  $\pi^* - n$ . Most likely the fluorescence is observed from a transition from the first excited state to the ground state.

Aromatic compounds containing functional group of low energy  $\pi \rightarrow \pi^*$ transition exhibit intense and useful fluorescence. Fluorescence is also observed in compounds containing aliphatic and acyclic carbonyl groups or conjugated double bonds.

It is found that the absorption maxima and the fluorescence peaks shift in benzene containing substitution (Table 36.1).

Wavelength fluorescence (nm)	Relative intensity of fluorescence	
270-310	10	
270-310	17	
275-345	7	
292-380	5	
_	0	
283-365	18	
310–400	10	
312–392	3	
_	0	
272–322	17	
	### Time	

**Table 36.1** Effect of Substitution on the fluorescence of substituted benzene derivatives

Heterocyclic compounds like pyridine, thiophene and pyrrole do not exhibit fluorescence. However, heterocyclic compounds with fused ring show fluorescence. This is because fusion of heterocyclic nuclues to a benzene ring causes increase in molar absorptivity of the absorption peak. It is found that heterocyclic compounds containing nitrogen do not fluorence due to the involvement of low energy transition ( $n \to \pi *$ ) system, which is rapidly converted into the triplet state.

#### **36.6 INSTRUMENTATION**

## 36.6.1 Instrument for Fluorimetric Analysis

The instruments which are used for the measurement of fluorescence are called *fluorimeters* or *fluorophotometers*. In these instruments, the wavelength of excitation is obtained by using filters. A fluoremeter is described as a manual instrument and is useful for the measurement at one or two wavelength since a filter has to be changed each time the wavelength is changed.

In a way fluorimeters are like absorptiometers. In both cases, the sample is irradiated and radiation leaving the sample is measured. However, in fluorimeters, the illumination is done at right angles to the direction of observation. A typical fluorimeter is shown in Fig. 36.1. It uses a mercury vapour lamp, a condensing lens, a primary filter, a container for the sample, a secondary filter and a photocell. Normally, primary filter selects ultraviolet but not visible radiation and the secondary filter transmits the visible fluorescent radiation and absorbs incident ultraviolet radiation.

The light from the mercury vapour lamp passes through a condensing lens and then through a primary filter. The radiation (ultraviolet) from the primary filter

passes through the sample. The ultraviolet and fluorescent radiations obtained from the sample passes through a second filter, which absorbs the primary radiant energy, the fluorescent radiations are transmitted, which are received by the photocell (placed at right angle to the incident beam). The output from the photocell is measured by a suitable device like a sensitive galvanometer.

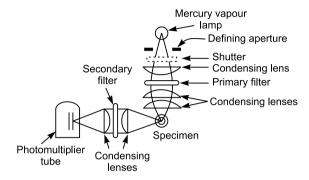


Fig. 36.1 A typical Fluorimeter showing various parts

Since the fluorescence intensity is proportional to the intensity of irradiation, the light source must be stable. The fluctuations in the intensity of irradiation are removed by using two photocells and the readings recorded on a potentiometer by balancing the photocells against each other. A double beam filter fluorimeter is shown in Fig. 36.2.

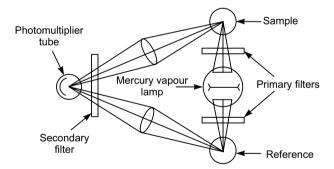


Fig. 36.2 A double beam filter fluorimeter

In the double-beam filter fluorimeter (Fig. 36.2) a specially designed mercury vapour lamp, having two anodes on opposite sides of a centre structure is used. This lamp works by alternating current, which enables two anodes to receive the discharge and so produces radiations on alternate half-cycles of the exciting voltage. In this way, the sample and the standard receive identical radiation. The two beams from the lamp pass through two primary filters. Subsequently, the beams simultaneously pass through sample and reference. The fluorescent radiation

from the sample and reference converge through a common secondary filter on to a single photomultiplier tube, which records the ratio of the two fluoroscent signals. In this system, there is minimum effect of the changes in temperature and variations in source and the detector.

### 36.6.2 Instrument for Phosphorimetric Analysis

It has already been stated that compounds with phosphorescence also show fluorescence and that phosphorescence is a type of delayed fluorescence (section 36.1). So a instrument for measurement of phosphorescence (a phosphorimeter) should be capable of making a distinction between the two. It is possible to achieve this by using a rotating shutter which is capable introducing a delay between the times during the time of irradiation of the sample and observation. A typical single-beam phosphorimeter with a rotating shutter is shown in Fig. 36.3.

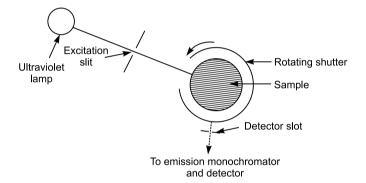


Fig. 36.3 A single-beam phosphorimeter with a rotating shutter

In the above instrument (Fig. 36.3) filters can be located in either the primary beam or secondary beam or both. The rotating shutter device is called a phosphoroscope and the sample system is maintained at liquid nitrogen temperature.

Different types of phosphoroscopes are available. A typical rotating can phosphoroscope is shown in Fig. 36.4.

The instrument (Fig. 36.4) has a hollow cylinder having one or more slits equally spaced in the circumference. A variable-speed motor rotates the cyclinder. On rotation of the rotating-can by a motor, first the sample is illuminated and then darkened. When there is dark, phosphorescence radiation passes to the emission monochromator and is measured. The type of rotating-can phosphoroscope is used in the Aminco spectrophosphorimeter.

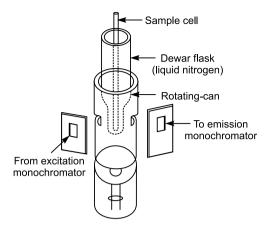


Fig. 36.4 Rotating-can phosphoroscope

#### 36.7 APPLICATIONS OF FLUORIMETRY

Some of the important applications of fluorimetry are given below:

- (i) Determination of Uranium in Salts: The sample containing uranium is oxidised by evaporating with nitric acid. The oxidised sample is fused with sodium fluoride to a melt, which is cooled to a solid glassy substance. This is examined is a specially designed fluorimeter. Using this method it is possible to determine uranium in concentration of  $5 \times 10^{-9}$  g/gram of solid sample.
- (ii) Analysis of Inorganic Ions: Normally inorganic ions do not exhibit fluorescence. It has however, been found that some of the inorganic ions form fluorescent chelates with non-fluorescent organic molecules. Most of the transition elements can be analysed by this procedure. Some examples include:
  - Determination of ruthenium ion in presence of other platinum metals.
    - Ruthenium forms a complex ion with 5-methyl-1,10-phenanthroline. The complex exhibits fluorescence strongly at pH 6. Using this method, ruthenium can be determined in the range 0.3 to  $2.0 \,\mu\text{g/mL}$  in presence of interfering elements of platinium group (present to the extent of  $30 \,\mu\text{g/mL}$ ).
  - Determination of aluminium in alloys.
    - Aluminium is known to form a complex with the dye, potochrome Blue-black RM at pH 4.8.
    - This complex fluoresces strongly. Using this method aluminium can be determined in the range 0.2 to 25 mg in a volume of 50 mL.

- Determination of traces of boron in steel: The boron in acid solution of steel is converted into boric acid, which is distilled with methanol. The resulting distillate having boric acid is neutralised (sodium hydroxide) and methanol distilled. The sodium salt of boric acid left in the distillation flask is treated with alcoholic solution of benzoin. The resulting fluorescent solution is examined by fluorimetry. Using this procedure, boron up to concentration of 100 µg in 50 mL can be determined.
- Determination of cadmium: It is estimated by precipitation with 2-(2-hydroxyphenyl) benzoxazole in the presence of tartarate. The solution of this complex in glacial acetic acid gives bright blue fluorence in uv light. The solution in acetic acid is used for the determination of cadmium by fluorimetry.
- (iii) Fluorescent Indicators: The colour and intensity of fluorescent indicators depends on the pH. These indicators are used in acid-base titrations. These can be used in the titration of coloured solutions in which it is defficult to see the change in colour. Same examples fluorescent indicators are given in Table 36.2.

IndicatorpH rangeColour ChangeFluorescein4.0–6.0Colourless→greenEosin3.0–6.0Colourless→greenAcridine5.2–6.6Green→violet blue2-Hydroxycinnamic acid7.2–9.0Colourless→green

Table 36.2 Some Fluorescent Indicators

(*iv*) Fluorometric Reagent: These are useful for the analysis of cations. Applications of some fluorometric reagents and their applications are given in Table 36.3.

