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B.Sc. CHEMISTRY

THIRD YEAR

PAPER – VII : ANALYTICAL CHEMISTRY

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UNIT – I

Analytical chemistry is a measurement science consisting of a set of powerful ideas and methods that are useful in all fields of science and medicine.

1. THE ROLE OF ANALYTICAL CHEMISTRY

(i) Analytical chemistry is applied through industry, medicine and all the sciences. Example, the concentrations of oxygen and of carbon dioxide are determined in millions of blood samples every day and used to diagnise and treat illnesses.

(ii) Quantities of hydrocarbons, nitrogen oxides and CO present in automobile exhaust gases are measured to assess the effectiveness of smog-control devices.

(iii) Quantitative measurements of ionized calcium in blood serum help to diagnise parathyroid disease in humans.

(iv) Quantitative analytical measurements also play a vital role in many research areas in chemistry, biochemistry, biology, geology, physics and the other science.

1.1. IMPORTANCE OF ANALYTICAL METHODS IN QUALITATIVE AND QUANTITATIVE ANALYSIS

Qualitative analysis

The process of identifying the components present in a sample is called a qualitative analysis. It follows that a qualitative analysis of a sample tells us what components are present in it.

For example,

A qualitative analysis of a mixture of inorganic salts gives us information about the presence of the basic and acidic radicals in the mixture. In qualitative analysis, it is essential to give importance to the questions of "How to analyse particular types of material" and "How to separate the components of different types of materials". It should be stated here that the quantitative analysis of a sample of matter does not give the numerical information about the amount of the components present. Nevertheless, a rough estimation on the amounts of the components can be made from the qualitative analysis.

Quantitative analysis

The process of determining the amounts of each of the components in a sample of matter is termed as quantitative analysis. In other words, a quantitative analysis gives much more precise information about the quantity of the desired component or components of the sample. The components of a

sample under analysis are called the analyte. In a quantitative analysis, the results of analysis are generally expressed in such relative terms as parts of analyte per hundred (i.e., percent), per thousand, per million (i.e., ppm) or per billion of the sample, and sometimes, in terms of the weight or volume of the analyte per unit volume of the sample, and also in mole fraction.

CHEMICAL AND INSTRUMENTAL METHODS OF ANALYSIS

We will now discuss the different analytical methods used to determine the physical properties of the substances under two categories.

1. Chemical methods

In fact, analytical methods developed after the introduction of the measuring instruments. Thus the introduction of analytical balance made the gravimetric method possible for the quantitative analysis of chemical compounds. After the development of the gravimetric analysis of the solutions, glass wares were calibrated for the volumetric measurements. It is interesting to note that until 1920 the quantitative analysis of chemical compounds was based upon the measurement of mass and volume. It was, therefore, the gravimetric and volumetric methods are referred to as classical or chemical methods.

(a) Gravimetric methods

This method involves the separation of desired component from the solution of the weighed sample in form of a pure weighable stable compound of known composition. As stated earlier, the gravimetric analysis can be carried out by three processes, e.g., precipitation, electrodeposition, and volatilization.

(b) Precipitation

In precipitation, the desired component is first precipitated by adding a precipitating agent to the solution of the sample followed by separation by filtration. The precipitate is then dried and finally weighed. From the known composition of the precipitate (i.e., the stable compound), the weight of the desired component is determined.

(c) Electrodeposition

In electrodeposition, as stated earlier, the weight of the desired component is determined from the difference in weight of the corresponding electrode taken before and after the electrodeposition.

(d) Volatilization

In volatilization, the amount of the desired component (if volatile) is determined from the difference in weight before and after volatilization of the sample.

(e) Volumetric method

This method involves the measurement of volume of a gaseous component or liquid component. Generally, volumetric analysis refers to the procedure what is termed as titration. Depending on the type of stoichiometric reaction, volumetric titrations are of four types, namely, acid – base titration (neutralization reaction). Oxidation – reduction titration (change of valency states), precipitation titration and complexometric titration.

A number of physical properties can be applied in analysis with a titration. These include conductometric, potentiometric, coulometric, spectrophotometric titrations etc.

2. Instrumental methods

Analytical methods employed for chemical analysis based on the physical properties of the substances need instruments available for the purpose. Such methods are known as instrumental methods of analysis. The optical, electrical, thermal, radiochemical properties, etc. can be detected by means of instrumental methods. Instrumental methods have some advantages over the chemical methods, i.e., instrumental methods are used to save time, to avoid chemical separations, and to get greater accuracy. Nevertheless, both the methods, chemical and instrumental, go side by side, in the sense that the accuracy in the instrumental methods of analysis depends on the improvement in the chemical methods of analysis. Moreover, various chemical steps, e. g., sampling, dissolution, separation of interfering substances, concentration, precipitation, etc., involved prior to analysis are an integral part of an instrumental method.

Functions of instrumental methods

As stated earlier, a physical property of a substance can be measured by an appropriate instrumental method. It follows that different instruments are needed for measuring different physical properties. In nearly all cases, the instrument does not give the quantitative data of the desired substance but provides information relative to a standard substance. It may be stated that the instrument simply converts the chemical composition in to information that is readily observable. Thus, an instrument may be regarded as a communication device acting either directly or indirectly as a comparison between the unknown and standard substance.

(a) Electroanalytical methods

These methods involve the measurement of such electrical properties as potential, current, resistance and quantity of electrical charge.

(b) Spectroscopic methods

It is mainly based on measurement of the interaction between electromagnetic radiation and analyte atoms or molecules or on the production of such radiation by analytes.

(c) Finally, a group of miscellaneous methods includes the measurement of such quantities as mass – to – charge ratio of molecules by mass spectrometry, rate of radioactive decay, heat of reaction, rate of reaction, sample thermal conductivity, optical activity and refractive index.

STEPS IN ANALYSIS

The properties of a substance can be described in terms of its colour, solubility in various liquids, response to heat, electrical and / or magnetic behaviour, ability to react with other chemical species, etc.

From the forgoing discussion, it is evident that several different physical properties may be involved in various steps in analytical procedure. So prior to chemical analysis, the following steps are taken in to consideration:

1. Separation of the sample in the pure form.
2. Preparation of the sample for the analysis.
3. Analytical procedures for the measurement (chemical or instrumental methods).
4. Assessed validity of the experimental result.

Analytical methods may be classified in to different methods according to the size or quantity of the sample, viz., macro method (quantity of the sample is minimum 100 mg), and semi – micro method (sample is about 10 mg). Similarly, when the quantity of the sample is still less, e.g., 1 mg, 1 μg or 0.01 μg, the methods are termed as micro, ultra micro or sub micro methods.

A typical quantitative analysis involves the sequence of steps shown in the flow diagram

Select method

↓

Acquire sample

↓

Process sample

↓

Eliminating interferences

↓

Calibrating and measuring concentrations

↓

Calculating Results

↓

Evaluating Results

We select a method, acquire and process the sample, dissolve the sample in a suitable solvent, measure a property of the analyte, calculate the results, and estimate the reliability of the results. Depending on the complexity of the sample and the chosen method, various other pathways may be necessary.

Select method

(i) The selected method usually represents a compromise between the accuracy required and the time and money available for the analysis.

(ii) Economic factors are the number of samples that will be analysed.

Acquire sample

An analysis must be performed on a sample that has the same composition as the bulk of material from which it was taken. When the bulk is large and heterogeneous, great effort is required to get a representive sample.

For example, if the sample is already a liquid, we can avoid the dissolution step.

Process sample

Under certain circumstances, no sample processing is required prior to the measurement step. For example, once a water sample is withdrawn from a stream, a lake or ocean, its pH can be measured directly. Under most circumstances, we must process the sample in any of a variety of different ways. The first step is often the preparation of a laboratory sample.

Eliminating interferences

An interference or interferent is a species that causes an error in an analysis by enhancing or attenuating (making smaller) the quantity being measured. A scheme must be devised to isolate the analytes from interferences before the final measurement is made.

Calibrating and measuring concentration

The measurement of the property is directly proportional to the concentration.

$$c_A = kX$$

Where k is proportionality constant. The process of determining k, an important step in most analyses, is termed a calibration.

Calculating and evaluating results

Computing analyte concentrations from experimental data is usually relatively easy, particularly with modern calculators or computers. These computations are based on the raw experimental data colleted in the measurement step, the characteristics of the measurement instruments, and the stoichiometry of the analytical reaction.

1.2. SAFETY MEASURES

Handling reagents

The following rules should be strictly observed in storage and handling of chemicals.

1. After using a chemical, the container should be tightly closed and returned to its original place.
2. Care should be taken to see if all the containers have the appropriate labels stuck to them. No bottle should be left unlabelled.
3. Corrosive chemicals like acids and alkalis should not be straightaway poured from their containers; they should be carefully siphoned avoiding spillage.
4. Corrosive chemicals should be stored in corrosion – resistant chambers.
5. Some chemicals, especially concentrated acids, produce copious fumes on exposing to the atmosphere and therefore it is advisable to handle these inside a fume – hood.
6. Flammable liquids should not be heated in an open vessel over a free flame. They should be heated in a flask fitted with a reflux condenser on a water or steam bath with the burner extinguished.
7. Flammable solvents like benzene, alcohol, ether and carbon disulphide should never be handled without turning off burners, heaters etc. Such liquids should not be kept in open beakers. Excess of these should be returned to the container and not poured in to the sink.
8. Volatile solvents must be removed by distillation and not by evaporation.
9. Some of the organic solvents like ether, because of their high volatility, build up a pressure in the containers. Such containers should be cooled in ice and then opened. Similarly, a bottle of ammonia should be opened only after cooling.

10. Only minimum quantity of reagents should be used; using excess of these is hazardous and also expensive.

Hazards in laboratory

Strong acids

Wash immediately with large quantities of water and then with a 5% solution of Na_2CO_3. Lightly dress with sterile boric acid powder and bandage with wet compress of saturated boric acid.

Strong alkalis

Wash quickly with large quantities of water and then with a 5% solution of acetic acid. Dress with boric ointment. Similar treatment is used for burns from sodium peroxide and metallic sodium.

Bromine

Wash immediately with large quantities of water and then apply gently concentrated sodium thiosulphate solution with a cotton wool repeatedly. Dry gently and apply carron oil (a mixture of linseed oil and lime water).

Phenol

Wash immediately with ethanol. If the burn is severe, dress with boric ointment or sterile Vaseline.

Waste disposal

Waste disposal in a chemical laboratory is an important aspect of health care. The following general rules should be observed in this regard.

1. Corrosive waste should be treated and made harmless before letting it down the sink. If it is directly discharged, the laboratory drainage fittings will be ruined. Concentrated acids and alkalis should never be poured in to the sink.
2. Toxic chemicals should never be washed down the sink. If this is done, their vapour will pollute the laboratory air, causing serious health damage. If treatment procedures are known, they should be treated before discharge. Otherwise, they must be poured in to special containers and kept aside for disposal at a far away place.
3. All waste products should be discarded only in to the container intended for these. It is advisable to remove waste matter from a laboratory as frequency as possible to avoid serious health problems.

Fume disposal

Formation of fumes is very common in chemical laboratories. Frequent exposure to chemical fumes is a sure way of contracting diseases connected with lungs. Therefore, every laboratory must have an efficient fume disposal

system. Exhaust fans can, to some extent suck out fumes from the laboratory. However, these are not efficient in removing all the fumes. It is desirable to carryout reactions producing fumes inside fume – hoods. Fume – hoods, fitted with proper motors can suck all the fumes from the laboratory and discharge the fumes in to the atmosphere above building. Any operation producing fumes should be done only inside the fume – hood, after switching on the motor. Chemicals which produce fumes on exposure to air should also be stored in fume – hoods. Attempts should be made to absorb the fumes produced during a reaction in some reagents. For example, if hydrogen chloride is evolved in a reaction, it should be trapped with water or aqueous alkali.

Freely let – out laboratory fumes cause great damage to the laboratory fittings and furniture in addition to posing health hazards.

1.3. DATA ANALYSIS

In analytical chemistry we are going to get a lot of data. How to analyse and interpret these data? This we shall see in this chapter.

Significant figures

They are figures in a number which contains only digits known with certainty plus the first uncertain one.

A measured value has some uncertainty about it. There is a convention to give the measured value as a number such that it contains only one figure about which there is uncertainty. The practice is called significant figure convention.

For example,

If the weight in a weighing is known with certainty only up to three decimals, the value should be reported only up to four decimals.

Its importance

In presentation scientific data, one comes across a set of values. For this set of values, one gives their mean or median as the best value. Now the uncertainty about this best value must also be indicated while presenting the data. To achieve this, usage of significant figures is very helpful. On several occasions one has to round off numbers to give meaningful results. For this, the practice of rounding off to a number which contains only one uncertain figure in it, is adopted. Thus significant figures become important.

Salient features of significant figures

The number of significant figures in a given number is found out by counting the number of figures from left to right in the number beginning with the first non-zero digit and containing till reaching the digit that contains the uncertainty.

Examples

1. Each of the following has three significant figures. 583, 0.234, 1.67, 0.00987 and 65.4
2. Zero is a significant figure when used as a number. It is not a significant figure when it is used to locate decimal points in very small and very large numbers. Example: 0.02750 has four significant figures. The two zeros before 2 are used to imply only the magnitude. So they are not significant. The zero beyond 7 is significant.
3. The value 6.030×10^{-4} has four significant figures while 1.45×10^{5} has three significant figures.

Using the above, one can determine the number of significant figures in a given number.

ACCURACY

It is the degree agreement between the measured value and the expected or true value of a property.

As per analytical data available, the solubility product of AgCl is 1.8×10^{-10}. If an analyst gets this same value while determining the same in an experiment, then, it is said that accuracy of the result is excellent. If it differs, we calculate the deviation. This deviation gives a measure of the accuracy of experiment.