<b>Table 36.3</b>	Some Fluorometric	reagents for	r inorganic species
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lon	Reagent	Fluorescence Wave length (nm)	Sensitivity (µg/mL)
Al <sup>3+</sup>	Alizarin garnet B	500	0.007
Li+	8-Hydroxy quinioline	580	0.2
Sn <sup>4+</sup>	Flavanol	470	0.1
Zn <sup>2+</sup>	Benzoin	green	10

- (v) Organic Analysis: Some of the important applications in organic analysis include determination of vitamin B<sub>1</sub> (thiamine) vitamin B<sub>2</sub> (riboflavin) and quinine.
  - (a) Determination of Vitamin B<sub>1</sub>: Vitamin B<sub>1</sub> (Thiamine) in non-fluorescent but its oxidation product, thiochromene gives blue fluorescence. This forms the basis of determination of vitamin B<sub>1</sub> in food stuffs. The method involves treatment of food sample with phosphatase in order to hydrolyse the phosphate esters. The clear filtrate is diluted to a known volume. From the filtrate equal volumes of two aliquots are taken-one for analysis and the other for blank. Both the aliquots are rendered alkaline with equal quantities of sodium hydroxide and equal volumes of isobutyl alcohol added. To the first aliquot is added an oxidising agent (e.g., potassium ferricyanide) solution shaken and alcoholic solution separated using a separatory funnel. The alcoholic solution is examined in the fluorimeter using a blank as reference. The total procedure is repeated using a standard thiamine solution.
  - (b) Determination of Vitamin B<sub>2</sub> (Riboflavin): Vitamin B<sub>2</sub> (riboflavin) is fluorescent but its oxidation products are nonfluorescence. This forms the basis for the determination of vitamin B<sub>2</sub>. In fact the fluorescent power of vitamin B<sub>2</sub> depends on the exact conditions and the amount and the nature of the impurities present. The fluorescence of a sample of the standard is measured in the same solution with the unknown in order to find if any impurities have the same effect upon unknown as standard. The method consists in extracting the sample (containing vitamin B<sub>2</sub>) with acids and precipitating the interfering salts using different reagents. The resulting solution is oxidised using dilute  $\mathrm{KMnO}_4$  solution as an oxidant. The residual fluorescence is measured; This is the blank value. Subsequently, a reducing agent (Na<sub>2</sub>S<sub>2</sub>O<sub>4</sub>) is added and the fluorescence again determind. Finally, a known volume of the standard added and fluorescence again determined. Following observations are recorded.

Oxidised sample (10 mL) +  $H_2O$  (1 mL) Fluorescence  $F_1$ Oxidised sample (10 mL) +  $Na_2S_2O_4$  Fluorescence  $F_2$ Oxidised sample (10 mL) + Standard (1 mL) Fluorescence  $F_3$ . Finally, the concentration of fluorescent material (vitamin  ${\bf B}_2$ ) is calculated by using the formula

$$\frac{F_2 - F_1}{F_3 - F_1} = \frac{m_x}{m_x + m_s}$$

where  $m_x$  and  $m_s$  are the masses of vitamin  $B_2$  in sample and standard.

(c) Fluorometric methods are also useful in the determination of adenine, anthranilic acid, aromatic polycyclic hydrocarbons, cysteine, guanidine, indole, skatole, tryptophan, uric acid, warfarin etc. Even medicinal products (e.g., adrenaline, chloroquin, pencillin, phenobarbital etc.) and plant products (e.g., chlorophyll, ergot, ascorbic acid, folic acid etc.) can be determined using, fluorometric methods.

#### 36.8 APPLICATIONS OF PHOSPHORIMETRY

Some of the important applications of phosphorimetry are given below:

- (i) Aspirin can be analysed to the extent of 0.02-1.00 mg/mL in blood serum. The metabolic product of aspirin is salicylic acid, which fluoresces but not phosphoresce. However, aspirin shows phosphorescence and not fluorescence. Thus, using the chloroform extract of serum, the amount of salicylic acid and aspirin are determind by flurometry and phosphorimetry respectively.
- (ii) Procane, cocaine, phenobarbital and chlorpromazine can be determined in low concentration in blood serum.
- (iii) In combination with thin layer chromatography, phosphorimetry has been used. A typical example is the separation of the alkaloids nicotine, nor nicotine and anabasine in crude tobacco by TLC on alumina. The separated drugs are scrapped from the TLC plate and determined by phosphorimetry.
- (iv) Cocaine and atropine in urine can be determined.

#### 36.9 COMPARISONOFFLUORIMETRYANDPHOSPHORIMETRY

Fluorimetry finds large number of applications than phosphorimetry. This is because phosphorimetry is experimentally more complicated. However, the two are complementary.

For the analysis of samples containing interfering substances, phosphorimetry is more selective and sensitive. This is because is phosphrimetry there are no scattering problems. Also, in phosphorescence, the quantum efficiencies are greater than in fluorescence. This is because phosphorimetric determinations are carried out at much lower temperatures (–196°C) compared to fluorimetric determinations which are carried out at room temperature. At low temperatures in phosphorimetry quenching does not create any serious problem than in fluorimetry.

Both fluorimetry and phosphorimetry are 10 to  $10^3$  times more sensitive than the absorption methods.



- 1. Give briefly differences between fluorescence and phosphorescence.
- 2. Give differences between:
  - (i) Fluorescence and absorption methods
  - (ii) Fluorimetry and phosphorimetry.
- **3.** Discuss the theoretical principles involved is fluorimetry and phosphorimetry.
- **4.** What types of transitions are involved in fluorescence giving examples?
- **5.** Discuss the instrument for fluorimetric analysis and phosphorimetric analysis. What is the difference between the two instruments?
- **6.** Explain singlet and triplet states.
- 7. What are fluorescence indicators?
- 8. How will you determine
  - (i) Uranium in salts
  - (ii) Determination of Vitamin B<sub>1</sub> and Vitamin B<sub>2</sub>
  - (iii) Analysis of blood serium for aspirin, procane, phenobarbital
  - (iv) Cocaine and atropine in urine.
- **9.** How is phosphorimetry used in combination with TLC giving an example?





# Chapter 37

# Nephelometric and Turbidimetric Techniques

#### **37.1 INTRODUCTION**

Both nephelometric and turbidimetric techniques of analysis are in a way closely allied to colorimetry. These are based on the scattering of light by particles suspended in a solution. The two techniques differ in the manner of measuring the scattered radiation.

In tubidemetric analysis, light is passed through a suspension. A part of the incident radiant energy is dissipated by absorption, reflection and refraction while the remaining incident radiant energy is transmitted. The measurement of the transmitted light as a function of concentration of the suspended particles is the basis of turbidimetric analysis (Fig. 37.1).

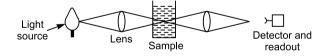


Fig. 37.1 Tubidimetric analysis

However, in nephelometry, though light is allowed to pass directly through the sample solution (as in the case of turbidimetry), the amount of radiation scattered by the particles is measured at an angle 90° to the incident light. Thus, the measurement of the intensity of the scattered light as a function of concentration of the dispressed phase is the basis of nephelometric analysis (Fig. 37.2).

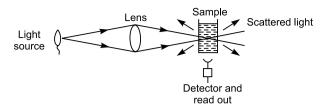


Fig. 37.2 Nephelometric analysis

#### **37.2 TURBIDIMETRY AND COLORIMETRY**

As already stated, turbidimetry is similar to colorimetry. Both involve measurement of transmitted light intensity through a medium. Both these differ in the sense that there is decrease in light intensity in turbidimetry and in coloimetry the light intensity decreases due to absorption. Both techniques may use similar or different apparatus.

#### 37.3 NEPHELOMETRY AND FLUORIMETRY

Both nephelometry and fluorimetric method involve measurement of scattered light and in this respect both are similar. However, the difference between the two is that in fluorimetry the scattering is elastic compared to inelastic in nephelometry. This implies that in nephelometry, both incident and scattered light are of the same wavelength and in fluorimetry the scattered light is of a longer wavelength than the incident light.

#### 37.4 CHOICE BETWEENNEPHELOMETRYANDTURBIDIMETRY

Choice between the two techniques (*viz.*, nephelometry and turbidimetry) depends on the amount of light scattered by suspended particles present in solution. For determining comparatively high concentration of supended particles, tubidimetry is preferred. This is because in concentrated solution extensive light scattering takes place due to the presence of many particles. However, when the suspension is less dense and decrease in power incident beam is small, nephelometry is well suited. In this case, accurate results are obtained due to small amount of scattered light which is measured against a back background.

# 37.5 BASIC PRINCIPLES OF NEPHELOMETRY AND TURBIDIMETRY

When a beam of light strikes a solution, reflection or scattering of light takes place depending on the size of the particles in solution. If the particles suspended in solution are larger than the wavelength of incident light, reflection takes place. However, if the size of the suspended particles in solution is of the same order

or smaller than the wavelength of the incident light, scattering will take place predominantly.

In nephelometry, the suspended particles should be small compared to the wavelength used. This is necessary so that scattering of light predominates. Smaller particles undergo scattering giving a symmetrical pattern of secondary rays having maximum intensity at 90° to the primary incident beam. It is due to this reason that most of the instruments for use is nephelometry involve measurement at 90° (see Fig. 37.2). In case the particle size is larger, measurements have to be made at angles smaller than 90°. However, if the particle size is too large or too small, the scattering efficiency falls.

In turbidimetry, the measurements depends on the total radiation from the primary beam and so particles larger than the wavelength of the incident light do not cause any complication.

A number of factors are responsible for the amount of radiation removed or deviated from the primary beam. These factors include concentration, particle geometry, wavelength of incident light and refractive index difference between the particle and its surrounding medium. These factors are discussed below:

(i) Concentration: As already stated turbidimetry involves measurement of transmittance of a primary beam of radiation. The transmittance of primary beam is defined as follows:

Transmittance: 
$$T = \frac{I}{I_0}$$

where  $I_0$  is the intensity of the incident light after passing through comparison cell containing solvent and I is the intensity of light after passing through the cell containing suspended particles in solution.

The transmittance is related to the concentration of the suspended material by the following equation which is similar to Beer's law.

$$S = \log \frac{I_0}{I} = kbc$$

where s is turbidence due to scattering,

b is the path length,

k is the proportionality constant known as turbidity constant, whose value depend of the particle size, wavelength and the refractive index of the suspended particles and the surrounding medium.