One must clearly understand the connotations of the terms precision and accuracy. An understanding of the two terms will make it clear that high precision does not imply accuracy. We can prove this with an example. In a volumetric estimation one may get concordant titre values. Yet the result may turn out to be wrong. Here, as concordant titre values have been got, the result is precise. But as the answer does not agree with the expected value, it is not accurate. Thus a precise value may not be an accurate value. The reason for this may be a determinate error like instrumental error or operative or personal error like personal carelessness or even some unknown indeterminate error.

Methods of expressing accuracy

Accuracy is expressed in terms of absolute error or relative error. The lower these values are, the more will be the accuracy.

ERRORS IN CHEMICAL ANALYSIS

Error

The difference between the measured value of a property and its accurate value is called the error.

During the chemical analysis we measure the value of a particular property. E.g. we measure the weight of an object or the volume of a solution. Accurate results will be got when persons with great skill do the measurements with best instruments. This is nearly impossible. Usually the measured value of the property will never be the accurate value of the property. The difference between these two is called the error. Such errors in measurement will affect the accuracy and precision of the property that is measured. So the analytical data so obtained becomes unreliable.

Absolute error (E)

It is defined as the difference between the accepted value (x_t) and the observed value (x_i)

$$E = x_i - x_t$$

Relative error (RE)

It is the error percentage of the accepted value.

$$RE = E/x_t \times 100$$

Advantage of relative error over absolute error

Absolute error depends on the reliability of the accepted value itself as a lot of uncertainty may be there about the accepted value. So relative error is used more often to express accuracy

TYPES OF ERRORS

The errors that arise in a chemical analysis are classified in to two types. They are

(i) Determinate errors or Systematic errors

(ii) Indeterminate errors or Random errors

(i) DETERMINATE OR SYSTEMATIC ERRORS

These are errors which have a definite value and an assignable cause. The analyst can measure and account for these errors. These can be avoided. They are unidirectional, i.e., the errors will be either more or less than the accurate value. From this, they can be identified.

Source of these errors

(i) Defective instruments

(ii) Careless operation

(iii) Procedural defects

Classification of Determinate errors

Determinate errors are classified in to three types.

(i) Instrument errors

(ii) Methodic errors

(iii) Personal errors

(i) Instrument errors

When we use balances, weights, pipettes, burettes etc., we must make sure that they are not defective. For example, a weight marked 10 g may not be 10g after all. So to avoid these errors one must use best instruments. Periodic calibration of apparatus and weights is a must. These errors may be identified by changing the instrument, the error will also change.

(ii) Methodic errors

These are introduced by defective experimental procedures. Eg.,

(i) Co-precipitation or post precipitation in gravimetric analysis

(ii) Usage of improper indicators in volumetric analysis.

These are difficulty to identify. So these are the most serious of the three types of determinate errors. So to avoid these, one must be thorough with the theoretical part of the experiment.

(iii) Personal errors

Personal errors also known as operative errors. These are introduced by personal defects or carelessness. The sources of this error are human defects in eyes, mind etc. In colorimetric experiments errors will be introduced by a person who is colour blind. A person with defective eyes will invariably note readings erroneously. Carelessness, fatigue and improper instructions from the teacher also introduce these errors. Several mistakes may creap in. E.g., Wrong calculations, wrong placement of decimals, nothing wrong signs, cooking results etc. These can be avoided if one works scrupulously in the laboratory. These are identified by the fact that these errors change when the measurement is repeated.

Determinate errors may also be classified as being either constant or proportional errors.

Constant errors

These are errors whose magnitude in independent of the size of the sample taken for analysis. For example, let us say that 0.5 mg of precipitate is lost when washed with 200 ml of the wash liquid. Now, if we wash 500 mg of precipitate with 200 ml of wash liquid 0.5 mg of the precipitate will be lost. So

the loss is (0/5 x 100) ÷ 500 = 0.1%. Let us assume that we wash 50 mg of the precipitate with 200 ml of wash liquid, here also 0.5 mg will be lost. So the loss is (0/5 x 100) ÷ 50 = 1%.

Thus we find that a constant error will become more serious as the size of the quantity measured decreases. So to minimize the effect of constant error, we have to use a large sample.

Proportional errors

These are errors whose magnitude increases or decreases in proportion to the size of the sample taken for analysis. Invariably impurities in the sample, if not removed, will cause a proportional error.

Correction of determinate errors

Determinate instrumental errors are corrected by calibrating the instruments concerned. The equipments are to be calibrated periodically. This is because instrumental errors arise due to wear, corrosion or mistreatment.

Determinate personal errors can be minimized by care and self discipline. Instrument readings, note book entries and calculations should be checked systematically.

Determinate method errors are particularly difficult to detect. They may be corrected by one or more of the following procedures.

Analysis of standard samples

A method may be tested for determinate error by analysis of a synthetic sample whose over all composition is known and which closely resembles the material to be tested by the particular method.

Independent analysis

When samples to be analysed are not available in a pure state this method is used. The sample is analysed in a particular method. Then it is analysed by a different method of established reliability.

Blank determinations

Constant errors affecting physical measurements can be frequently evaluated with a blank determination, in which all steps of the analysis are performed in the absence of the sample. The result is then applied as a correction to the actual measurement. This method is useful to correct errors that are due to the introduction of interfering contaminants from reagents and vessels employed in the analysis. This method is useful to correct titration data in volumetric analysis.

By taking large sample size

We know that a constant error decreases as the size of the sample is increased. So to correct such type of errors large sample size is used for analysis.

(ii) INDERMINATE OR RANDOM OR ACCIDENTAL ERRORS

They are errors arising from uncertainties in a measurement that are unknown and not controlled by the person doing an experiment.

Sources

(i) Instrument uncertainties

(ii) Method uncertainties

(iii) Personal uncertainties

Identification

Identification of indeterminate errors is difficult. Scatter of data about the mean is the effect of an indeterminate error.

ERROR ANALYSIS

When indeterminate error or deviation from mean (dm) is plotted against its frequency (f) we get a curve as shown in figure.1. This bell shaped curve is called Gaussian or normal error curve. The properties of this normal curve are

(i) The frequency is maximum where the indeterminate error is nil.

(ii) There is symmetry about this maximum, suggesting that positive and negative errors occur with equal frequency.

(iii) As the magnitude of the error increases, the frequency decreases exponentially.

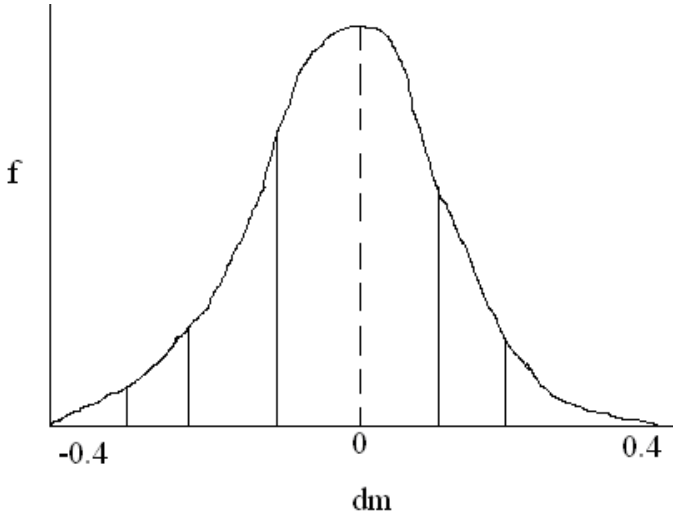


Figure. 1

In chemical analysis, indeterminate errors follow this Gaussian type distribution. For example, if the deviation from the mean (dm) of hundreds of repetitive weighing measurements on a single object, are plotted against the frequency (f) of occurrence of each deviation we get a curve as shown in figure.2. This curve proves the fact that a number of small independent and uncontrolled are there in our normal measurements. These uncertainties manifest in the result.

This Gaussian distribution of most analytical data permits us to use statistical techniques to estimate the extent of indeterminate error. Thus we use several statistical techniques like mean, median, average deviation, standard deviation etc., while analyzing our experimental results.

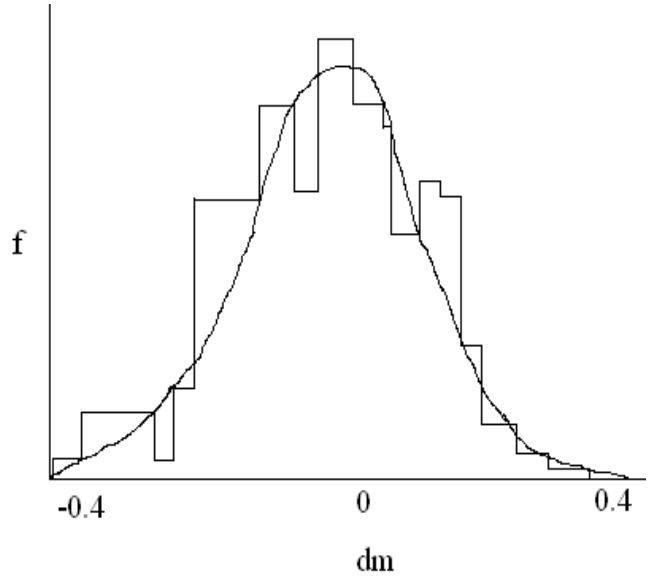


Figure. 2

Minimising error

From the above discussion it is clear that, if we want to minimize error we have to minimize determinate as well as indeterminate errors.

To minimize determinate errors we will have to use standard, internationally accepted instruments. The various measuring aids must be calibrated periodically and got certified according to international standards. All reagents must be properly maintained. Dependable procedures must be adopted to avoid methodic errors. To avoid personal errors one must be careful and honest in recording the observation. It is human tendency to manipulate results to get high degree of precision. This tendency must be avoided.

We know that indeterminate errors are uncontrollable. So to minimize these errors, we repeat the experiments several times and adopt statistical techniques to get maximum precision.

The following are some other specific suggestions to minimize error.

1. Blanks experiments are to be conducted along with regular ones.
2. Almost care is taken to avoid personal errors. Arithmetic mistakes etc., should not be committed.
3. While taking readings, one must be very careful to note the correct reading.
4. Without getting proper and complete instructions, the experiment should not be done.
5. When one becomes tired, the experiment must be stopped in a convenient place and continued after taking sufficient rest.

PRECISION

It is the degree of agreement between two or more measured values of a property, measured under identical conditions.

Let the weight of a beaker be 20.0891 g when weighed in a particular set of conditions. If the same beaker is weighed under identical conditions and if the weight got is the same 20.0891 g, then we say that there is precision in the weighing. Thus if a value is reproducible, then it called a precise value.

The difference between the highest and lowest values in a set of values is called the range (w). This range is a measure of precision. If the range is wide, then it means that the measurement is less precise.

Precision is expressed by two methods

(i) Absolute method

(ii) Relative method

(i) Absolute method

In this method the precision is expressed in terms of average mean deviation. The smaller the value of average mean deviation the greater will be the precision.

(ii) Relative method

In this method the precision is expressed in terms of percentage deviation from the mean of a set of values. i.e.,

$$\text{Percentage deviation from the mean} = \frac{\text{Mean of a set of values minus a particular value}}{\text{The Mean}} \times 100$$

$$= \frac{X - X_i}{\bar{X}} \times 100$$

MEAN / AVERAGE

Mean is the quotient obtained when the sum of a set of replicate measurements by the number of individual results in the set.

Example

Calculate the mean for the following set of values 20.20, 20.08, 20.01

Solution

Mean = (20.20 + 20.08 + 20.01) ÷ 3 = 60.29 ÷ 3

= 20.096667

MEDIAN

It is the value about which all other values are equally distributed. Half the values will be greater and the other half smaller than the median.

The median is obtained by arranging the set of values in increasing or decreasing order. If there is odd number of values then the middle value gives the median. If there is even number of values then the average of the middle pair gives the median.

Example

(i) Calculate the median for 20.20, 20.08 and 20.01

Solution

Arranging the values in increasing order we get 20.01, 20.08 and 20.20. This set contains odd number of values. Therefore the median of this set is the middle value i.e., **20.08**

(ii**)** Calculate the median for the following set of values: 20.21, 20.04, 20.13 and 20.19

Solution

Arranging the values in increasing order we get 20.04, 20.13, 20.19 and 20.21. This set contains even number of values. Therefore the median of this set is the average of the middle pair of values.

i.e., (20.13 + 20.19) ÷ 2 **=** 40.32 ÷ 2 = 20.16.

MEAN DEVIATION

The average deviation of a set of values is the average of the deviations of all the individual values from their average.

Explanation

To get the average deviation

(i) The average of the given set of values is calculated

(ii) The deviation of each value from the average is calculated.

(iii) The average of all these deviations (ignoring sign) gives the average deviation.

Example

Calculate the mean deviation for the following set of values: 20.21, 20.04, 20.13 and 20.19

Solution

Average of this set = (20.21 + 20.04 + 20.13 + 20.19) ÷ 4

= 80.57 ÷ 4 = 20.14

Value	**Deviation from average**
20.21	| 20.14 – 20.21| = 0.07
20.04	| 20.14 – 20.04| = 0.10
20.13	| 20.14 – 20.13| = 0.01
20.19	| 20.14 – 20.19| = 0.05

Total of deviation from average = 0.07 + 0.10 + 0.01 +0.05 = 0.23

Mean deviation = 0.23 ÷ 4 = 0.0575.

Its usefulness

If the mean deviation of a set of measurements is small it means that the average of that set is nearly precise.

STANDARD DEVIATION

It is the square root of the quotient obtained by dividing the sum of the squares of the individual deviations from their mean by the number of measurements made.

(Or)

$$\text{Standard deviation } \sigma = \sqrt{\text{Sum of squares of individual deviations from Their mean} \div \text{Number of measurements made}}$$

Explanation

To obtain standard deviation:

(i) The average ($\bar{x}$) of the measurement (x_i) is calculated

(ii) The individual deviation of each measurement from the average $(x_i - \bar{x})$ is calculated.