A calibration curve is prepared by plotting *s vs.* known concentration of scattering material. Unknown concentration are found from the calibration curve by knowing their *s* values from the tubidimeter.

In nephelometry, the scattered intensity is dependable on a number of factors like properties of scattering suspenson, the angle and geometry 534

of the instrument used for measuring. Due to this, it is not possible to relate the scattering to the concentration. In this case a equation which relates the scattered intensity,  $I_s$  to the concentration of suspended particles is useful.

$$I_{s} = K_{s}I_{o}C$$

where  $K_s$  is a emprical constant for a particular system and  $I_0$  is the incident intensity. Identical conditions are used for all measurements.

A working curve is obtained by plotting the concentration of suspended particles against  $I_a/I_a$  under controlled conditions.

- (ii) Particle Geometry: Particle shape and size are important factors both in turbidimetric and nephelometric analysis. As far as possible all samples and calibration solutions should possess the same distribution of small, medium and large particles. So both the sample solutions and standards must be prepared under identical conditions. However, this is very difficult. The conditions include concentration of reactants, temperature, mixing, pH and the order of mixing of reactants and the time for the growth of particles. In the absence of these conditions errors in turbidimetry and nephelometry are introduced.
- (iii) Wavelength of Incident Light: The wavelength of the incident light is an important factor in turbidimetry. It is best to select such a wavelength so that no absorption takes place in the sample solution. Thus, for a colourless sample solution, the incident light used is of the same colour. However, if the clear solution has dark particles, the incident light in the red or even infrared region should be used where there is maximum absorption.

Absorption is much less problem in nephelometry. In such case, as a convenience, white light is generally used.

(iv) Refractive-index Differences: As far as possible, there should be considerable refractive index differences between the particles and its surrounding medium. This gives the best results. If necessary, it may be useful to change the solvent so that the refractive-index differences increase.

#### **37.6 INSTRUMENTATION**

The special features of both turbidimeters and nephelometers are given below.

(i) Source of Light: It is advantageous to use monochromatic radiation (in nephelometer white light can also be used). The monochromatic radiations minimise absorption in turbidimeters. However, in any case,

high intensity monochromatic radiations is used and if possible short wavelengths are used, which increase efficiency of Rayleigh scattering. A most convenient source is a mercury arc or a laser with suitable filter combinations in order to isolate one of its emission lines. However, for determination of the concentration of a particular material, it is best to use a polychromatic source such as tungsten lamp. In the later case, best results are obtaind by using blue spectral region and using a filter for blocking other wavelengths.

(ii) **Detectors:** Since the intensity of scattered readiation is normally very small in naphelometry, so in nephelometry a photomultiplier tube is used as a detector. The detector is generally fixed at 90° to the primary beam in nephelometers. However, it is desirable to vary the detector angle (which is normally close to the primary beam for maximum versatility and sensitivity. In some naphelometers, the detector is mounted on a circular disc. This permits measurement of a number of angles, *i.e.*, at 0° and from 30° to 135°. In this case the outer edge of the circular disc is graduated in degree so that it can be read from outside.

In tubidimeters, ordinary detectors like phototubes are conveniently used.

(iii) Cells: The cells to be used should minimise reflections and multiple scattering. Generally a cell with a rectangular cross section is used. For measurements at angles other than 90°, semioctagonal cells (Fig 37.3) and used. The octagonal faces (Fig. 37.4) permits measurements to be made at 0°, 45°, 90° or 135° to the primary beam. The walls of the cell through which light beams do not pass are painted black in order to absorb unwanted radiation and minimise any stray radiation.

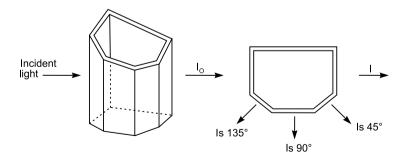


Fig. 37.3 Semioctagonal cell Fig. 37.4 (Octagonal faces of a semioctagonal cell)

The instruments used in turbidimetry and nephelometry are discussed below.

#### 37.6.1 Turbidimeters

Different types of tribidimeters are available for use. A common and interesting turbidimeter is the well known Du Pont model 430. This is considerably more sensitive to low concentrations of suspended particles than the ordinary turbidimeters. The Du Pont model 430 instrument (Fig. 37.5) is a double-beam instrument. For its operation, it depends on the relative degree of polarisation of transmitted and scattered light.

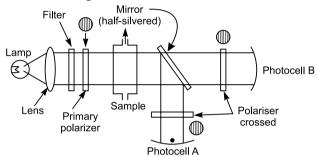


Fig. 37.5 A tubidimeter (Du Pont Model 430)

In the above tubidimeter (Du point model 430) (Fig. 37.5), the scattering by suspended particles in solution causes changes in the plane of polarisation of light. The beam of light from the lamp passes through a primary polarizer, which causes the incident beam to be plane-polarised, which in turn passes through the sample solution. The resultant beam gets splitted into two parts with the help of half-silvered mirror. These are detected by two separate photocells A and B. In case the sample solution does not have suspended particles, photocell A shows maximum response and the photocell B shows zero or minimum response. The measure of concentration of suspended particles is obtained from the ratio of two signals is photocells A and B. Increase in concentration of suspended particles, there is increase in the response of photocell B and decrease in the response of photocell A. So, the ratio of the two is a measure of the turbidity.

The Du Pont model 430 tubidimeter cannot be used in case solution contains optically active substances.

## 37.6.2 Nephelometers

Ordinarily, fluorimeters can be used for nephelometric measurements. Even spectrophotomers can be used as nephelometers in some cases. A typical precision nephelometer which works at low intensities is shown in Fig. 37.6.

In the nephelometer (Fig. 37.6), the multiplier phototube used as a receiver is mounted on a turnable so that it can be positioned at any desired angle (0 to 180°) relative to the exit beam. However, in most of the nephelometric measurements

it is positioned at 45° or 90° to the primary beam. The undeviated beam passed into a tube at the back called a light trap. This nephelometer can be used for determination of size of particle, its shape and molecular weight in addition to the usual nephelometeric measurements.

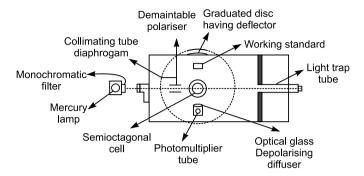


Fig. 37.6 A pricision Nephelometer

#### **37.7 APPLICATIONS**

Turbidimetric and nephelometric methods have been used for finding the suspended material in liquids in nature and in manufacturing processes. These methods find applications in water treatment plants, sewage treatment processes, in steam generating plants, petroleum refining and bottling industries. Some of the important applications are given below:

(a) Inorganic Analysis: It has been found that in a number of cases, the precipitates due to their colloidal nature are difficult to filter making gravimetric estimations difficult. In such cases nephelometry and turbidimetry can be conveniently used by converting the precipitates into ideal suspension under controlled conditions. This is necessary since the scattering of light depends on the size and number of particles as well as their concentrations.

For usual quantitative analysis, it is necessary to prepare callibration curves using known metal concentrations. The amount of suspensions of unknown solutions are obtained by referring to the calibration curves.

Using nephelometry and turbidimetry, it is possible to determine sulphate and carbonate as BaSO<sub>4</sub>, BaCO<sub>3</sub> respectively. Also, chlorides, fluorides, cyanides and calcium are determined as AgCl, CaF<sub>2</sub>, AgCN and calcium oxalate respectively. The determination of sulphur in Coke, Coal, oils, rubbers, plastics etc. are useful applications of nephelometry and turbidimetry. The procedure used for the determination of sulphur involves its conversion into sulphate and subsequent shaking with NaCl and excess of solid BaCl<sub>2</sub> to get a suspension of BaSO<sub>4</sub>, which is then subjected to nephelometry or turbidimetry as the case may be. Finally, the concentration of suspension is determined from the calibration curve.

For the determination of CO<sub>2</sub>, the gas is bubbled through an alkaline solution of a barium salt and the formed BaCO<sub>3</sub> in suspension is determined by nephelometry or turbidimetry. Phosphate can be estimated in as low concentration as 1 part in more than 300 million parts of water as a precipitate with strychnine molybdate reagent. Also ammonia in concentration of 1 part in 160 million parts of air can be detected by addition of Nesslers reagent. Some typical turbidimetric and nephelometric methods are given in Table 37.1.

Element	Methods	Suspension	Reagent	Interference
Ag	N, T	AgCl	NaCl	_
As	Т	As	K <sub>2</sub> H <sub>2</sub> PO <sub>2</sub>	Se, Te
Au	Т	Au	SnCl <sub>2</sub>	Ag, Hg, Pd, Pb,
				Ru, Se, Te
Ca	Т	CaC <sub>2</sub> O <sub>4</sub>	$H_2C_2O_4$	Mg, Na, SO <sub>4</sub> <sup>2-</sup>
CI-	T, N	AgCl	AgNO <sub>3</sub>	Br, I <sup>-</sup>
Se	Т	Se	SnCl <sub>2</sub>	Те
Те	Т	Те	NaH <sub>2</sub> PO <sub>2</sub>	Se

**Table 37.1** Determination of some typical metals by tubidimetric and nephelometric methods

T = Turbidimetric, N = nephelometric

- (b) Organic Analysis: Turbidimetric procedure can be used for determination of turbidity in sugar products, citrius Juices etc. Determination of benzene in alcohol can be done by dilution with water in order to get an immiscible suspension.
- (c) Biochemical Analysis: An important application in biochemical analysis is measurement of growth of bacterium in nutrient median and finding the amounts of amino acids, vitamins and antibiotics. Determination of proteins, yeast, glycogen and beta and gamma globurin in blood and plasma can also be done.
- (d) Environmental Pollution: Continuous monitoring of air and water pollution can be done by turbidimetry and nephelometry. In air, dust and smoke are monitered and in water turbidity is monitored.