(iii) Each individual deviation is squared $(x_i - \bar{x})^2$

(iv) All the individual deviation squares are added $\sum(x_i - \bar{x})^2$

(v) The value obtained in step (iv) is divided by the number of measurements made $\sum(x_i - \bar{x})^2 \div N$

(vi) The square root of the value obtained in step (v) gives the standard deviation.

Thus standard deviation $\sigma = \sqrt{\sum(x_i - \bar{x})^2 \div N}$

σ is applicable only when the number of measurements is large. But in analytical chemistry we make only a small number of measurements. So in step (v) instead of dividing by N the value obtained in step (iv) is divided by (N-1). The standard deviation for a small number of measurements.

$$s = \sqrt{\sum(x_i - \bar{x})^2 \div (N-1)}$$

Example

Find the standard deviation for a subset having the following six values: 7.720, 7.725, 7.736, 7.719, 7.742 and 7.751

Solution

(i) Calculation of average $\bar{x}$

$(7.720 + 7.725 + 7.736 + 7.719 + 7.742 + 7.751) \div 6 = 46.393 \div 6$

$= 7.732$

(ii) Calculation of $\sum|x_i - \bar{x}|^2$

x_i	$\lvert x_i - \bar{x}\rvert$	$\lvert x_i - \bar{x}\rvert^2$
7.720	0.012	1.44×10^{-4}
7.725	0.007	0.49×10^{-4}
7.736	0.004	0.16×10^{-4}
7.719	0.013	1.69×10^{-4}
7.742	0.010	1.00×10^{-4}
7.751	0.019	3.61×10^{-4}

(iii) $\sum |x_i - \bar{x}|^2$ = 8.39×10^{-4}

(iv) $\sum |x_i - \bar{x}|^2 \div N\text{-}1$ = $8.39 \times 10^{-4} \div 5 =$ 1.678×10^{-4}

(v) $\sqrt{\Sigma(x_i - \bar{x})^2 \div (N\text{-}1)}$ = $\sqrt{1.678 \times 10^{-4}}$ = 0.013

Therefore, Standard deviation= 0.013

Advantages

Standard deviation is more reliable than average or mean deviation to express precision as it has theoretical foundation.

If the standard deviation of a set of measurements is small, it means that the average of the set is nearly precise.

CONFIDENCE LIMITS

Definition

They are the limits, which may be set, about the experimentally measured mean ($\bar{X}$), with in which we may expect to find the true mean (μ) with a given degree of probability.

Only the mean of an infinite number of measurements can be a true or accurate mean. This true mean is represented by μ. This is unattainable as an infinite number of measurements is impossible. In practice, we make a finite number of measurements and calculate the experimental mean ($\bar{X}$). How near or how far away is $\bar{X}$ from μ. This can be determined by setting limits within these limits we can find μ with a given degree of probability. The interval between these limits is called the confidence interval. The size of this interval depends on the degree of probability that we need. If we want 99% probability, the interval will be small. 99% probability means that 99 times out of 100, the true mean will be within this interval. Similarly we can choose 95% probability, 90% probability etc., depending upon our needs. This percentage probability is called confidence level.

When the standard deviation is for a small number of measurements,

s = σ (standard deviation, applicable only when the number of measurements is large) the confidence limit for a single measurement for μ is given by

$$\mu = (\bar{X}) \pm z\,\sigma$$

where,

$$z = \frac{\bar{X} - \mu}{\sigma}$$

The confidence limit for the μ of measurements is given by

$$\mu = \bar{x} \pm \frac{z\sigma}{\sqrt{N}}$$

When σ is unknown: Confidence limit for μ of N measurements is given by

$$\mu = \bar{x} \pm \frac{ts}{\sqrt{N}}$$

Where

$$t = \frac{\bar{x} - \mu}{s}$$

Thus we find by applying statistical methods we can fix the confidence limits within which the true average of a set of experimental results can be found for various confidence levels of probabilities. For this we must know the values of z and t which are readily available in literature.

1.4. CHEMICAL AND SINGLE PAN BALANCE

An analytical balance used in the chemistry laboratories can provide weight data very accurately. An analytical balance is normally designed to weight to one ten thousandth of a gram viz., 0.0001 g. There are two types of analytical balances, the equal arm or double – pan balance and the unequal arm or single – pan balance.

Single Pan Balance

A single – pan balance is an electrically operated device for carrying out speedy weighing. This balance has only two knife edges whereas a two – pan has three knife edges. In a single – pan balance the knife edge is not placed at the centre of the beam but such that one balance arm is approximately twice the length of the other. The pan is suspended from the shorter arm of the beam. A set of weights is placed on this side of the knife – edge. A constant weight is placed at the opposite end of the beam to counterpoise the weights and the weight of pan. When an object is placed on the pan, the beam is heavier on the side of the knife – edge due to the weight of the object. Weights are removed incremently from that side of the beam until the sum of the weights removed is exactly equal to the weight of the object whereupon balance is restored.

The load on the beam when weighing object is thus the same as when there is no object on the pan and no weights have been removed. Therefore, this type of balance is called a constant load balance.

Ring weights are permanently in position on the carrier when the balance is at rest. These are removed from the carrier to compensate for the weight of the object. The weight removed is read on an optical scale. In this technique weighing is done by substitution, i.e., the weight of the object on the pan is substituted for the weights removed from the carrier. The weights are removed by gently rotating a set of dials at the front of the balance.

Weighing in a single – pan balance

Rotate the arrest – knob to the full release position and adjust the zero adjusting control until the illuminated scale indicates a reading of zero. Arrest the balance and place the object to be weighed on the pan. After turning the arrest – knob to its partial release position, rotate the dial controlling the heaviest likely weight for the object until the illuminated scale changes position. The removal of the correct range of weight is indicated by the gentle movement of the illuminated scale being in view. Inadequate or excessive removal will make the illuminated scale disappear from the illuminated window. When weights equivalent to the weight of the object have been removed, turn the arrest – knob to its full release position and allow the balance to achieve equilibrium. The weight of the object is found by taking the sum of the weights indicated on the dials and that which appears on the illuminated scale. A vernier is used in reading the scale to nearest tenth of a milligram.

Advantages of using Single – Pan Balance

1. The mechanical manipulation of the weights by turning them knobs in a single pan balance instead of placing individual weights on the pan leads to speedy weighing.
2. The load on the beam is maintained constant and therefore unlike with a two – pan balance the sensitivity remains constant.
3. Errors arising from any inequality in the two arms of the ordinary balance are avoided with a single – pan balance.
4. The use of two knife edges instead of three enhances the accuracy of weighing. This is because it is easier to achieve co – planarity of two knife edges than of three.

Rules for the use of Single – Pan Balance

In addition to the rules mentioned for using the two – pan balance, the Following rules for use of the single – pan balance must be observed.

1. The dials must be rotated as gently as possible; rule handling of the dials will dislodge the ring – weights from the beam.
2. In a single – pan balance, the beam is always under maximum load and hence increased wear of knife edges is likely. This is minimized by making use of controlled or partial release position of the beam. In the partially released position of the beam, the beam swings only partially on the knife – edge. Only at the end, for recording the final weight the beam should be fully released.
3. The balance case must be closed as soon as the object has been placed on the pan.

ERRORS IN WEIGHING

1. Correction for buoyancy

(i) Moisture and absorption effects

Absorption of water by the material being weighed can lead to wrong weights. This is prevented by weighing the substance in a closed container. Sometimes water can be adsorbed on the surface of the container from the surrounding air. This can be prevented by maintaining the weighing bottle inside a desiccator. A large glass container would probably weigh more on a humid day than on a dry day.

Interaction of the object being weighed with atmospheric moisture, CO_2 or oxygen does not signify any shortcomings in the weighing apparatus but rather means that the material being weighed is not entirely what one desires.

(ii) Electrical effects

Electrification of the container used in weighing can cause serious error. Wiping a glass container with a dry cloth tends to impart a static charge to it and this charge is transferred from the object to the balance causing the balance to swing erratically. The charged pan is attracted to the uncharged floor of the balance and hence the apparent weight of the object would be recorded as greater than the correct weight. This effect is more pronounced when the surrounding humidity is low. Therefore, rubbing the container vigorously with a cloth should be avoided. The electric charge is slowly dissipated on standing. Ionization of the air in the balance case by a piece of pitchblende or a similar feebly radioactive substance will also remove the charge.

(iii) Air Buoyancy

The apparent weight of an object immersed in a fluid is less than its true weight in vaccum by a quantity equal to the weight of the fluid displaced by the object (the Archimedes Principle).

In weighing with a two – pan balance the object on one pan is buoyed by the weight of air it displaces and these weights on the other pan are buoyed by the weight of air they displace. These two buoyancies cancel each other only if the object and the weights are of the same density so that they displace equal volumes of air. When the densities are different, an error in weighing results from buoyancy effect.

Generally, for most of the analytical determinations the buoyancy error is insignificant and can be ignored. In a double – pan balance, using a counter – weight glassware similar to the container permits some self – compensation for the influence of buoyancy. This technique for the self – compensation of buoyancy effect is not possible on a single pan balance.

2. Temperature effects

Errors are caused in weighing when the temperature fluctuates during weighing. Such errors are due to change in the densities of air, object and weights with temperature. However, this error is significant.

A more serious temperature effect is encountered when the object to be weighed is not at the same temperature as the balance and its surroundings. In this situation, air convection currents are induced which lead to either false weights or erratic pointer movement. These errors can be of appreciable magnitude and hence should be avoided.

3. Calibration of weights

A set of weights should be calibrated before it is used in the laboratory. It has to be recalibrated every year. The calibration of weights is carried out by composition of each piece of the set against the corresponding piece from a standard set which has been calibrated and certified by a recognized institution. In our country, National Physical Laboratory, New Delhi checks and issues certificates for the accuracy of standard weights.

1.5. GRAVIMETRIC ANALYSIS

1.5.1. Principle

Gravimetric analysis is an analytical technique by which one estimate the amount of a substance present in a given sample by determining the weight of precipitate obtained from that sample.

Example

To determine the amount of chloride present in a given solution it is treated with enough $AgNO_3$ solution to precipitate the chloride completely. The precipitate is filtered off, dried and weighed. From the weight, AgCl precipitate, amount of chloride present in the whole of the given solution is calculated.

THEORIES OF PRECIPITATION

Precipitating agents

A reagent added to a solution of a substance to be estimated so as to precipitate that substance is called a ***precipitating agent.***

Example

To estimate barium in a solution of barium chloride we use potassium chromate solution. Thus potassium chromate solution is the precipitating agent or the precipitant.

An ideal precipitating agent should react specifically with the ion or substance to be estimated gravimetrically and give a precipitate which

(i) Has sufficiently low solubility so that a loss, due to solubility of the solid is negligible.

(ii) Is readily filtered and washed free of contaminants.

(iii) Is unreactive and of known composition after drying or if necessary ignition.

Advantages of organic precipitants

(1) They give precipitates which are insoluble in water. So quantitative precipitation is possible.

(2) The formula weight of the precipitate is large with respect to the formula weight of the metal ion to be estimated.

(3) They give precipitates with large particle size. This makes filtration and washing efficient.

(4) Drying of precipitates is easy as organic compounds have least tendency to retain water in them.

Disadvantages

(1) Drying makes the composition of the precipitate uncertain as organic substances are decomposed or volatalise on heating.

(2) Preparation of the solution of the precipitating agents is difficult as the solubilities of these reagents in water is less.

Example

The solution of DMG has to be prepared in alcohol as it is insoluble in water.

Conditions of precipitation

(1) The ion or substance to be estimated gravimetrically should be precipitated quantitatively so that the filtrate does not contain that substance which is precipitated. i.e., solubility must be very low.

(2) The precipitate must be in the form of large particles so that it is readily filtered.

(3) The precipitate must be pure. It should not be contaminated with other substances. Even if it contains some impurities, they must be removable by washing and drying.

(4) The precipitate must have a constant composition, i.e., it should not change its composition during filtration, drying or if necessary ignition.

Techniques of precipitation

To fulfill the conditions of precipitation the following techniques are adopted.

(i) The solution of the analyte (substance to be estimated) is diluted. This will enable us to precipitate the analyte quantitatively.

(ii) The precipitation added must be chosen in such a way that it forms large particles as precipitate.

(iii) Precipitation must be done in hot condition. In hot solution the solubility increases reducing super saturation. Precipitation is hastened. Colloid formation is avoided. We get well formed precipitate.

(iv) The precipitant should be added slowly and in small lots with constant stirring. This will lead to quantitative precipitation of the analyte.

(v) Precipitation is done from homogeneous solution so that super saturation is minimized and helps the growth of the size of the precipitate.

(vi) Slight excess of the precipitant should be added to ensure quantitative precipitation.

(vii) The precipitate should be washed with suitable wash liquid (containing suitable electrolyte) to avoid peptization (dissolution of some precipitate).

MECHANISM / THEORY OF PRECIPITATION

There are two competing precipitation mechanisms

(i) Nucleation

(ii) Particle growth

The size of the precipitate depends on the fact whether (i) and (ii) dominates. Nucleation is process in which four or five ions or molecules unite to form a single particle called a *nucleus*. Further precipitation occurs either by

generation of additional nuclei or by the deposition of more and more of ions or molecules on the nuclei that have already been produced. If nucleation predominates, precipitates containing a large number of small particles result. If particle growth predominates, the precipitates containing a small number of large particles result.

Factors affecting the particle size of a precipitate

Particle size depends on chemical composition of a precipitate and the existing conditions during its formation. Solubility of the precipitate in the medium in which it is being formed, reactants concentration, the rate at which the reactants are mixed, temperature at which the precipitation is conducted are some of the factors which influence the particle size of the precipitate. The effect of these factors can be accounted for, qualitatively, by assuming that the particles size is related to the relative super saturation of the system.

$$\text{Relative super saturation} = \frac{Q - S}{S}$$

Where Q = Concentration of the solute at a given instant,

S = its equilibrium solubility.