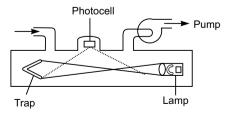


Fig. 37.7 Nephelometric analyser for atmospheric particulates

In the atmosphere, smokes and fogs are made visible due to scattering of light. An instrument for measuring these are used in monitoring atmospheric pollution (Fig. 37.7).

(e) Turbidimetric Titrations: In turbidimetric titrations, the absorbance is plotted against the volume of titrant added. As the volume of titrant increases, the concentration of precipitate increases resulting in increase in absorbance. When the precipitation is complete, the absorbance becomes constant and there is abrupt change in the slope indicating the end-point (Fig. 37.8).

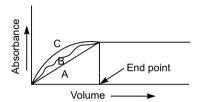


Fig. 37.8 Turbidimetric titration curves

In the above titration, curve A is an ideal curve. The other two curves, B and C result from precipitates with mixed particle size and poor stirring etc. In case of curves B and C the detection of end point is complicated. Some examples of turbidimetric titrations include determination of  $SO_4^{2-}$  as  $CaSO_4$ ,  $Ag^+$  as halide  $Ca^{2+}$  as oxalate. These titrations can be used in the concentration range  $10^{-5}$  to  $10^{-6}$ .

The apparatus used is simple. It consists of a light source and a photocell placed on the opposite side of the titration vissel.

(f) Phase Titrations: Turbidimetry has been successfully used for the titration of mixture of two liquids by a third liquid, which is miscible with one and immiscible with the other. Thus, addition of a sufficient quantity of the third liquid results in separation of phase causing turbidity. The results can be interpreted on the basis of phase diagram of three-component system or by titrating unknown solution with a known mixture. The titration curve for titration of water-pyridine by chloroform is shown in Fig. 34.9.

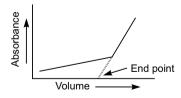


Fig. 37.9 Titration curve of titration of water-pyridine by chloroform

(g) Molecular Weight of High Polymers: The procedure for the determination of molecular weight of high polymers involves the measurement of the intensity of

light scattered by solution of polymers. The relationship given below holds good between molecular weight and turbidity.

$$\underset{C \to O}{\text{Limit}} \frac{HC}{\text{Turbidity}} = \frac{1}{M}$$

H is a constant for a given polymer and a dispersion medium.

C is the concentration of the solution (g/mL) and turbidity is the fraction of incident light scattered per cm length of the solution thought which it passes.

For the determination of the molecular weight of a polymer, turbidity is measured at different concentrations of its solution in a suitable solvent. Finally, the plot of (HC/turbidity) on extrapolation to zero concentration (as shown in Fig. 37.10) give the value of 1/M.

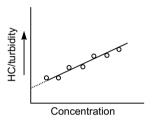


Fig. 37.10 Plot of HC/turbidity against concentration of polymer in solution



- 1. Give the basic principles of nephelometry and turbidimetry.
- 2. Compare turbidimetry with calorimetry and nephelometry with fluorimetry.
- 3. Discuss the comparison of nephelometer, turbidimeter and spectrophotometer.
- 4. How with you decide whether nephelometry or turbidimetry procedure has to be used in a particular case?

- **5.** Discuss the instrumentation for a turbidimeter and a nephelometer.
- **6.** Write note on:
  - (a) Turbidimetric Titrations
  - (b) Phase titrations
  - (c) Determination of molecular weight of High Polymers.



# Chapter 38

# Refractometry and Interferometry

#### 38.1 INTRODUCTION

The **refractive index**, a useful physical characteristic of liquids, is used for their identification. Thus, comparison of the experimentally measured refractive index with the literature value for a pure substance is used to determine the purity of the liquid. The liquid sample under examination is considered to be pure if the measured refractive index is closer to the literature value.

The refractive index (n) of a substance is defined as the ratio of the velocity of light in air (or vacuum) to that in the substance

$$n = \frac{\text{Velocity of light in substance}}{\text{Velocity of light in air}}$$

When a ray of light passes from air into a liquid, its direction changes. This change of direction is

called refraction (Fig. 38.1).

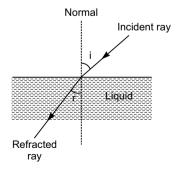


Fig. 38.1 Refraction of light

The refractive index of the liquid with respect to air is given by **Snell's law**. According to this,

$$n = \frac{\sin i}{\sin r}$$

where i is the angle of incidence and r is the angle of refraction.

The refractive index of a liquid depends on the temperature of the liquid and the wavelength of the incident ray. Firstly, the density of a medium changes with change of temperature and so the speed of light in medium also changes. Secondly, beams of light with different wavelengths are refracted differently in the same medium thus giving different refractive indices for the medium. Generally, the refractive indices are measured at 20° using a sodium discharge lamp as an illumination source. The sodium lamp is known to give yellow light of 589 nm wavelength. Thus, the refractive index is represented by the symbol.

Since the refractive index is a ratio, it has no units.

The refractive index is reported in four decimal places. However, it is sensitive to even very small amount of impurity. So the substance must be extensively purified failing which it is not possible to reproduce the last two decimal places given in literature.

#### 38.2 SPECIFIC ROTATION

On the basis of theoretical considerations, Lorenz and Lorenz (1880) derived the following relation for the refractive power of substance

$$R_s = \frac{n^2 - 1}{n^2 + 2} \cdot \frac{1}{d}$$

where  $R_s$  is the specific rotation, d the density and n the refractive index. The value it  $R_s$  is constant at all temperatures.

#### 38.3 MOLAR REFRACTION

The molar refraction  $(R_M)$  is an additive property. Thus, molar refraction of a molecule is the sum of molar refraction of its constituent atoms. Using molar refraction, it is possible to determene the constitution of a compound. The molar refraction values are calculated for various possible structures and the formula in accordance with the calculated molar refraction is the correct structure (or formula) of that compound. The molar refraction values for some atoms groups and bonds are given in Table 38.1.

Table 38.1         Molar refraction of some important atoms, groups and bonds					
Atoms	R <sub>M</sub>	Group	R <sub>M</sub>	Bonds	R <sub>M</sub>
С	2.42	_C=O	4.60	—C—C— double bond	1.73
Н	1.10	—ОН	2.55	_C≡C—	2.40
CI	5.97	—СООН	7.25	(triple bond)	
Br	8.86	—NO <sub>2</sub>	4.71	3 membered ring	0.48
I	13.90	—CH <sub>3</sub>	5.65	4 membered ring	0.71
O in OH	0.53	—C <sub>2</sub> H <sub>5</sub>	10.30	5 membered ring	0.19
O in R – OR	1.64	(Increase for every CH <sub>2</sub> )	4.65	6 membered ring	0.15
O in C = O	2.21	—C≡N	5.46		
N(Primary aliphatic amine	2.322	NO <sub>2</sub> (aromatic)	7.30		
N (sec. aliphatic amine)	2.499				
N (tert. aliphatic amine)	2.840				
N (Primary aromatic amine)	3.21				
N (sec. aromatic amine	3.59				
N (tert. aromatic	4.36				

Let us consider the example of ethyl alcohol, (CH<sub>3</sub>CH<sub>2</sub>OH). Its refractive index at 295.9 K for D-line is 1.3611 and its density is 0.7885g cm<sup>3</sup>. The molar mass of ethyl alcohol is 46 (2  $\times$  12 + 1  $\times$  6 + 1  $\times$  16). The molar refraction is calculated by using the relation

$$R_M = \frac{n^2 - 1}{n^2 + 2} \cdot \frac{M}{d}$$

amine) N (amide)

2.65

$$= \frac{(1.3611)^2 - 1}{(1.3611)^2 + 2} \times \frac{46}{0.7885}$$
$$= 12.9105 \text{ cm}^3 \text{ mol}^{-1}.$$

The computed value of molar refraction on the basis of values given in Table 38.1 is 12.916 cm<sup>3</sup> mol<sup>-1</sup>.

Contribution of 6 hydrogens = 
$$6 \times 100 = 6.600$$
  
Contribution of 2 carbons =  $2 \times 2.418 = 4.836$   
Contribution of O in OH group =  $1.525$   
Total Contribution =  $1.2916 \text{ cm}^3 \text{ mol}^{-1}$ .

As seen, the computed value is in agreement with the value calculated above. So the correct structure is CH<sub>3</sub>CH<sub>2</sub>OH.

The molar refraction  $R_m$  of a solution is given by

$$R_M = \frac{n^2 - 1}{n^2 + 2} \left( \frac{x_1 M_1 + x_2 M_2}{d} \right)$$

where  $x_1$  and  $x_2$  are mole fraction of the solvent and solute with molecular masses  $M_1$  and  $M_2$  respectively, n is the refractive index of the solution and d is the density of the solution.

The molar refraction of a solid is determined by dissolving the solid in a suitable solvent so that a solution of known concentration is obtained. Finally, the molar refraction is determined as in the case of liquid.

#### 38.4 DETERMINATION OF REFRACTIVE INDEX

The refractive index of a liquid is determined using an instrument called **Abbe Refractometer** (Fig. 38.2).