When (Q - S) / S is large, the precipitate tends to be colloidal. When this parameter is small we get coarse, crystalline particles.

Conditions to get precipitate in the form of large particles

(i) Precipitation is to be conducted at elevated temperatures, this increases S.

(ii) Precipitation is done from dilute solutions. This decreases Q.

(iii) The precipitating agent is to be added slowly. This also decreases Q

(iv) While adding the precipitant, the solution is to be stirred well. This also decreases.

SOLUBILITY PRODUCT

Definition

The solubility product of a sparingly soluble salt is the product of the concentrations of its ions in the saturated solution. It is a constant at a given temperature. It is denoted as K_{sp}.

Explanation

When a slightly soluble ionic solid such as silver chloride is placed in water, Ag^+ ions and Cl^- ions from the solid phase pass in to solution till the

solution becomes saturated. Now there exists equilibrium between the ions present in the saturated solution and the solid phase. Thus

$$AgCl \rightleftharpoons Ag^{+} + Cl^{-}$$

(solid) (In solution)

Applying the law of chemical equilibrium, the value of equilibrium constant.

$$K = \frac{a_{Ag+}\ a_{Cl}^{-}}{a_{AgCl}}$$

Since the activity of a solid is taken as unity, the above expression may be written as

$$Ka_{AgCl} = a_{Ag+}\ a_{Cl}^{-}$$

Or

$$K_{sp} = a_{Ag+}\ a_{Cl}^{-}$$

K_{sp} is called the solubility product

Ionic product

Ionic product of a substance in solution is the product of the concentrations of its ions in that solution.

Explanation

When an ionic solid is dissolved in water, it dissolves. It ionizes in solution. Now, the product of the concentrations of the ions present in the solution is the ionic product of that substance.

Example

Let us take an ionic solid NaCl. When it is dissolved in water it ionizes as follows:

$$NaCl \rightarrow Na^{+} + Cl^{-}$$

Let the concentration of Na^{+} be $[Na^{+}]$ and that of Cl^{-} be $[Cl^{-}]$. Now the ionic product of NaCl in that solution is given by

$$\text{Ionic product of NaCl} = [Na^{+}][Cl^{-}]$$

When the ionic product of a substance in its solution exceeds its solubility product then it is precipitated.

We know that solubility product of a substance is actually the ionic product of that substance in its saturated solution. So when the value of the ionic product of a substance in its solution exceeds its solubility product, it is precipitated, because it contains more substance than necessary to make it a saturated solution.

Application

We use this solubility product principle in our gravimetric analysis. We add a slight excess of the precipitating agent to the solution of the analyte so that the ionic product of the substance to be estimated in solution exceeds its solubility product and gets precipitated quantitatively.

Use of masking or sequestering agents

They are reagents added to eliminate the interference by a substance in estimation. These are also called masking agents.

If we want to estimate Mg^{2+} gravimetrically in presence of Cu^{2+} using oxine, then we have to eliminate Cu^{2+}. For this we use CN- which forms a soluble complex with Cu^{2+} as shown below;

$$Cu^{2+} + 4CN^{-} \rightarrow [Cu(CN)_4]^{2-}$$

Thus Cu^{2+} is prevented from reacting with oxine. Now CN^{-} is the sequestering agent. The process of eliminating an ion or substance using a chemical agent so that it does not interfere in the estimation of another ion or substance is called sequestration.

Uses

1. Mg^{2+} can be estimated gravimetrically with oxine in presence of Cu^{2+} using CN^{-} as the masking agents.
2. Ca^{2+} can be estimated with suitable reagent in presence of Cu^{2+} using cyanide ion as sequestering agents.
3. In the presence of EDTA, beryllium may be precipitated with ammonia in presence of chromium, cobalt, cadmium, iron, copper, lead, zinc, bismuth etc.
4. Copper has been masked with EDTA in the polarographic determination of antimony in alloys.
5. Uranium can be separated from numerous other ions by precipitation with oxine from an EDTA solution at pH 5.3.

CO-PRECIPITATION

Definition

It is a process by which normally soluble components of a solution are carried down during the formation of a precipitate.

In the determination of barium ions by precipitation as barium sulphate other barium salts such as barium nitrate and barium chloride occlude on barium sulphate precipitate. We know barium nitrate and barium chloride are soluble. Still, during the precipitation of barium sulphate, these soluble

compounds occlude on barium sulphate precipitate. Such a process is known as co-precipitation.

Theory of co-precipitation

Co-precipitation occurs either by surface adsorption, inclusion or occlusion. Gelatinous precipitates co-precipitate by adsorption while crystalline precipitates co-precipitate by inclusion or occlusion. Adsorption is a surface phenomenon. Inclusion involves the random distribution of the contaminant throughout the interior of the solid; occlusion involves a non-homogeneous distribution of the contaminant within the imperfections in the crystal lattice of the precipitate.

Effect of co-precipitation

Co-precipitation positive or negative error may result depending on whether the co-precipitated substance has more or less molecular weight than that of the precipitate. For example, during the precipitation of $BaSO_4$, if barium nitrate is co-precipitated, a positive error is observed as the molecular weight of barium nitrate is more than that of $BaSO_4$. On the other hand, if barium chloride is co-precipitated, a negative error arises as the molecular weight of barium chloride is less than that of $BaSO_4$.

POST PRECIPITATION

Definition

It is a process in which an impurity is precipitated from its supersaturated solution some time after the appearance of the substance to be estimated as precipitate.

When calcium is determined as its oxalate, any magnesium present in the solution will be precipitated slowly on calcium oxalate. Similarly when Cu^{2+} or Hg^{2+} is determined as its sulphide, any Zn^{2+} present in the solution will be precipitated slowly on CuS or HgS. Post precipitation becomes pronounced when the precipitate is permitted to be in connecting with the mother liquor.

Theory of post-precipitation

It occurs with sparingly soluble substances which form super saturated solutions. These substances usually have an ion in common with the analyte. Example: Post precipitation of magnesium oxalate while estimating calcium by precipitating it as calcium oxalate.

Effects of post precipitation

It will lead to positive error.

PEPTIZATION

Definition

It is a process in which a coagulated colloid reverts to its colloidal state.

Explanation

During the precipitation of AgCl, if the precipitate is washed with water some of the precipitated AgCl goes back in to the solution so that a negative error is introduced.

Difference between Co-precipitation and Post-precipitation

Co-precipitation	Post-precipitation
1. It decreases with time when the precipitate is allowed to be in contact with its mother.	It increases.
2. It decreases when the solution is stirred or heated.	It increases
3. The amount of the substance co-precipitated is far less.	It is much more.

REDUCTION OF ERROR

Co-precipitation, post precipitation and peptization will lead to errors. The first two will introduce a positive error while the third, a negative error.

Minimize co-precipitation

1. The solution, from which precipitation is to be carried out, should be diluted.
2. The precipitant is to be added in small quantities with constant stirring so that the precipitation takes place as slowly as possible.
3. Precipitation is carried out from hot solutions only so that, the solubility of the impurity would be increased, formation of colloidal particles would be decreased and the attraction between the precipitate and impurity would be decreased.
4. The precipitate should be digested well. These methods would decrease co-precipitation due to surface adsorption.
5. The precipitate is dried at high temperatures so that the occluded impurity could escape.
6. Crystal growth must be slow so that occlusion is less.

Remedy of post-precipitation

1. The precipitate is washed with suitable solution so as to remove the post precipitated impurity.
2. The precipitate should be filtered off as soon as the precipitation is over.

Methods to minimize peptization

1. The precipitate should not be washed with pure water.
2. The precipitate should be washed with suitable wash liquid such that peptization is minimised. Usually washing is done with a solution containing a volatile electrolyte that can subsequently be removed from the precipitate by heating. Example. AgCl precipitate is washed with dilute HNO_3 solution. HNO_3 escapes on heating AgCl precipitate.

PRECIPITATION FROM HOMOGENEOUS SOLUTION

Definition

This is a process in which the precipitant is slowly and chemically generated in the reacting solution itself.

Ba^{2+} is precipitated as $BaSO_4$ by mixing Ba^{2+} solution containing a calculated excess of sulphamic acid. Sulphamic acid solution slowly generates SO_4^{2-} ions in solution by hydrolysis.

$$NH_2SO_3H + H_2O \rightarrow NH_4^+ + H^+ + SO_4^{2-}$$

These SO_4^{2-} ions react with Ba^{2+} ion and from $BaSO_4$ which is precipitated. Dimethyl sulphate is also used to precipitate Ba^{2+}.

Advantages

Precipitation from homogeneous media is done to avoid local reagent excesses, as the precipitating agent appears slowly and homogeneously, throughout the entire solution. The relative super saturation is thus kept low. This method results in marked increase in particle size of the precipitate and its purity.

1.5.2. CHOICE OF PRECIPITANTS

Precipitating agents may be inorganic or organic. Some inorganic precipitating agents used are

(ii) barium chloride to estimate ion

(iii) H_2SO_4 or K_2CrO_4 to estimate Ba^{2+} ions.

(iv) HCl to estimate Ag^+ ions.

(v) $AgNO_3$ to estimate Cl^- ions etc.

Some organic reagents used are

(i) DMG (dimethyl glyoxime) to estimate Ni^{2+} ions

(ii) Oxine (8-hydroxyquinoline) to estimate aluminium etc

The choice of precipitants for estimating metal ions is limited, as only one or two precipitates are there which satisfy all the requirements of a good precipitant.

For example

For estimating barium we have H_2SO_4 or K_2CrO_4 only. However we can choose between an organic precipitant and an inorganic precipitant. For example, to estimate aluminium we can use ammonium hydroxide or oxine as precipitant. While using inorganic precipitants we may come across co - precipitation or post precipitation.

SPECIFIC PRECIPITANTS

Specific precipitants precipitate quantitatively a particular ion or a substance from a solution. There are no specific precipitants in reality. However some precipitants may be mentioned which are nearly specific.

Example

Dimethyl glyoxime – DMG – is used to precipitate nickel specifically.

SELECTIVE PRECIPITANTS

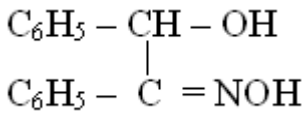
Selective precipitants precipitate a small group of ions from solutions containing several ions. Most of the gravimetric precipitants are only selective.

Example

OH^- precipitates a few metal ions as their hydroxides from solution containing several metal ions. The metal ions which are not precipitated also form hydroxides but they are soluble and so are not precipitated. S^{2-}, CO_3^{2-} are some other example of selective precipitants. When controlled conditions are adopted, selectivity of precipitants may be increased.

Cupron (Benzoin-α-oxime)

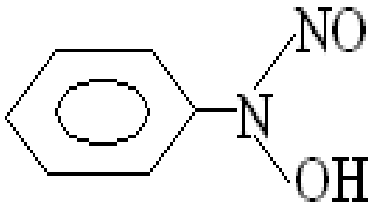
It is used to estimate copper in dilute ammoniacal solution in presence of tartrate.



A green precipitate is got it is dired at 110°C and weighed.

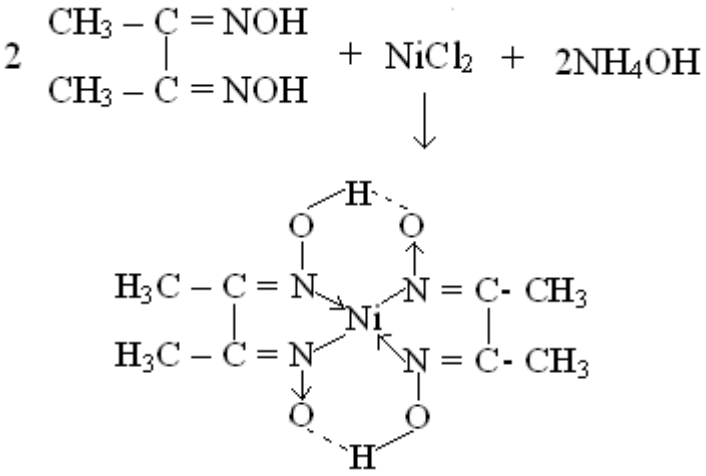
CUPFERRON

It is the ammonium salt of N-nitroso-N-phenyl hydroxyl amine. It is used to estimate Cu (II) and Fe (II) in cold aqueous acid medium. Its structure is



D.M.G. (DIMETHYL GLYOXIME)

It is a specific precipitant for estimating nickel (in alkaline medium) and palladium (in acid solution). Its reaction with Ni^{2+} ions is given below:

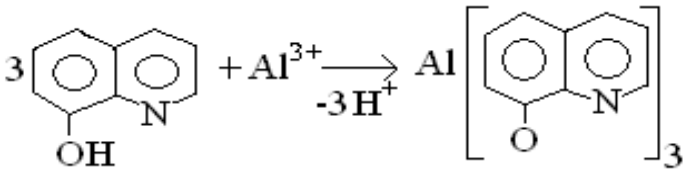


ETHYLENE DIAMINE

It is 1, 2-diaminoethane. It is used to estimate Cu (II), Hg (II) and Cd (II) ions.

8-HYDROXY QUINOLINE

About 24 cations give precipitates with oxime. It is used to estimate aluminium from a solution with pH around 3, its reaction with Al^{3+} ions is given below:



Some other metals that are precipitated by oxine are Mg, Cu, Zn etc. The solubilities of metal oxinates vary from cation to cation and are pH

dependent. So through pH control a considerable degree of selectivity can be achieved.

SALICYLALDOXIME

It is also used to estimate Cu (II) in presence of acetic acid at pH 2.6.

1.5.3. CRUCIBLES

Different types of crucibles are used for collecting and weighing the precipitates.

Types care and use of crucibles

These are mainly of two types.

1. Crucible made of silica or porcelain is used when the precipitates are ignited to constant weights.
2. Sintered crucibles are used for precipitates which after drying in air oven.