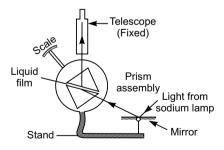


Fig. 38.2 Abbe Refractometer

A thin film of the liquid is placed between the two prisms. Light from a sodium lamp falls on the lower side of the lower prism with the help of a mirror. The hypotenuse surface of the lower prism is ground, and so light enters the liquid

at all angles of incidence. No ray can enter the upper prism with greater angle of refraction than the grazing incidence (*i.e.*, atom angle slightly less than 90°). Thus, a view in the telescope appears to be divided into two bands, one bright and one dark. The prism assembly on rotation with the help of a side knob till the cross wire of the telescope coincides with the edge of the bright band (Fig. 38.3) A pointer attached to the prism assembly indicates the refractive index on the callibrated scale.



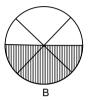


Fig. 38.3 A. Refractrometer incorrectly adjusted B. Refractrometer correctly adjusted

#### 38.5 APPLICATIONS OF REFRACTOMETRY

Refractometry has been used for qualitative and quantitative analysis and also for structural investigations.

It has been found that in a homologues series the molar refraction increases in regular increment with the number of carbon atoms. Thus, molecular refraction of a compound is considered to be sum of atomic increments. The contribution of each carbon is approximately the same in all molecules. As an example, the atomic increments for a saturated carbon, non-polar hydrogen and an ether hydrogen are 2.42, 1.10 and 1.64 respectively (*see* Table 38.1).

Thus, the calculated molar fraction for diethyl ether ( $C_2H_5OC_2H_5$ ) is

$$4 \times 2.42 + 10 \times 1.10 + 1 \times 1.64 = 22.32$$

The molar refraction values of a few important atoms, groups and bonds are given in Table 38.1. The applications of molar refractivity are given below using suitable example:

(i) Kekule's structure of benzene.

Benzene, C<sub>6</sub>H<sub>6</sub> contains three double bonds. Molar refraction due to

6 carbon atoms  $6 \times 2.42 = 14.52$ 

6 Hydrogen atoms  $6 \times 1.10 = 6.60$ 

3 double bond	$3 \times 1.73 = 5.19$
1 six-membered ring	$1\times0.15  =  0.15$
	26.16

The experimental value of molar refraction 25.95 close to the calculated value support the kekule structure of benzene.

(ii)	Acetylene dibromide	BrCH	BrCH = CHBr	
	Two carbons	$2 \times 2.42$	= 4.84	
	Two hydrogens	$2 \times 1.10$	= 2.20	
	Two bromine	$2 \times 8.86$	= 17.62	
	One double bond	1 × 1.73	= 1.73	
			26.89	

On the basis of observed value, the formula of acetylene dibromide is correct.

(iii) Differentiation between structures having the same molecular formula. As an example methyl formate and acetic acid have the same molecular formula C<sub>2</sub>H<sub>4</sub>O<sub>2</sub>. These can be identified on the basis at their molar refraction

Acetic acid CH₃COOH		Methyl formats HCOOCH <sub>3</sub>	
2C	4.836	2C	4.836
4H	4.400	4H	4.400
1 carbonyl oxygen	2.211	1 carbonyl oxygen	2.211
1 hydroxyl oxgen	1.525	1 ester oxygen	1.643
	12.972		13.090

The difference between the two structures is clear in the molar fraction values.

(iv) It is possible to determine the composition of solution of sodium chloride and potassium chloride. A curve is drawn by plotting the percentage of sodium chloride against index of refraction. By this procedure a fair degree of accuracy is obtained.

## 38.6. OPTICAL EXALTATION

In case of compounds containing two or more double bonds in conjugation, as in —CH—CH—CH—CH—, the molar refraction is much higher than that calculated from the data in Table 38.1.

The difference between the observed and calculated values is called optical exaltation. As an example, the isomeric hydrocarbons,  $C_6H_{10}$ , which have two types of double bonds (conjugated and unconjugated) have different  $R_M$  values.

	R <sub>M</sub> (Observed)	R <sub>M</sub> (Calculated)
CH <sub>2</sub> =CH-CH <sub>2</sub> -CH <sub>2</sub> -CH=CH <sub>2</sub> unconjugated	28.77	21.99
CH <sub>3</sub> CH—CH—CH—CH <sub>3</sub> conjugated	28.87	28.99

Conjugated double bonds in a ring compound (e.g., benzene) do not exhibit optical exaltion.

It has also been found that the presence of a carbonyl group along with a double bond or a triple bond also causes optical exaltation. As an example, phonone is experementally found to have molar refraction 45.39 compared to calculated value of 42.85

(CH<sub>3</sub>)<sub>2</sub>C = CH − CO − CH = C (CH<sub>3</sub>)<sub>2</sub>

Phonone

$$R_M$$

experimental value calculated value

 $45.39$  42.85

∴ Optical exaltion =  $45.39 - 42.85 = 2.54$ .

## 38.7 INTERFEROMETRY

It has already been stated that refractometry depends on the inteferance (diffraction) of light on passing through a medium (solution). However, in interferometry there is no diffraction. The light enters and leaves the solution at right angles. The instrument used is called **interferometer**. In a simple form, an interferometer is represented as in Fig. 38.4.

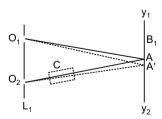


Fig 38.4 Optical Principle of an interferometer

The method consists in passing parallel light through two small openining  $O_1$ , and  $O_2$  (as shown in Fig. 38.4) In this case  $O_1$  A and  $O_2$ A are of equal lengths and two beam reach at A in phase resulting in a bright spot. At other points on the screen

548

 $(y_1y_2)$ , the two beams are unequal. Thus, at some point  $B_1$ , there is a difference of half a wavelength in the two beams. A dark spot is produced at  $B_1$  due to the interference of the two beams. A little point further along  $y_1$ , the difference is one wavelength resulting in the formation of a bright spot. The succession of light and dark spots (maximum and minimum) continues indefinitely with monochromatic light. If a substance of slightly greater refractive index is placed at C. The optical length of the beam  $O_2A$  increases by an amount  $\Delta b$  due to velocity of light being decreased through C. The magnitude of this increase is dependent on the thickness of the sample and its refractive index.

$$\Delta b = b(n - n_0)$$

where

b is the thickness of the sample n is the refractive index of sample  $n_0$  is the refractive index of medium (air).

The velocities of light in two media are proportional to there refractive index.

In the later case when the substance is placed at C, the two beams will not arrive at A, but at some other point A', which is optically equidistant from  $O_1$  and  $O_2$ . The band system shifts by this amount (A - A'). For a light of wavelength  $\lambda$ , the distance between A and A', measured in number of fringes, N (each made up of a dark and a light band) is

$$N=rac{\Delta b}{\lambda}$$
 or 
$$N=rac{b(n-n_0)}{\lambda}$$

If N is more than 1, all bands will be alike, and so it is impossible to tell how many whole number of bands greater it is. This problem is overcome by using white light in place of monochromatic light. In the later case, the central band is the only one which is white. Due to different wavelengths that make up white light, the band on either side of the maximum of the first order are fringed with blue towards the centre of the system and red along the outer edge. After about six bands, the diffusion is great and the rest of the field again becomes uniformly white. Thus, when a substance at C is placed in the path of one of the beams, it is possible to determine the value of b ( $n - n_0$ ) by counting the number of bands which the central band has been shifted. Any of these terms can be calculated knowing the other terms.

In case two plates of equal thickness are placed in the two beams, the number of bands that the central band shifts could be used to calculate the refractive index of one of the plates if the value of the other one is known. The interferometer is used not for the measurement of the index of refraction but for comparison and measurement of the concentrations of solutions and gases.

One type of interferometer is shown in Fig. 38.5.

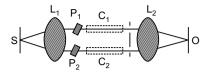


Fig. 38.5 Optical schematic representation of a type of interferometer

In this inteferometer, the optical length of the two beams is equalized by means of a glass plate in the path of the two beams  $P_1$  and  $P_2$  at an angle of approximately 45° to the beam. One of the plate is fixed and other plate is attached to a lever so that it can be rotated in order to increase or decrease its effective thickness. A micrometer screw is used to measure the movement. The micrometer screw is turned until the central achromatic bands of the two systems correspond meaning that the optical path of the two beams is of the same length. This instrument is capable of measuring quantities of gases like  $CO_2$  and  $CH_4$  present in air in amounts as low as 0.02%.

## 38.7.1 APPLICATIONS OF INTERFEROMETER

The use of interferometer in analysis of gases has already been stated. It can also be used to determine the permeability of baloon fabrics to helium and hydrogen.

Using a 1-m chamber it is possible to determine 0.02% of CO<sub>2</sub> or CH<sub>4</sub> in air.

Interferometer can also be used for the investigation of sea water and chart ocean currents and also for the analysis of dilute solutions used for the determination of freezing point.

Determination of potassium and sodium in a mixture of their chlorides or sulfates can be achieved with a precision of  $\pm$  0.1 mg in a 50 mg sample. The procedure consists in dissolving the mixture in 200 times its weight of water and comparison with a standard solution of pure potassium sulfate dissolved in 200 times its weight of water and making a calibration curve.

Interferometer has also been used for analysis of sewage and milk. In biological problems like measurement of serums,  ${\rm CO_2}$  in blood, ethyl alcohol in blood and concentration of heavy water.



- 1. Explain the term Refractive index. How it is represented?
- 2. Define specific Rotation and Molar Refraction.
- **3.** How is refractive index determined?
- **4.** What are the applications of Refractometry?