(i) **Crucibles are expensive.** They should be handled properly for obtaining reliable gravimetric data. They should never be touched but must always be handled with a pair of tongs. They should not be placed on the table but only on a clean tile.

(ii) **Porcelain crucibles.** They are usually of 3 to 4 cm in diameter. These should be carefully heated with a small flame in order to avoid fracture before being heated to a high temperature. These are suited for heating the precipitates to very high temperatures. Porcelain crucibles are suitable for use up to temperatures 1000-1200^{o}C. HF, NaOH, sodium peroxide and alkali carbonates attack porcelain and therefore substances containing these materials should not be handled in such crucibles. These crucibles can be cleaned with warm concentrated acids.

(iii) **Silica crucibles.** These are more expensive than porcelain crucibles. However, these may be safely exposed to sudden changes of temperature without any risk of fracture. Silica crucibles must not be used for alkalis or HF because these chemicals attack silica. Certain precipitates tend to adhere strongly to these crucibles and hence these crucibles should be carefully cleaned immediately after use.

(iv) **Platinum crucibles.** Platinum crucibles are marketed in various shapes and sizes. The choice of the crucible for an experiment is decided by the quantity of precipitate handled. These are very expensive and hence their use is very much restricted to cases where they are indispensable. These crucibles can be more readily and more uniformly heated to redness than porcelain crucibles.

(v) **Rose crucible.** A Rose crucible is used when a precipitate has to be ignited in an atmosphere of a gas, say, hydrogen, carbon dioxide or oxygen.

(vi) **The Gooch crucible.** A Gooch crucible is made of porcelain with the bottom perforated with a number of small holes. It is usually fitted in to a glass adopter (Gooch funnel) by means of a narrow rubber ring, the adopter passing through the rubber stopper of a filter flask. The bottom of the crucible is covered with asbestos. This covering is done by shaking asbestos fibre bits with water and pouring the mixture in to the crucible. The water drains leaving the asbestos as a thin cover at the bottom. A Gooch crucible is used usually for drying precipitates only up to about 250^{o}C, because above this temperature the asbestos in it loses its weight. This crucible is preferable to filter paper for filtering solutions such as $KMnO_4$, which attack paper.

Important Questions

1. Write the important chemical methods used in quantitative and qualitative analysis.
2. Write various instrumental methods used in qualitative and quantitative analysis
3. Discuss the various steps involved in analysis
4. What are the safety measures to be taken in handling acids and alkalis?
5. How will you handle bromine water and phenol?
6. Write a short note on disposal of wastes.
7. Write the significance and importance of significant figures
8. Distinguish between precision and accuracy
9. Discuss the various types of errors and minimization of errors
10. What is the effect of temperature on weighing?
11. Write various theories of precipitation?
12. Write notes on solubility product.
13. What are the factors affecting solubility?
14. Write notes on co and post precipitation.
15. How will you reduce errors in gravimetric analysis
16. Write notes on specific and selective precipitants
17. What are the various types of crucibles used in gravimetric analysis

NOTES

UNIT – II

CHROMATOGRAPHIC TECHNIQUES

Chromatography is an analytical technique for separating compounds on the basis of the differences in their affinity for a stationary phase and a mobile phase.

2.1. COLUMN CHROMATOGRAPHY

Definition

Column chromatography is defined as a separation process involving the uniform percolation of a solution through a column packed with finely divided material.

Principle

The separation in the column is effected either by direct interaction between the solute components and the surface of the stationary phase or by adsorption of solute by the stationary phase. Column chromatography involves adsorption, partition or ion exchange phenomena. In adsorption column chromatography, the substances are preferentially adsorbed by the adsorbent, packed in the column. In partition column chromatography of components of a mixture distribute themselves in different ratios between two different solvents and thus get separated. In this method the column is packed with silica gel or cellulose which contains significant amounts of water. In ion exchange column chromatography, ions are exchanged between the mobile phase and stationary phase.

The efficiency of column depends on

i) The particle size of the solid adsorbent. It should be uniform, it should not be too fine to affect the rate of separation or too coarse so that separation becomes

ii) It should have high specific area. This helps attainment of equilibrium of solute between stationary and mobile phase.

iii) There should be no air gaps. Air gaps lead to mixing of separated zones and allow the solution of the mixture to pass through without effecting separation of the mixture.

Adsorption

We know that the separation of a mixture by column chromatography involves adsorption, partition or ion-exchange. The method of separation is the same for all the three types. The difference is that the column must be packed suitably.

A solution of the mixture to be separated is prepared in a relatively non-polar solvent. It is introduced in to the column in a stepwise manner with the help of a pipette. The stopcock at the bottom is opened slightly to allow the solvent to run – out until a small amount of the solution remains in the column covering the top of the packed material. First the components of the mixture are adsorbed at the top of the column.

Development

When all the solution has been poured, the developing solvent is then introduced. It is allowed to flow steadily through the column. When the developer percolates through the column, the various substances are separated. This process is known as development. If the components are coloured, different colour zones are got in the column. As development continues, the separation becomes more and more pronounced. The well developed column is called chromatogram in below figure.

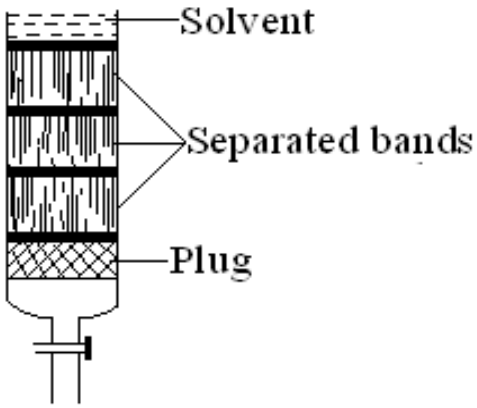


Figure.2. Development column

Elution

It is the process of recovering the various constituents of a mixture from the well developed chromatogram using a suitable solvent. The solvents used for this purpose are called eluents.

Requirements of an eluting solvent

1. It should be less polar than the components of the mixture so that it is not more strongly adsorbed than the components of the mixture on the adsorbed in the column.
2. It should dissolve the components and not the adsorbent so that separation is efficient.
3. The solvent is chosen by trial and error method.

Recovery of substances

This can be done in two ways:

i) The eluent is passed through the column till all the zones are washed out completely. The eluted solvent is collected in a separate vessel. Different vessels are used to collect the eluted solvents for different zones.

ii) Elution is carried out till the separation is complete, i.e., till we get well defined bands. Now the adsorbent with the zones is taken out. Each zone is separated with the help of a knife. Then each zone is dissolved in a suitable solvent and then analysed by usual methods.

Identification of compounds

Column chromatography is applicable to both coloured and colourless substances. If the substances are coloured, they are identified easily. If the substance is colourless, the situation is rather difficult. In practice, the elute is analysed by usual methods. The eluent with the compound is evaporated to remove the solvent. The pure compound is isolated. It is analysed.

Several physical and chemical methods may be used to identify the compound. Many colourless compounds fluorescence strongly in UV light. The eluent itself is directly led in to a device which measures the absorbed UV light by the eluent and produces peaks on the graph. Thus the fractions containing useful material can be readily located. This method is not suitable in cases where the compounds are affected by UV light such as sterols and silver salts.

Chemical methods are used where physical methods could not be used. The fractions are treated with appropriate reagents to produce coloured products. E.g. Vitamin A gives blue colour when treated with antimony chloride in chloroform.

Applications

1. Quantitative of two or more compounds of a mixture.

   Example

   i) A mixture of Pb^{2+}, Ag^{+}, Zn^{2+} ions can be separated on alumina. Here the solvent is water and the eluent is water and a solution of hydrogen sulphide. The following coloured bands are got.

      a) Lead sulphides – black at the top.

      b) Silver sulphides – grey in the middle.

      c) Zinc sulphide – dirty white near the bottom.

   ii) A mixture of ferric alum and cupric sulphate can be separated on alumina. Here the solvent is water. The eluent is also water. We get

a blue band of Cu^{2+} below a brown band of Fe^{3+}. Now the bands ar eluted with 1 to 2% solution of potassium ferricyanide. The lower band becomes brown with the formation of cupric ferrocyanide. The upper band becomes deep blue due to the presence of ferricferrocyanide.

2. Purification of substances from their contaminates.

2.2. THIN LAYER CHROMATOGRAPHY

Definition

It is a type of adsorption chromatographic technique of separation of mixtures of compounds and identification of the constituents.

Principle

In thin layer chromatography some adsorbents like silica gel, alumina etc., are supported as thin layers on glass plates (called chromatoplates). In this technique we have a wider choice of the media. So we can separate compounds by partition, adsorption and ion-exchange.

Explanation

In thin layer chromatography (TLC) some adsorbents like silica gel, alumina etc., are separated as thin layers on glass plates (called chromatoplates). This technique is similar in some aspects to both column and paper chromatography. Just as in column chromatography the moving substance is attracted by the polar sites on the surface of the adsorbent by electrostatic forces. This binding is reversible. There is a three fold interaction interaction between

i) Solvent and the adsorbent

ii) The solvent and the compound

iii) The compound and adsorbent.

Choice of adsorbents

There are a number of materials which are used as adsorbents. They are as follows:

1. Silica gel

This is the most widely used. This is used along with a binding agent like plaster of paris.

2. Alumina

This is basic in nature. It may be applied with or without plaster of paris.

3. Kieselghur

This is neutral in nature. Its adsorbing capacity is less than either silica gel or alumina. It may be applied with or without plaster of paris.

4. Cellulose

Several cellulose powders with a variety of ion exchange properties are available. They may be applied with or without plaster of paris.

5. Commercial adsorbents

Commercially the following adsorbents are used.

i) Polyamide powder

ii) Calcium sulphate

iii) Magnesium silicate

iv) Powdered glass.

Choice of solvent

The choice of solvent depends on the nature of the substances to be separated and on the adsorbent used. Generally the polarity of the solvent and the substance is matched and then the choice of solvent is made. More polar solvents produce greater migration and thus give better separation. The choice of solvent should be such that the position of the compounds must be half way between its point of application and the solvent front. Combination of two solvents always gives good separation than a single solvent. The choice is usually made from the following solvents: petroleum ether, CCl_4, benzene, pyridine, acetone, water etc.

Preparation of chromatogram or chromatoplate

Square or rectangular glass plates with sizes ranging from 2.5X20 cm to 20 X 20 cm are used as adsorbent supports in TLC. Plastic and metal foils can also be used instead of glass plates. The most widely used adsorbent is silica gel. A weight amount of the adsorbent is taken in a bottle. Water is added to it. The bottle is stirred or shaken vigorously until we get homogeneous, thick, mobile slurry. Two plates are put together. They are held by the thumb and forefinger at one end. Now they are dipped in the slurry, taken out and are held vertically. The solvent dries up. The dry plates are separated. Now we get two chromatoplates. There are also other apparatus for the preparation of chromatoplates.

Activation: TLC involves adsorption. So water or other polar solvents affect the development. So they must be removed from the chromatoplates. This is known as activation. This is done by drying the plats in an oven at 100 – 150^{o}C for about 2 hours.

Sample application

A small amount of the sample is dissolved in a small volume of a volatile solvent such as benzene, ether or ethanol. The choice of solvent depends on

i) The nature of the substance to be separated.

ii) The nature of the adsorbent.

For polar substances polar solvents are used. Less polar substances should be dissolved in a suitable non-aqueous solvent. Activated silica gel or alumina should be used as the adsorbent.

A base line is drawn, about 2.5 cm from one edge of the plate. The samples are applied in small spots, 1 cm apart, on the base line. The solvent is evaporated. Solutions of standard substances are also applied by the side of the test samples.

Development of the chromatogram

The chromatogram is usually developed by ascending method in a developing chamber called tank. The atmosphere in the tank is saturated with the solvent vapour by placing a paper impregnated with the solvent around the side of the tank. The chromatoplate is placed in the tank and the tank is filled with the solvent to a depth of about 1 cm. The tank is closed firmly with a lid. After a certain time when the solvent has moved to about 10-20 cm above the origin, the plate is removed from the tank and the solvent front is carefully marked. The solvent is evaporated.

Location of compound on the chromatogram

Coloured compounds can be identified by visual inspection. To identify colourless compounds physical or chemical methods are adopted.

a) Water is sprayed on the chromatogram. Hydrophobic compounds show their presence as optically dense waxy areas.

b) The developed chromatogram is inspected under UV light in a darkened box. Many compounds appear as dark spots in a light background.

c) Crystals of iodine are placed in the tank during development. It imparts a dark brown colour to the spots.

d) The sample is mixed with a very small amount of a radioactive isotope and applied on the plate. The radioactivity is measured by a Geiger Muller counter after development. They may be detected by autoradiography also.

Applications

TLC may be used basically for qualitative identification, quantitative separation in the preparation of organic and inorganic compounds. The technique is extremely suited for analysis of compounds which are available in traces only. A large number of inorganic compounds have been separated, identified and quantitatively analysed. The applications of TLC include the detection of by-products in synthetic processes, determination of the presence of impurity, removal of impurities and isolation of pure compounds and analysis of inorganic cations and anions.

For example

Ni, Mn, Co and Zn may be separated using TLC (or PC). A mixture of acetone and HCl is used as the solvent for development. A suitable spraying agent may be used.

Separation of a mixture of dyes

The chromatoplate is prepared with kieselghur or silica gel as adsorbent. It is activated in an oven at 100-150^{o}C for about half an hour. The solvent used for development of the chromatogram is a mixture of methyl alcohol, acetic acid and isoprophyl alcohol in the ratio 2:2:1. The solvent is taken in a rimless beaker to a depth of about 1 cm. A drop of the mixture dyes is placed 2 cm above the bottom of the chromatoplate. It is placed in the solvent for sufficient time till the solvent reaches the top of the chromatoplate. The plate is removed and dried. Various spots would be noticed showing that the dyes have been separated.

R_F - VALUES

Definition

R_f - value is defined as the ratio of the distance traveled by the compound at its point of maximum concentration to the distance traveled by the solvent.

Significance of R_f values

Both distances are measured from the point of application of the sample.

$$R_f = \frac{\text{Distance moved by the sample}}{\text{Distance moved by the solvent}}$$

In many cases the solvent moves beyond the end of the paper. So another term R_x is used. R_x is the ratio of the distance travelled by the sample to the distance traveled by a chemically similar standard substance.

$$R_f = \frac{\text{Distance moved by the sample}}{\text{Distance moved by standard substance}}$$

The movement of the solvent is always greater than the solute. Therefore, R_x value is always less than 1. Every compound has a different R_f value. R_f values depend on the solvent used. So R_f value of a particular compound is given with reference to the solvent used.

Factors affecting the R_f – values

Factors on which R_f value depends on

1. The solvent used. The R_f value is directly proportional to the polarity of the solvent. Thus R_f values are given with reference to the solvent used.
2. The pH of the solution.
3. The direction of the fibre placements in the paper.
4. Method of development, drying etc.
5. Irreversible adsorption on paper.

Two types of forces operate when a drop of a solution is applied on the chromatoplate and treated with a solvent.

I. The propelling force drags the substance in the direction of the flow of solvent. At a certain temperature, different components of a mixture will dissolve differently in a given solvent. The compound with higher solubility will move rapidly along the chromatoplate than the less soluble one. This leads to a separation.

II. The retarding force drags the substance behind towards its point of application. This retardation depends on the adsorption and partition. Thus when a drop of solution is treated with the solvent on the chromatoplate the more strongly chromatoplate adsorbed component remains at the point of application while the less strongly adsorbed will move along with the solvent. The process of partition is also operative on chromatoplate. The adsorbent always contains a small amount of water. Partition of the substance takes place between water in the adsorbent and the mobile organic solvent. This also causes separation of substances.

2.3. PAPER CHROMATOGRAPHY

Definition

It is a chromatographic technique to analyse an unknown substance and to separate mixtures of substances by flow of solvents on specially designated filter paper.

Principle

In paper chromatography the separation of unknown materials is effected by the differential migration of different components on a specially designated filter paper. This separation occurs due to difference in partition co-efficient. It is a type of partition chromatography. Here the mobile phase is an organic solvent and the stationary phase is water absorbed on the surface of the paper. Alternatively, the paper may be impregnated with anhydrous silica, alumina or ion exchange resin. Here partition occurs as a consequence of solid – liquid or ion exchange equilibria.

Paper as medium

In paper chromatography, a course paper serves the role of a packed column chromatography or the role of a coated plate in TLC. In column chromatography and in TLC silica gel acts as the solid support for the polar phase. In paper chromatography a filter paper serves this purpose. Thus paper serves as medium for partition to take place between a stationary liquid and a mobile liquid.

Solvent used

As paper chromatography is based on partition of substances between two liquids, we have to use suitable solvents for stationary and mobile phases.

1. Stationary phase

The following classes of solvents are used as the stationary phase.

a) Aqueous solvents

Water is held firmly by the paper. So it is used as the stationary phase. The filter paper is suspended in water placed in a closed chamber, whose atmosphere is saturated with water vapour and equilibrium is allowed to be established.

b) Hydrophilic solvents

A hydrophilic organic solvent can be used as the stationary phase. Eg., Methanol, Formamide, Glycol, Glycerol etc.

c) Hydrophobic solvents

A hydrophobic solvent also can be used.

E.g., Kerosene, Hydrocarbons, Dimethyl formamide etc.

2. Mobile phase

Several combinations are possible for the mobile phase. A mixture of two, three or more solvents, solution of salts, buffers etc., is generally used.

Example

1) Isopropyl alcohol, water and ammonia mixture in the ratio 9:2:1.
2) n – Butyl alcohol, water and acetic acid mixture in the ratio 4:5:1.

The choice of solvents depends on the nature of the substance to be separated. The choice is usually made by referring to the literature or by trial and error method.

Mechanism

In this method, the dissolved substances are applied as a small spot on cellulose bound filter paper. The paper is then dipped in to a vessel containing the mobile phase. The mixtures are partitioned between the solvent held on the paper (stationary phase) and organic solvent (mobile phase). The separation is effected by the differential migration of the mixture of substance.

Two types of forces operate when a drop of solution is applied on the filter paper and treated with a solvent.

1. The propelling force drags the substance in the direction of the flow of the solvent. The propelling force depends on the solvent flow and the solubility of substances in the solvent. The compound with higher solubility will move rapidly along the paper than the less soluble one. This leads to a separation.
2. The retarding force drags the substance behind towards its point of application. The retardation depends on the adsorption and partition. When a drop of the solute is treated with the olvent on the strip of a paper, the more strongly adsorbed component remains at the point of application while the less strongly adsorbed component will move along the paper with the solvent. The process of partition is also operative on the paper, since the cellulose of the paper always contains a small amount of water. Partition of the substance takes place between water in the cellulose and the mobile organic solvent. This also causes separation of substances.

SEPARATION OF AMINO ACID MIXTURES

We shall explain the use of paper chromatography in the separation of mixtures taking a mixture of amino acids as an example. The various operations involved are

a) Choice of the filter paper

b) Preparation of the solution

c) Application of the sample to the paper

d) Choice of the solvent

e) Development of the chromatogram

f) Drying the paper

g) Location of the substance on paper chromatogram

h) The quantitative estimation

a) Choice of the filter paper

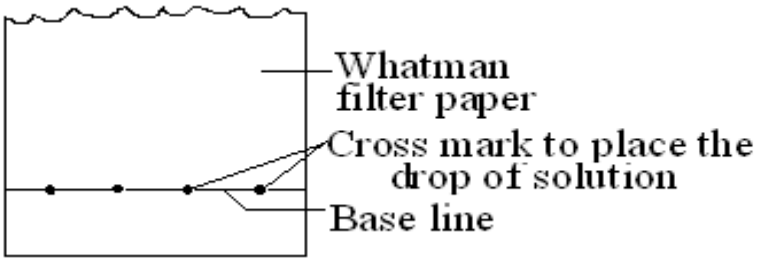
Whatman number 1 is taken (20 X 30 cm).

b) Preparation of the solution

A solution of the mixture of amino acids is prepared.

c) Application of the solution to the paper

A pencil line is drawn about 5 cm from one end of the paper below figure. This line is called the base line. A cross mark is made with a pencil on the base line. A drop of the concentrated solution of the mixture of amino acids is placed on the cross mark using a capillary pipette. The solvent is allowed to evaporate or if necessary, dried quickly with a dryer.



d) Choice of solvent

Ethanol, water and ammonia are mixed in the ratio 8:1:1. This mixture is used as the solvent.

E) Development of the chromatogram

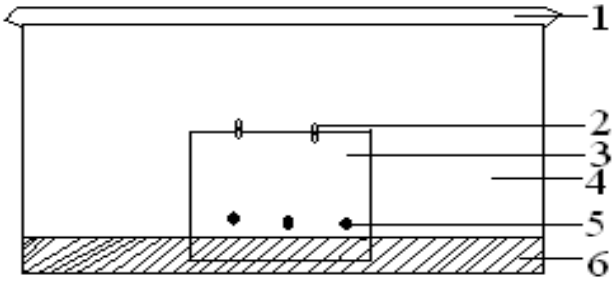
The chromatogram can be developed by

1. The ascending technique
2. The descending technique
3. The radial development

1. Ascending technique

Here the solvent is allowed to travel up the paper. The solvent is placed at the bottom of a tank and the paper is suspended from the top, with the help of

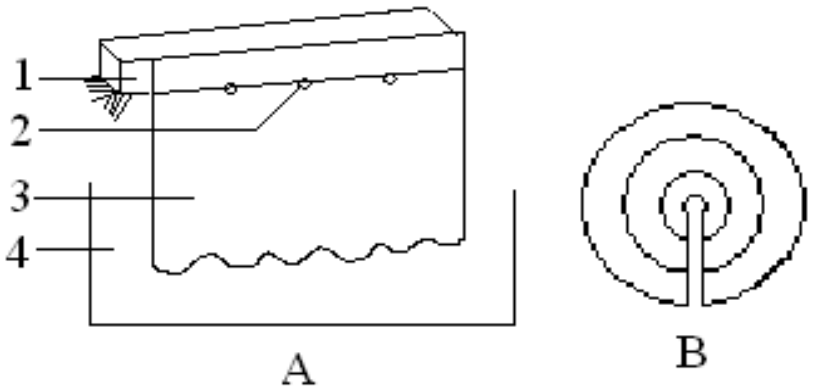
a clip, so that the lower end of the paper containing the spots is well above the solvent in below figure. The paper is made in to a cylinder. The two ends are clipped together. It is then placed in to the solvent tank. The tank is always closed with a lid. The solvent moves up the paper. The solute is partitioned between water in the filter paper and the solvent. Equilibrium is established between the two phases.



1. Lid
2. Wire clip
3. Paper
4. Tank
5. Spot
6. Solvent

2. Descending technique

Here, the solvent is placed at the top of the tank. The paper is hanged such that solvent flows down the paper. A small amount of the solvent is placed at the bottom of the tank to replace air inside the tank with the vapour of the solvent. In this method, the solvent moves down the paper. Since the solvent can be allowed to run off the paper, the separation can be improved by increasing the length of the paper in below figure



1. Rod, 2. Spot, 3. Paper, 4. Trough

3. Radial or disc development

This technique is used in special cases. In this method a circular paper is marked with a pencil as shown in figure. A wick is cut parallel to the radius from the edge to the centre. The sample solution is applied at the upper end of the wick in the centre. The paper is dried. It is placed over a petridish containing the developing solvent with the wick dipping in the solvent. The solvent flows through the wick to the sample spot and carries the solute with it. Thus the different components of a mixture are separated. This technique is called circular chromatography.

f) Drying the chromatogram

After the solvent has moved a certain distance for a certain time, the chromatogram is taken out from the tank and the position of the solvent front is marked with a pencil. The chromatogram is now dried by blowing hot air from a hair dryer or by any other suitable method.

g) Location of amino acids

300 mg of ninhydrin dissolved in 100 ml acetone is used as the locating agent. Different amino acids appear as blue spots. The R_f values are recorded and compared with the literature for identification. Thus the mixture of amino acids is separated and identified.

PAPER ELECTROPHORESIS

Definition

The migration of colloidal particles under the influence of an electric field is called electrophoresis.

Principle

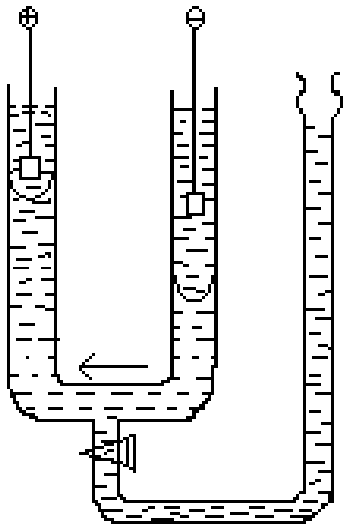
When current is passed through solutions, positively charged particles move towards the cathode and negative by charged particles move towards the anode. The movement is stopped at a suitable time so as to effect separation.

Explanation / Illustration

This principle is also used to separate substance in paper chromatography. This can be illustrated as follows: A mixture of amino acids can be separated in to individual acids by applying the principle of electrophoresis to paper chromatography.

Chromatographic Whatmann paper of suitable size is taken. The paper is soaked in a buffer solution containing pyridine and acetic acid. The apparatus (figure) consists of an anode compartment and a cathode compartment. The paper soaked in buffer solution is placed in such a way that it touches the buffer solutions in both the compartments. The mixture to be separated is applied to the positions marked 1, 2, 3, 4 and 5 on the paper. When current is switched on

the various amino acids move either to the anode or the cathode depending upon the charges possessed by them. After two or three hours, the current is switched off. The paper, called electrophoretogram, is removed and dried. The paper is sprayed with ninhydrin (locating agent) and heated. Blue – lilac spots appear at different places showing that separation has taken place. The paper is cut and dissolved in suitable solvents to get the separated amino acids.



Applications

1. This method is used for separation of large biochemical ions like proteins, poly saccharides, nucleic acids and other complex substance.
2. The principle is also used to separate substances in thin layer chromatography also. The procedure is similar to the one used in paper chromatography. Instead of paper soaked in buffer, here a thin glass plate coated with a thin layer of silica is used.

RADIAL PAPER CHROMATOGRAPHY (CIRCULAR CHROMATOGRAPHY)

In this technique the equipment consists of two glass plates and a circular filter paper. This technique is the same as other paper chromatographic techniques except that the development is done by using the circular disc.

Comparison between paper and thin layer chromatography

Similarities

1. Both techniques can be adopted to separate the same mixture.
2. Development of chromatogram, solvents used for development, spraying agents etc., is similar.

Differences

Paper chromatography	Thin layer chromatography
1. Separation is based on partition.	Separation is based on partition, adsorption and ion exchange
2. Stationary phase is water on the surface of paper.	Stationary phase is some adsorbents like silica gel, alumina which are supported as thin layers on glass plates.

Superiority of TLC over PC

1. Since TLC can be used for separations based on partition, adsorption and ion exchange, a wide variety of mixtures can be separated, identified and estimated. But since PC is based only on partition, it has limited applications only.
2. Paper as a medium is weaker than a glass plate. So damages may occur while performing the experiment. So extreme care is to be taken to avoid tearing of the wet paper. In TLC this risk is not there.
3. Corrosive reagents and acid can be sprayed on TLC chromatoplates without any damage. This is not possible in PC.