**5.** An organic compound is found to have a molar refraction of 12.972. Find out whether the compound is acetic acid or methyl formate (use the values given in Table 38.1).

Ans. Acetic acid

6. Calculate the molar refraction of carbon tetrachloride. Given refractive index of CCl₄at 20° is 1.45 73 and density at 20° is 1.595g cm<sup>-3</sup>.

**Ans.** 26.51

7. If the molar refraction of a compound is 16.97. Find whether it is allyl alcohol or acetone.

Ans. Allyl alcohol.

- **8.** What do you understand by optical exaltation giving two examples?
- **9.** Calculate the specific refraction and molar refraction of the following compounds:

	$n_D^{20}$	$d^{20}$
Benzene	1.4979	0.879
Ethanol	1.3590	0.788
Nitrobenzene	1.5524	1.21

10. The refractive index of a solution of  $D_2O$  and  $H_2O$  is 1.32980. Calculate the per cent of  $D_2O$ .

Given  $D_2O$ ,  $n_D^{20} = 1.32830$ ; and for water  $n_D^{20} = 1.33280$ .





# Chapter 39

# X-Ray Methods

## 39.1 INTRODUCTION

Three types of *X*-ray method, *viz.*, *X*-ray absorption, *X*-ray fluorescence and *X*-ray diffraction methods are normally available.

- (i) X-ray Absorption Methods: This method consists in passing a beam of X-rays through a sample under investigation. A fraction of X-ray photons are absorbed. This is proportional to the concentration of the absorbing substance. This method finds application in the detection of imperfection in the internal structure of the substance through which it passes. As compared to other X-ray methods, these have least applications.
- (ii) X-ray Diffraction Methods: These methods are based on the scattering of X-rays by crystals. These methods find application in identification of the crystalline structure of solid compounds. These methods are much more important than the X-ray absorption methods or X-ray fluorescence methods.
- (iii) X-ray Fluorescence Methods: The X-rays in these methods are generated within the sample. By measurement of the wavelength and intensity of the generated X-rays, it is possible to perform qualitative and quantitative analysis.

# 39.2 THEORETICAL CONSIDERATION

X-rays were discovered by German Physicist while investigating the effect of cathode rays, on a fluorescent screen. During investigations, Roentgen observed an invisible type of radiation of very short wavelength, which he called X-rays. For the discovery of X-rays, Roentgen won Nobel Prize in Physics in 1901.

X-rays are penetrating electromagnetic radiation, usually generated by accelerating electrons to high velocity and suddenly stopping them by collision with a solid metallic body. The wavelength ranges from about 10<sup>-5</sup> angstrom to  $10^{-3}$  angstrom. Earlier X-rays were used for diagnosis for the detection of fractures and cracks in bones. In Industry, these are used for testing welded metal Joints. The X-rays are also known as X-radiation and Roentgen rays. The generation of X-rays can be understood by considering the Bohr's theory of atomic structure.

It is known that each atom consists of a dense positively charged nucleus made up of neutrons and Protons and that negatively charged electrons orbit at different energy levels (called electron shells) surrounding the nucleus. The different shells are known as  $K, L, M \dots$  shells. As an example, a sodium atom contains completely filled K and L shells (2 and 8 electrons respectively) and one electron in the next M shell. The electronic structure of an atom is shown in Fig. 39.1.

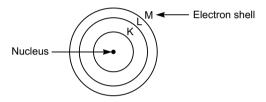


Fig. 39.1 Electronic Structure of an atom

A fast moving electron on striking an atom knocks out an electron from one of the inner shells of the atom. Due to the loss of one inner-shell electron, one of the electron from the outer shell falls into the vacated orbit, simultaneously emitting an X-ray photon. The energy of the emitted X-ray photon is found to be equal to the difference in energy between the two levels involved. Thus, as an example, if a K-shell loses one electron and is replaced by an electron from the L-shell, the resulting X-ray is called KX-ray and its energy  $(E'_k)$  is given by the expression

$$E'_k = E_L - E_K$$

where  $E_L$  and  $E_K$  are the energies of L- and K- shell electrons respectively.

These K lines are further subdivided into  $K_{\alpha}$  and  $K_{\beta}$  depending on whether the electron ejected is replaced by electron from the K shell is from the closest shell or from the next shell (*L* or *M* shell).

In a similar way, when an electron from L shell loses one electron and is replaced by an electron from the outer shell. In this case, the resulting X-ray is called L X-ray. These L-lines (as in the case it K lines) are further divided into  $L_{\alpha}$ and  $L_{\rm R}$  depending on whether the electron ejected (from L shall) is replaced by electron from the closest shell or from the next nearer shell (M or N shell)

Thus, in case an electron is lost from K shell, it may be replaced by an electron from L or M shell. The electrons on descending from the shell is expected to emit X-rays with a frequency.

$$v = \frac{E_L - E_K}{h}$$

The lines originating from transitions between K and L shells are designated as K lines. There is, however, a slight difference in the energy levels of different electrons in the L-series. This results in the emission lines at different frequencies. These lines are designated  $K_{\alpha 1}$  and  $K_{\alpha 2}$ .

Electrons descending from M shell to L shell are expected to emit X-rays with frequency

$$v = \frac{E_M - E_L}{h}$$

These transition emit L lines.

In case primary X-rays are used for bombarding a target element, a series of sharp characteristic lines are obtained (as discussed above). However, bombardment with high-energy electrons a complicated spectrum of X-rays with sharp spectral lines superimposed on the continuum is obtained.

The X-rays intract with matter by absorptions, scattering or diffraction.

(a) Interaction of X-rays by absorption: On passing a beam of X-rays through matter, the energy (of X-rays) are lost partly by absorption and partily by scattering; the latter process is a loss of small part of the X-rays.

In the former case, *i.e.*, absorption of *X*-rays, the electrons of the atoms in the matter absorb energy from the *X*-rays and inturn get excited. The excited electrons emit secondary radiation, which are characteristic of these atoms.

(b) Interaction of X-rays by Scattering and diffraction: When a beam of X-rays strikes a substance its atoms behave as small oscillators. These atoms oscillate at the same frequency as that of the beam of striking X-rays and emit electromagnetic radiations in all directions. The frequency of the emitted electromagnetic radiation is the same as that of the beam of striking X-rays. The scattered waves originate from electrons, which as we know, are arranged is a regular manner in a crystal lattice and then travel in certain directions. It can be said that the scattered waves are diffracted by the crystal plane. The scattered X-rays are characteristic of the substance under examination.

The study of crystal structure by X-ray diffraction technique is called **X-ray crystallography**. Thus, when X-rays strike crystals, they get scattered, the intensity and the direction of the scattered rays are dependable on the distance between the atoms in the crystal. An analysis of the image produced as a result of interference of diffracted rays reval the crystal structure.

# (c) Fluorescence

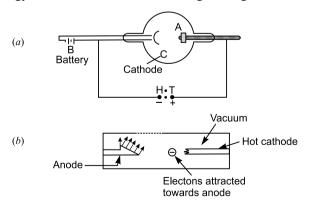
Some times, irradiation of a sample with an X-ray beam emits other X-ray beams, whose wavelength is different from the incident X-ray beam. This process is known as fluorescence. The wavelength of the X-ray fluorescence is used to determine what elements are present in the sample. The amount of the element is determined by the intensity of the emitted X-rays. This technique provides a method for elemental analysis, which is independent of the molecular structure.

# 39.3 INSTRUMENTATION

As already stated, X-ray spectroscopy constitutes three main fields, viz, X-ray absorption, X-ray diffraction and X-ray fluorescence. In all these the optical components of the equipment are the same but the optical system varies. The main components are:

(i) Production of X-rays: We know now that X-rays are generated when high velocity electrons strike a metal surface. On an average, about 1 per cent of the total energy of electron beam is converted into X-radiation and the remaining energy is dissipated as heat. A number of different types of X-ray tubes are available for the production of X-rays. A typical tube along with schematic representation of an X-ray source are shown in Fig. 39.2.

As seen a cathode is a filament of tungsten. It is heated by a battery B to generate thermionic electrons, which constitutes the cathode ray stream. On Keeping a positive ray voltage in the form of anode, which has a target A near these electrons, the electrons in turn are accelerated towards the target. As the electrons strike the target, these transfer their energy to the metallic surface resulting in the generation of X-rays.



**Fig. 39.2** (a) A typical tube (Colidge tube) for the production of X-rays; (b) Schematic representation of a X-rays source

The choice of the target (A) material is dependable on the sample to be analysed. Generally, a target material should have atomic number greater than the atomic number of the element to be examined in the sample. Also, the energy of X-rays emitted by the target material should be greater than that required for the excitation of the elements to be irradiated.

It is observed that generally the target gets hot during use, In order to solve this problem, the tube is cooled with water. Alternatively, the target is rotated at high speed in order to reduce the production of localised heating. The alternative procedure permits the generation of X-rays of intensity higher than that obtained from the stationary target. Use of a spherical electrostatic lens within the electron gun of the X-ray tube in helpful to produce electron beam less than 1 micron in diameter, which is necessary for X-ray analysis.

(ii) Collimator: A collimeter is helpful to get a narrow beam of X-rays. The X-rays generated as explained in (i) above are passed through a collimator, consisting of two sets of closely packed metal plates which are separated by a small gap. All the X-rays except the narrow beam that passes through the gap are absorbed. A typical collimator assembly for achieving a narrow beam of X-rays is shown in Fig. 39.3.