Superiority of TLC over other techniques

1. Identification and separation by TLC can be done within 20 – 40 minutes.
2. It can be applied to a wide variety of compounds, both organic and inorganic.
3. The medium in TLC is a thin layer. The particle size is very small. So we get improved resolution and compact spot.
4. It is applicable to the compounds which are decomposed by heat.
5. TLC is sensitive and gives sharper zones.
6. Here also, as in column chromatography, the moving substance is attracted by the polar sites on the surface of the adsorbent by electrostatic forces. This binding is reversible. There is a three fold interaction between
   a) The solvent and the adsorbent
   b) The solvent and the compound
   c) The compound and the adsorbent. This produces good separation of compounds.

2.4. ION EXCHANGE CHROMATOGRAPHY

Definition

It is a special category of column chromatography in which the stationary phase is an ion exchange resin.

Principle

Ion exchange is a process in which an interchange of ions of like signs takes place between a solution and an insoluble solid (ion exchanger) in contact with the solution. This process is utilized to separate a mixture of ions. Here, a reversible exchange of ions takes place between ions in a liquid phase (mobile phase) and an ion exchange resin (an insoluble substance containing ionic sites) which is the stationary phase.

Types Of Resins

Ion exchange resins granular insoluble organic compounds with giant molecules with exchangeable ions. The ion exchange resins are of two types. They are

1. Cation exchange resin
2. Anion exchange resin

1. Cation exchange resin

A cation exchange resin can be represented as (Res A^-)B^+ where Res is the acidic polymer of the resin to which is attached the anion A^- and mobile cation B^+. The cation exchanger exchanges its mobile cation with the cation of the solution.

Example

$$2(\text{Res } SO_3^-)\ Na^+ + Ca^{2+} \rightleftharpoons (\text{Res } SO_3^-)_2\ Ca^{2+} + 2Na^+$$

2. **Anion exchange resin**

A anion exchange resin can be represented as (Res B^+) A^-. Anion exchanger exchanges its mobile anion with the anion of the solution. **Example**

$$[\text{Res } N(CH_3)_3^+)]OH^- + H^+Cl^- \rightleftharpoons [\text{Res } N\ (CH_3)_3^+)]Cl^- + H_2O$$

Each of these is subdivided as follows:

i) Strongly acidic cation exchange resins

Example: Sulphonated polystyrene resins. They are useful in the pH range of 1-14. These are used to separate cations, inorganic compounds, lanthanides, vitamins, peptides and amino acids.

ii) Weakly acidic cation exchange resins

Example: Carboxylic polymethacrylate resins. They are useful in the pH range of 5-14. These are used to separate cations, biochemical compounds, transition elements, amino acids, antibiotics and organic bases.

iii) Strongly basic anion exchange resins

Example: Quarternary ammonium polystyrene resins. These are useful in the pH range 0-12. These are used to separate anions, halogens, alkaloids, vitamin B complex, fatty acids etc.

iv) Weakly basic anion exchange resins

Example: Phenol formaldehyde and polyamine polystyrene resins. These are useful in the pH range of 0-9. These are used to separate anionic complexes of metals, anions of different valencies, vitamins and amino acids.

Requirements of a good resin

1. It must be sufficiently cross linked to have only a negligible solubility.
2. It must be sufficiently hydrophilic so as to permit diffusion of ions through the structure at a constant and finite rate.
3. The swollen resin must be denser than water.
4. It should be chemically stable.
5. It must contain a sufficient number of accessible ionic exchange groups.

Action of ion exchange resins

Ion exchange resins behave as a porous network, carrying a surplus electric charge, which is distributed over the surface and throughout the pores. The surplus charge is compensated by ions of opposite charge. Thus ion exchange resins comprise of static ions attached to the resin-part and mobile ions. These mobile ions are exchanged with similar ions during ion exchange process. In this ion exchange process, no chemical bonds are formed as the available heat of exchange is low. The actual ion exchange process is taking place by diffusion occurring in two different ways.

1. Film diffusion

In this method diffusion of ions takes place across the liquid film which is adjacent to the resin particle. This method dominates in dilute solutions and with small ions.

2. Particle diffusion

In this method diffusion of ions takes place within the pores of the resin particles. This method dominates in concentrated solutions and with large ions.

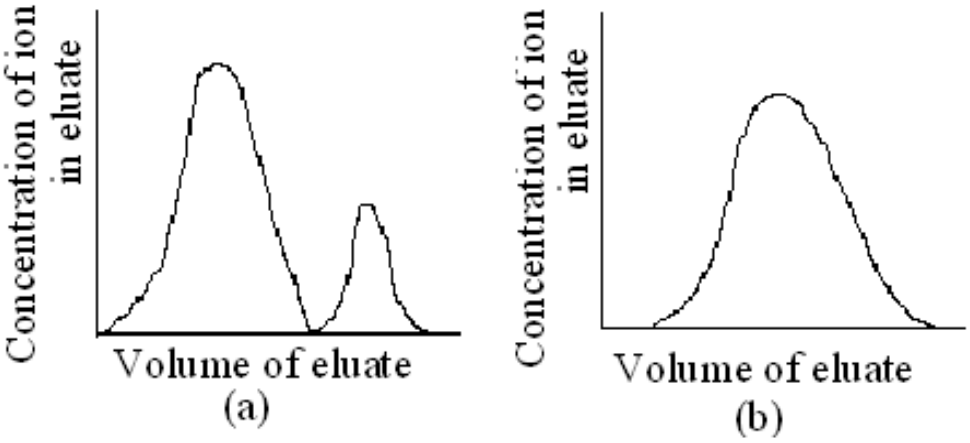
EXPERIMENTAL TECHNIQUES

The ion – exchange chromatography is carried out in a chromatographic column. The column consists of a burette provided with a sintered glass disc or glass wool plug at the lower end. The column is packed with wet ion – exchange resin uniformly. The top of the resin bed is covered with a glass wool pad. The column should never be allowed to drain out.

Let us suppose that we wish to replace the Cl^- ions by OH^- ions. The resin column is washed with a concentrated solution of NaOH to ensure the column only OH^- ions and no other anion. The solution containing Cl^- ions is run through the column. Cl^- is exchanged by OH^-. The effluent will contain a quantitative yield of the hydroxide compound. Thus same process occurs when a cation exchanger is used.

If a mixture of small quantities of two or more different cations X, Y etc., is passed through an ion-exchange column, they get separated. If cation X is held more firmly by the exchange resin than cation Y, Y will flow out of the bottom of the column before X. This separation technique is called ion-exchange chromatography. The liquid entering the column is called influent. The liquid leaving the column is called the effluent. The process by which the absorbed ions are removed from the column is known as elution. The solution used for elution is elutent and the solution obtained as a result of elution is called eluate.

If a solution of suitable eluent is passed through columns containing an ion X, the effluent is continuously analysed, and the concentration of X is plotted against the volume of eluate, an elution curve is obtained in below figure.b.



If the column contains several ions of similar charge, Y,Z etc., elution curves are obtained for each ion by the use of suitable eluents. It the elution

curves are sufficiently far apart, (fig.a) quantitative separation is possible. If the elution curves overlap, only incomplete separation is possible.

APPLICATIONS

Ion – exchange chromatography is used in the separation of rare – earths, actinides, transition metals, phosphates, complex ions and even isotopes. It serves as a standard method in the discovery and isolation of trace amount of actinides. Industrially, it is used in the metal finishing industry, extraction of metals from ores and separation of rare earths.

Some specific examples are given below:

1. Separation of rare earths

A column is packed with an acid resin and treated with HCl first to ascertain that all exchange centres were occupied by hydrogen ions. A mixture of rare earths (lanthanides) as their chlorides is sent down the column. The rare earth ions get exchanged with hydrogen ions. Later, the rare earth ions are eluted with a solution of citric acid. The cations from complexes with citric acid. Depending upon the stability of these complexes, these ions are eluted at different rates.

2. Separation of chloride and bromide ions

Principle

An anion exchange resin originally in the chloride form is converted in to the nitrate form by washing it with sodium nitrate solution. A concentrated solution of the chloride and bromide mixture is introduced at the top of the column. The halide ions exchange rapidly with nitrate ions in the resin forming a band at the top of the column. Now this band is eluted with sodium nitrate solution. Chloride ions are more rapidly eluted than bromide ions. Thus they are separated.

Procedure

A column of the anion exchange resin using 40 g of zeolite in the chloride form is prepared. The column is washed with 0.6 M sodium nitrate until the effluent contains no chloride ions. Now the column is washed with 50 cc of 0.3 M sodium nitrate solution. The mixture of chloride and bromide is mixed with 0.3 M sodium bromide and the mixture is placed at the top of the column. 0.3 M sodium nitrate is passed through the column at the rate of 1 cc / minute. Chloride ions are eluted first. Bromide ions are eluted next. Thus they are separated.

3. Separation of Co and Ni

Principle

Cobalt forms a monovalent complex anion (probably $[CoCl_3]^-$) in 9 M HCl while nickel does not. This anion is rapidly extracted from its solution by a strongly basic anion exchanger such as zerolite. The anionic chloro complex of nickel is not retained by the resin, as it is not stable. It can be washed out of the column with 9 M HCl. Later, the column is eluted with water. The cobalt complex is decomposed and passes out in the effluent as cobalt (III) chloride.

Procedure

A column of the anion exchange resin using 30 g of zerolit FF in the chloride form is prepared. 50 cc of 9 M HCl is passed through the column. 10 cc of the mixture of Co and Ni is placed at the top of the column with the aid of a pipette. A small amount of 9 M HCl is added. The mixture reaches the upper part of the column. 100 cc of 9 M HCl is passed through the column. Nickel is eluted out. It is collected. Then 50 cc water is passed through the column at the rate of 5 cc / minute. The anionic cobalt chloro complex is decomposed and comes out of the column as cobalt (III) chloride. It is collected. Thus cobalt and nickel are separated.

Advantages of ion – exchange chromatography

1. The exchange capacity of the resins is very high. So separation can be effected fairly quickly.
2. The recovery of ions from the column is nearly 100% complete. So expensive materials can be separated by this technique so that there would be no loss. Also, quantitative works could be carried out since separation is nearly 100% complete.

Factors determining the distribution of ions between an ion – exchange resin and a solution

1. Nature of exchanging ion

At low aqueous concentrations and at ordinary temperatures, the extent of exchange increases with increasing valency of the exchanging ion.

i.e., $Na^+ < Ca^{2+} < Al^{3+} < Th^{4+}$

2. Under similar conditions and constant valency, for univalent ions, the extent of exchange increases with decreases in size of the hydrated cation,

i.e., $Li^+ < H^+ < Na^+ < NH_4^+ < K^+ < Rb^+ < Cs^+$, while for divalent ions, the ionic size is an important factor but the incomplete dissociation of salts of bivalent metals also plays a part. The order is:

$Cd^{2+} < Be^{2+} < Mn^{2+} < Mg^{2+} = Zn^{2+} < Cu^{2+} = Ni^{2+} < Co^{2+} < Ca^{2+} < Sr^{2+} < Pb^{2+} < Ba^{2+}$.

2.5. GAS CHROMATOGRAPHY

Theory or principle

We know that chromatography refers to the physical method of separation based on the distribution of components between a mobile phase and a stationary phase. If the mobile phase is a gas, it is called gas chromato graphy (G.C). If the stationary phase is a solid, then it is called gas – solid chromatography (GSC). If the stationary phase is a liquid, then it is called gas – liquid chromatography (GLC). Gas – liquid chromatography is an important technique which has a liquid as a stationary phase distributed over the surface of a solid support. The technique is suitable for separation of materials which are volatile without decomposition.

EXPERIMENTAL TECHNIQUES

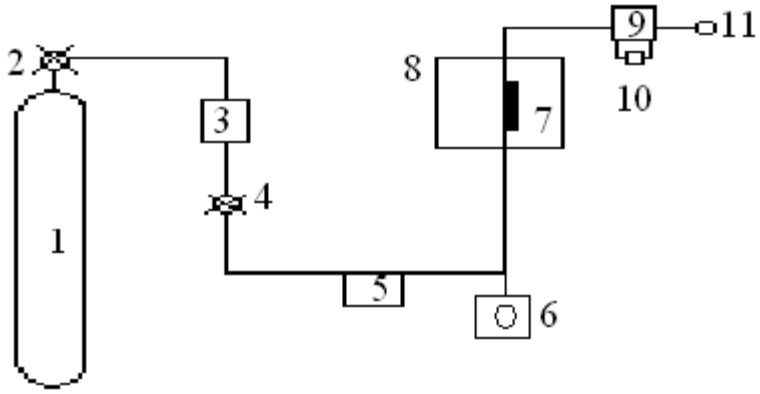


Figure. Schematic Diagram of a Gas Chromatography

1. Carrier gas cylinder
2. Gas valve
3. Desiccant
4. Fine – control valve
5. Flow meter
6. Injection port
7. Column
8. Oven
9. Detector
10. Recorder
11. Exit port

The sample is introduced in to the moving carrier gas stream in above figure. It is carried by the gas through the column. The column contains a liquid of low vapour pressure held upon an inert support or only a liquid in a capillary column. The non-volatile liquid forms the stationary phase. The carrier gas forms the mobile phase. The components of the mixture sample distribute themselves between the two phases. The solubility of different components in the liquid phase will be different. Therefore these components are carried along the column at different rates. They finally emerge at the end of the column in distinct zones (peaks) separated by the carrier gas. On emerging, the vapours of the components are detected by suitable methods and are recorded.

Procedure

There are six essential parts of a laboratory gas chromatography (GC).

(i) A gas cylinder containing a carrier gas

(ii) A sample injection system

(iii) The column

(iv) The thermal compartment

(v) The detection system

(vi) The recorder

(i) Carrier gas supply

The most commonly used carrier gases are He, H_2, N_2 and CO_2. The carrier gas should be pure. The flow rate of the carrier gas should be constant.

(ii) Sample injection system

A small amount of the sample is introduced in to the carrier gas with a syringe. Solid sample can be dissolved in a suitable solvent and introduced by a syringe.