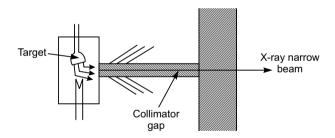


Fig. 39.3 A typical collimator assembly

(iii) Filters: Filters are used to obtain a monochromatic beam. A filter is normally a window of a suitable material which absorbs the undesirable radiation but permits radiation of required wavelength to pass through. For example, a Zirconium filter is used for molybdenum radiation. On passing X-rays emitted from molybdenum through zirconium filter, the radiation of molybdenum at short wavelength are strongly absorbed by zirconium, but the  $K_{\alpha}$  lines of molybdenum are weakly absorbed. Thus, Zirconium allows  $K_{\beta}$  lines to pass. Table 39.1 gives some suitable filters for some target elements.

		'	
Target element	β <b>-Filter</b>	Thickness	Percentage loss in $\mathbf{K}_{\alpha}$ activity
Со	FI	0.012	39
Cu	Ni	0.015	45
Fe	Mn	0.011	38
Мо	Zr	0.081	57
Ni	Со	0.013	42

**Table 39.1** β-Filters for some common Target elements to give an Intensity Ratio  $K_g/K_α$  of 1/100

Alternatively, crystals can be employed as a grating monochromator in order to obtain greater degree of monochromaticity.

(*iv*) **Detectors:** The intensity of *X*-rays are measured and recorded by photographic or counter methods.

**Photographic Methods**. In these methods, a plain or cylindrical photographic film is used to record the position and intensity of the X-ray beam. After exposing the film to X-rays, it is developed. The blackening of the developed film is expressed as

$$D = \log \frac{I_0}{I}$$

Where D is expressed in density units, represents the blackening of the developed film,

 $I_{\rm o}$  and I refer to incident and transmitted intensities of X-rays.

The quantity of D is related to total X-ray energy that is responsible for the blackening of the photographic film. D is measured by a densitometer.

The method reveals the entire diffraction pattern on a single plate and so is used in diffraction studies. However, the method takes more time. Also, the densitometer operation is time consuming and is a cause of considerable error. So, this method is seldom used in quantitative measurement.

#### **Counter Methods**

Different types of counters are a available. A Geiger-Mullar tube counter is commonly used. It consists of a Geiger tube filled with an inert gas like argon. A central wire anode is maintained at a positive potential of 800–2500V. The ray on entering the Geiger tube collides with the inert gas filling resulting in the production of an ion pair—the produced electrons moving towards the central anode and positive ion moving towards the outer electrode. Due to the potential gradient, the electron is accelereated causing the ionization of a number of argon atoms. This

results in the production of an avalanche of electrons travelling towards the central anode. The net result is the out pulse of 1 to 10V which is measured easily using a simple circuit (Fig. 39.4).

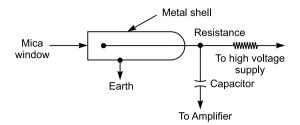


Fig. 39.4 A Geiger tube-schematic representation

The Geiger tube is cheap and gives higher signal for a given *X*-ray intensity.

## 39.4 INSTRUMENT FOR X-RAY ABSORPTION

The instrument (Fig. 39.5) has a tungsten X-ray tube, which operates at 15–45KV. A synchronous motor-driven chopper, kept between cell and attenuator alternately interrupts half of the X-ray beam. A variable—thickness aluminium attenuator is kept between the chopper and reference sample compartment. Both halves of X-ray beam are allowed to fall on a phosphor-coated photomultiplier tube, protected from visible light by a thin metallic filter.

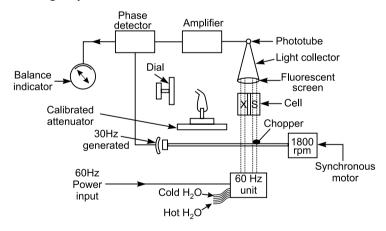


Fig. 39.5 X-ray Absorptiometer

The procedure involves following steps:

- (i) The reference sample is taken in the reference cell and the sample to be analysed is kept in the sample cell.
- (ii) The attenuator is adjusted till the absorption in the two *X*-ray beams is brought into balance.

The change in thickness of aluminium (required for various samples) is a function of difference in composition.

# **Application**

X-ray absorption finds application to determine a sample containing one element markedly heavier than the others. This method has been used to determine chlorine in hydrogen, plastics and hydrocarbons. Also it has been used to determine barium fluoride in carbon and barium/lead in special type of glasses.

## 39.5 INSTRUMENT FOR X-RAY DIFFRACTION

The X-ray diffraction procedures are used for the investigation of internal structures. A number of methods are used, some of which are given below:

# 39.5.1 Laue Method

The equipment for Laue Method (Fig. 39.6) consists of a source (A) of X-rays, which emits a beam of continuous wavelength (known as white radiation) obtained from a target (made of tungsten) at about 60,000 volts. The X-rays obtained from the source are passed through pinhole collimator (B) giving a fine pencil of X-rays. A smaller diameter of the pinchole collimator gives sharper interference. The X-rays from B pass through a crystal (C) whose internal structure is under investigation. The orientation of the crystal is achieved by setting it on a holder. The X-ray which pass through the crystal strike film (D) which is fixed on a rigid base and records the diffraction pattern as shown in Fig. 39.7.

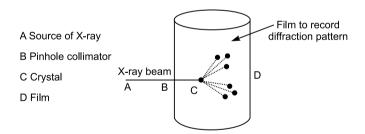


Fig. 39.6 Equipment for Laue's transmission method

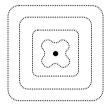


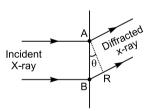
Fig. 39.7 Spots in Laue photograph of a simple cubic crystal

The spots in the Laue photograp occur as the position expected from the reflection law. On passing the *X*-ray beam along the axis of symmetry of the crystal, the Laue pattern contains a series of spots. The Laue patterns can be successfully used to orient crystals for solid state experiments.

# 39.5.2 Rotating Crystal Method

It is the most widely used method for the study of crystal strucure. The method consist in penetrating a beam of homogeneous *X*-ray into small crystals at right angles; the crystal is rotated round an axis parallel to one of the axes of the crystal. Due to rotation of the crystal, various planes come successively into suitable positions in order the diffraction to occur resulting in observing corresponding spots on a photographic plate (Fig. 39.8).

In Fig 39.8 A and B refer to points on two successive lattice planes. A diffraction maxima occurs the difference (BR) in the path of two diffracted rates is equal to a whole number of wavelength, *i.e.*, equal to  $n\lambda$ . As the value of n is dependent on the angle  $\theta$  (BAR), the direction of diffraction depending on increasing value of n is obtained on the photographic plate. In fact, horizontal lines (called layer lines) result for all lattice planes which have the same spacing (AB) in the direction parallel to the axis of rotation. In case the wavelength of the incident X-rays is known and the distance of between the crystal and photographic and the vertical distance between the layers is determined, it is possible to calculate the angle  $\theta$  and hence the spacing of the planes AB. The lattice spacings and the size of the unit cells can be determined by observing the set of spots in the transverse direction (Fig. 39.9).



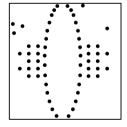


Fig. 39.8 Rotating Crystal Method

Fig. 39.9 X-ray Rotation Photograph

# 39.6 APPLICATION OF X-RAY DIFFRACTION

X-ray Diffraction is most useful of all the three X-ray procedures. Some of the applications of X-ray Diffraction are given below:

(i) Structure of Crystals: The most important use of X-ray diffraction is to determine the size of crystal planes. This is possible due to the fact that patterns obtained are characteristic of the particular compound from which crystal is formed. As an illustration, NaCl crystals and

KCl crystals give different diffraction patterns (Fig. 39.10). However, a mixture of 1% KCl in NaCl shows a diffraction pattern of NaCl with a weak pattern of KCl. In a similar way, a mixture of 1% NaCl in KCl shows the differaction of pattern of KCl with a weak pattern of NaCl (Fig. 39.10).

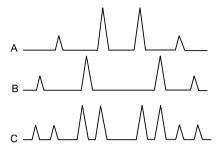


Fig. 39.10 X-ray pattern of two hypotical salts

- (A) X-ray pattern of salt-A
- (B) X-ray pattern of salt-B
- (C) X-ray pattern of a mixture of A and B; in this case the peaks match with those in (A) and (B)

In case of a mixed crystal of sodium potassium choride containing Na and K ions in the same crystal lattice, there will be changes in the crystal's lattice size as shown in Fig. 39.11.



**Fig. 39.11** X-ray pattern of powdered mixture of A and B; the peaks fall between those in Fig. 39.10C, indicating that the lattice size does not concide with that of Fig. 39.10(a) or 39.10(b), but is inbetween them

Identification of unknown crystalline compounds is possible by the comparison of diffraction pattern with patterns from crystals of known compounds.

Instruments are now available which use computers for the identification of unknown sample. The procedure consisting in putting the unknown sample into the instrument and record the *X*-ray diffraction pattern. This pattern is then fed into a computer memory bank which already has the diffraction patterns of a number of unknown compounds. The computer on instruction compares the diffraction pattern of unknown compound with those of known compounds and displays a short list of compounds. On comparison of the *X*-ray diffraction pattern with those of displayed compounds (for visual inspection) it is possible to find the unknown compound.

(ii) Characterisation of Polymers: X-rays Diffraction method has been used for the characterisation of polymers. A crystalline portion of the polymer give non-continuous diffraction line while non-crystalline polymer scatters the X-rays beam to give continuous background. The ratio of the area of diffraction peaks to scattered radiation gives the ratio of crystalline to non-crystalline material in the polymer.

The result obtained can be confirmed by using standard polymers with known amounts of crystalline and non-crystalline materials and basing the calculations on the known ratio of diffraction due to crystalline portion to scattering due to amorphous portion.

- (iii) X-ray Diffraction of Metals: Most of the metallic parts in an equipment are well-annealed. Due to annealing the crystals in metals are in well-ordered form and give sharp diffraction lines. However, on subjecting metals to harmmering, bending etc. they become 'fatigued'; in this condition the crystals become broken and the X-ray diffraction pattern becomes diffuse. In fact, due to fatigue nature the metal may break. In view of this, it is advised to check the moving parts (especially airplane wings) for metal fatigue. Also the parts of engine and bridges should be checked. This can be carried out by X-ray diffraction without removing the part from its position.
- (iv) Nature of Complexes: The *cis* and *trans* iosmers of a complex can be determined by X-ray diffraction studies. As as example, the structure of *bis* (pyridine-2-carboxamido) nickel (II) dihydrate (abbreviated as Ni(pia)<sub>2</sub>. 2H<sub>2</sub>O) can be determined. This complex obtained by the addition of aqueous solution of nickel (II) chloride to an alkaline solution of the ligand. The complex is obtained as orange-red, diamagnetic crystals of composition Ni(Pia)<sub>2</sub>.2H<sub>2</sub>O. On the basis of X-rays studies, it is found that the complex possesses a *trans* planar structure; the two ligands are coordinated with nickel atom *via* four nitrogen atoms.

It is found that the two water molecules are not coordinated directly with nickel atom and are situated between the complex molecules; these are hydrogen-bonded with each other and are linked to the amido oxygen atom of the complex (Fig. 39.2).

Fig. 39.12 Structure of bis (pyridine-2-carboxamido) nickel

X-ray diffraction method has also been used to determine the nature of the linkage in complexes. As an example, the reaction of biuret (abbreviated as biu) with copper (II) ion in alkaline medium to form a complex potassium bis (biureto) cuprate (II) tetrahydrate,  $K_2[Cu(biu^{2-})_2]$ .  $4H_2O$ . On the basis of X-ray diffraction studies, it has been found (Freeman, Smith and Taylor) that in the complex, two biuret anions are coordinated as didentate ligands through the N atoms of the amido group and the four N atoms are at the four positions of an square.

(v) Applications to Organic Molecules: Since many organic compounds are not obtained in crystalline state, it was believed that X-diffraction method cannot be used for such compounds. It has, however, been possible to convert some of the organic compounds into crystalline derivatives which have characteristic X-ray pattern. Using this procedure identification of dibasic acids, aldehydes and ketones (as 2,4-dinitrophenylhydrazones), fatty acids (as p-bromoanilides) and amines (as picrates) have been determined.

Bond lengths and angles of a number of organic compounds, predicted by new quantum mechanical theories of valency were confirmed by X-ray diffraction methods. The molecules includes hexamethylbenzene, naphthalene and anthracene. The structure of penicillin and vitamin  $B_{12}$ , were assigned (D.C. Hodgkin and her associates) on the basis of X-ray studies. In a similar way, structure of a number at complex alkaloids were assigned.

On the basis of X-ray studies, Kendrew and Perutz and their associates assigned structures to proteins—myoglobin and haemoglobin. These authors were Jointly awarded Nobel prize in chemistry in 1962 for their work.

X-ray work on some nucleotides and nucleosides showed that the heterocyclic ring in pyrimidines and purines is not perfectly planner as thought to be earlier.

(vi) Structure of Diamond and Graphite: On the basis of X-ray diffraction study of Diamond (W.H. Bragg and W.L. Bragg) it was shwon that in diamond all C atoms are surrounded by four other atoms situated at the corners of a regular tetrahedron. The C—C distance was found to be 1.54Å corresponding to single covalent bond in aliphatic organic compounds.

In graphite the C atoms are arranged in flat 3.14Å apart, each atom is attached to only three others and the distance between the centres of two adjacent carbon atom is each layer is 1.42Å. This type of arrangement corresponds to that found in benzene.

The intensity of each wavelength is recorded as the detector rotates in an arc around the analysing crystal.

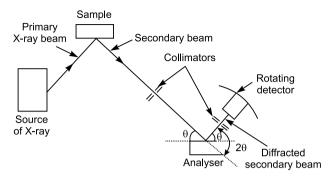


Fig. 39.13 Schematic representation of an X-ray fluorescence system

In fluorescence instrument, the intensity of X-ray source should be high and for this, the voltage to the source should be high (as high as 100 KV). Another requirement is that the wavelength of radiation from source should be shorter than the wavelength necessary to excite the sample. Due to this requirement, the choice of the target to be used in the source is important. Generally, tungsten, (which has a high atomic number) is used as target element. For this a shorter wavelength of X-ray radiation is required.

For selective X-ray fluorescence, an X-ray beam having a wavelength that excites one element only is used. The fluorescence intensity of the first element is measured and then with a second source. The difference in intensity is dependent an how much element of interest is present in the sample.

# 39.7 X-RAY FLUORESCENCE

On placing an element in a beam of X-rays, the X-rays are absorbed. The resultant excited atoms of the element emit X-rays of characteristic wavelength. This process is called X-ray fluorescence. As the wavelength of the fluorescence is characteristic of an element, the measurement of this wavelength is helpful to identify the fluorescing element. It is possible for the quantitative determination of an element by measuring the intensity of fluorescence.

The wavelength,  $\lambda$ , of fluorescence is calculated (by measuring the angle of diffraction,  $\theta$ ) by using the Bragg equation

$$n\lambda = 2d \sin \theta$$
.

where d is spacing between the crystal layers of the analysing crystal and is known. Knowing the wavelength of fluorescence,  $\lambda$  it is possible to identify the element

564

by referring to the chart of the fluorescence wavelengths of all known elements. *X*-ray Fluorescence is not dependent of the chemical forms of the element under examination.

# 39.7.1 Instrumentation

The instrument for *X*-ray fluorescence is diagrammatically represented as shown in Fig. 39.13. The filtered primary *X*-rays from an *X*-ray source is allowed to hit the sample. The produced *X*-ray fluorescence radiation is characteristic of the element under examination. The radiation is passed through a collimator, and the relative wavelengths are separated by the diffracting crystal.

The monochromator reacts with X-rays is the same way as a grating monochromator reacts with visible light. The function of the crystal is to separate X-rays of different wavelengths by diffracting them at different angles. Best results are obtained by using perfect crystals. For better sensitivity, a curved crystal is used. A number of curved crystals made from different salts (like NaCl, LiF, quartz, aluminium etc.) are available. The choice of the crystal depends on the wavelength range to be investigated.

Differentiation and measurement of X-ray fluorescence without using analyzing crystal monochromator system is possible by using a proportional counter. In these counters, the height of the peak is determined by the number of electrons per pulse and is proportional to the energy, hv, of the photon, i.e., a 2.00Å photon will produces pulse peak height which is one-half of a 1.00Å photon.

# 39.7.2 Applications of X-ray Fluorescence Spectroscopy

Some of the important application of X-ray fluorescence spectroscopy are given below.

(i) Elemental Analysis: X-ray fluorescence spectroscopy is most useful for the analysis of both metals and non metals having atomic number more than 12. This is not possible by emission spectroscopy or atomic absorption.

It is possible to carry out qualitative analysis by the measurement of the angle of diffraction of the fluorescent X-rays, from which the wavelength of fluorescence can be calculated. Since each element fluoresces at its characteristic wavelength, the element can be easily identified by referring to available tables.

Quantitative analysis can be carried out by the measurement of the intensity of fluorescence of wavelength, which is characteristic of the element to be determined. This procedure is used for the quantitative analysis of most of the elements in the periodic table.

(ii) In Agriculture: X-ray fluorescence spectroscopy is used to determine the trace elements in plants, foods and the detection of the presence of insecticides on fruits and leaves.

- (iii) In Medicine: Direct determination of sulfur in proteins can be accomplished. X-ray Fluorescence can indicate protein distribution a diagnostic link for the medical practitioner. Other applications include determination of chloride in blood serum, determination of strontium in blood stream and bone tissues and elemental analysis of tissues, bones and body fluids. It is appropriate to state the content of strontium in bone and blood is the result of radioactive fallout.
- (iv) In Mining and Metallurgy: X-ray fluorescence is helpful for the analysis of ores, tailings and drilled cores, determination of silica in flowing slurries of ores, determination of zinc in flowing slurries of zinc concentrates, determination of lead in lead-tin alloys, determination of Cu, Zn and Sn in copper alloys etc.
- (v) Nature of the oils used in automobiles can be ascertained by determining the additives like Ba, Zn, P, Ca, Cl and Pb, S in gasoline.
- (vi) Sulphur, which is used as vulcanizing element in rubber industry can be determined by *X*-ray fluorescence.



- **1.** How are *X*-rays produced?
- **2.** Describe different types of *X*-ray methods.
- **3.** Discuss the theoretical consideration involved in *X*-ray analysis
- **4.** Explain the interation of *X*-rays by absorption, scattering and diffraction.
- **5.** What do you understand by *X*-ray crystallography.
- **6.** Describe the instrumation for *X*-rays absorption, *X*-ray diffraction and *X*-ray fluorescence.
- 7. Discuss the applications of *X*-ray absorption, *X*-ray diffraction and *X*-ray fluorescence.



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