(iii) The column

The columns are between 120 cm to 500 cm in length and 2-10 mm in diameter. They are made from stainless steel, copper, glass or plastic. They may be coiled, or bent in V or W – shape. Partition columns are used in GLC. The partition columns are packed with an inert support like finely divided celite, ground fire bricks or glass beeds. These inert supports carry a non – volatile liquid phase. The choice of liquid phase depends upon the nature of the substance to be separated. Some liquids used are silicon oil, greases etc.

Capillary columns are open tubes made of nylon, glass, copper or stainless steel. They are 0.1 to 1 mm in diameter and 30 to 300 mm in length. The inside of the capillary tube is coated with a liquid partitioner.

(iv) Thermal compartments

The temperature of the column can be maintained uniformly by the use of vapour jacket containing benzene, toluene etc.

(v) Detection system

Detectors measure either the concentration of the solute in mole fraction in the carrier gas or the flow rate of the solute, (in moles per unit time). Some of the detectors which are commonly used are

a) Thermal conductivity detector

b) Gas density detector

c) Flame ionization detector etc.

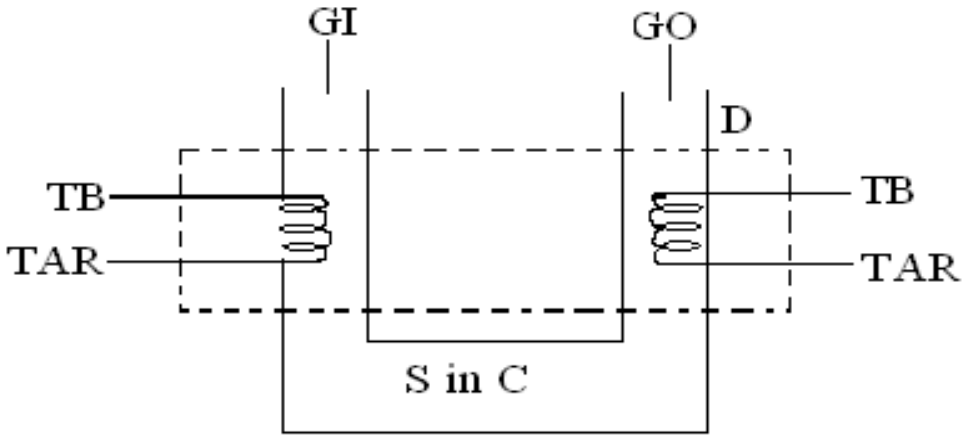
a) Thermal conductivity detectors

Principle

Thermal conductivities of gases are different. So a change in composition of the gas changes thermal conductivity. Thermal conductivity detectors are based on the change in the thermal conductivity of the gas stream. For this purpose, an instrument called Katharometer is used.

Procedure

A schematic diagram is shown below:



D = Detector; GI = Gas in; GO = Gas out; S in C = Sample in column; TB = To battery; TAR = To amplifier and recorder

A current is passed through the instrument, the thermal conductivity of the surrounding gas changes with a change in the temperature of the wire. The resistance of the wire is measured. From that, the thermal conductivity of the gas can be calculated and the change in the gas mixture in the effluent stream is monitored.

b) Flame ionization detector

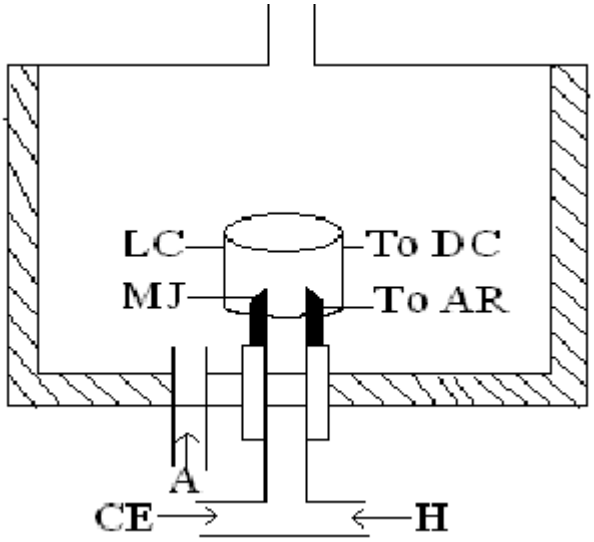
Principle

Organic compounds are pyrolysed when introduced in to a hydrogen or oxygen flame. As a result of pyrolysis, ions are produced. These are collected and the resulting current is measured.

Procedure

A hydrogen flame ionization detector is shown in below figure. The column effluent gas is mixed with hydrogen and burnt at a metal jet which acts as cathode. A loop of cylinder acts as the anode. As the composition of the gas in the flame changes, the number of ions and electrons will also change. Thus the current flow will change with the change in composition of the gas eluted from gas chromatographic column.

This is the most popular detector.



CE = Column effluent; H = Hydrogen; To DC = To source of direct current; To AR = To Amplifier and recorder; A = Air; MJ = Metal Jet; LC = Loop of Cylinder

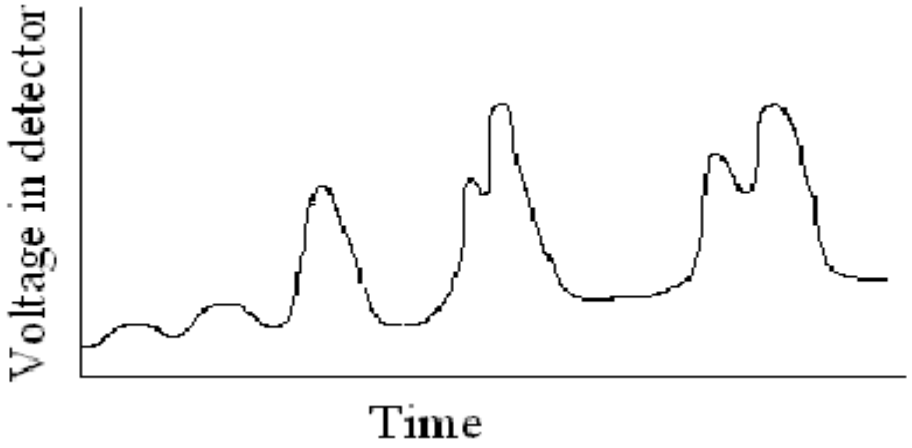
(vi) Recorder

All the detectors give rise to a small and weak electrical signal. These are passed through an amplifier and fed in to the recorder. The amplified signals drive the recording chart strip. We get a series of peaks on the paper.

Working Technique

The carrier gas obtained from a steel gas cylinder is passed through a flow regulator. This enters in to the sample injector. A small amount of the sample is introduced in to the sample injector using a syringe. The temperature of the sample injector is maintained slightly above the boiling point of the

highest boiling component of the sample. The carrier gas enters the sample injector. Its sweep off the vapourised sample and passes through the column. The components of the sample are distributed between the stationary and the mobile phases. They pass down the column at different rates. This results in the separation of the components of the sample. The carrier gas with the separated components enters the detector. The detector measures the change in concentration of the carrier gas. This change is amplified, fed in to a recorder. We get a chromatogram in below figure.



Interpretation of GC

The peak is generally bell shaped. Usually the time required for components to travel through the column is compared with that of known compounds. The area of the peak is a direct measure of the concentration of each compound present in the sample.

APPLICATIONS OF GAS CHROMATOGRAPHY

We can quantitatively determine the amount of a component in a mixture with GLC. The peak area is measured. This is a quantitative measure of a particular component.

Example

β – Diketo ligands like acetylene, trifluoroacetyl acetone (TFA) and hexafluoroacetylacetone (HFA) form stable volatile chelates with Al, Be, Cr (III) and a number of other metal ions. These can be identified, separated and estimated by GLC.

1. Fatty acids have been separated by this technique. The other mixtures separated by this technique are
    a) Benzene and Toluene,
    b) Chloromethanes

c) Species with similar boiling points but with different polar characteristics.

2. It can be used for quantitative analysis of mixtures also. Peak height, peak areas are compared with those of known substances and their quantity determined.
3. It has been used in elemental carbon, hydrogen and nitrogen analysis of organic compounds. They are converted to CO_2, H_2O and N_2 by burning them in the presence of oxidizing catalysts like MnO_2. The gases are detected.
4. In petroleum industry, it has been used in the analysis of crude petroleum products, fractions, gasolines, waxes, LPG, sulphur, nitrogen compounds etc.
5. In food industry, it has been used to account for the colour and flavor of food. It has been used for the determination of residual solvents in spices, oleoresins and for pesticides in food.
6. In cosmetics and perfume industry, it is used to determine the composition of the various cosmetics, the quality of ingredients and components of subtle fragrances.
7. In plastic industry, it is used to identity plastics, determination of esters in acrylic copolymers. In many research fields in the above industries and in many other fields it finds use.

2.6. HIGH PRESSURE LIQUID CHROMATOGRAPHY

Principle

HPLC involves the use of tightly packed columns containing small particles (a few micrometer diameters) of the stationary phase. The use of small particles (unlike the big particles used in usual column chromatography) provides greater surface area for the sample components; this increases the separation efficiency of the column. In other respects, it is similar to gas chromatography.

Instrumentation

As the column is packed with fine particles, the flow of solvent through the column would be very slow. To overcome this problem, in HPLC pressure (6000 psi) is applied on the inlet of the column with a pump; the outlet is atmospheric pressure (This method is also known as high pressure liquid chromatography, as it employs high pressure). Using the pump, the flow – rate is adjusted to be about 0.01 to 10 cm^3 per minute. The below figure describes a HPLC instrument.

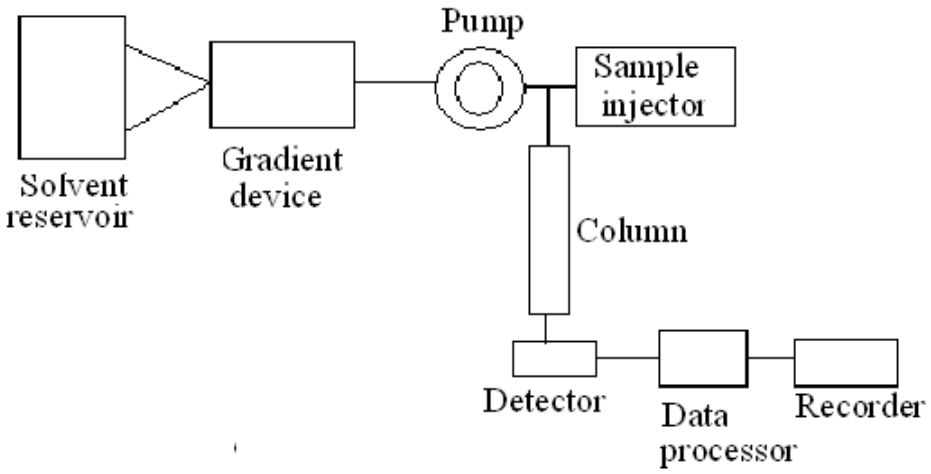


Figure. HPLC Block diagram

Several separation mechanisms can be employed in HPLC; these are partition, adsorption, ion exchange, and size exclusion. However, partition mechanism is the most widely used in this method.

The adsorption – HPLC separation of the isomeric nitroanilines on silica column is depicted in below figure. Ion exchange – HPLC can be used for separating metal ions. For example, a mixture of Cu^{2+}, Zn^{2+} and Cd^{2+} can be separated by this method.

Advantages of HPLC

1. It is extremely efficient in separation.
2. A wide variety of materials can be analysed by this method.
3. It requires only a few microlitres or micrograms of the analyte. However, the HPLC instrument is expensive.

2.7. PURIFICATION TECHNIQUES

Purification of organic compounds

Solvent extraction

It is a separation technique adopted to separate a solid or liquid present in a mixture by extracting it with a solvent.

Principle

The substance to be extracted should be soluble in a particular solvent while all the other constituents in the mixture should be insoluble. After extraction the solvent should be easily separable.

Superiority of solvent extraction over precipitation

1. 100% separation is achievable in solvent extraction method. In precipitation methods there is a possibility of some substance

remaining in the mixture, i.e., efficiency is more in solvent extraction.

2. The procedural part is simple.
3. In solvent extraction method a small amount of solvent is enough as it is recovered and recycled.

Uses

Solvent extraction is used to separate

(i) Dissolved substances from their solutions

(ii) One constituent from a solid mixture

(iii) Unwanted impurities from substances

(i) Separation of a substance from a solution in a solvent from other dissolved substances using another solvent

The principle used is Nernst distribution law. According to this law, at constant temperature, a solute distributes itself between two immiscible solvents only in a particular ratio. The ratio of the concentrations in the solvents is called the partition coefficient or distribution coefficient.

If C_A and C_B are the concentrations in liquids A and B then at constant temperature, $C_A / C_B = K$ = Constant. Larger the value of K, more efficient is the extraction.

When a substance distributes itself between two solvents without the complications of dissociation or association, it is possible to calculate the weight of the substance which can be removed by a series of extractions. If V_1 ml of a solution contains W g of a substance and if the substance is repeatedly extracted with V_2 ml of another solvent, the weight of the substance W_n remaining in the first solvent alter n extraction is given by

$$W_n = W (KV_1/KV_1 + V_2)^n$$

Where K is the distribution co-efficient.

(ii) Separation of one constituent from a solid mixture

If one constituent in a solid mixture is soluble in a solvnt while the other constituents are not, then that soluble solid can be separated from that mixture by solvent extraction.

(iii) Separation of unwanted impurities from substances

When the impurities are soluble in a solvent then may be extracted with that solvent.

Purification of solid organic compounds

Organic solids can be purified by the following methods:

Extraction - Use of immiscible solvents

When an organic solid is present as solution in water it can be recovered from the solution. For this, the aqueous solution is shaken with an immiscible organic solvent in which the solute is more soluble. The solvent layer is separated by means of a separating funnel. The organic substance is then recovered from it by distilling off the solvent.

Soxhlet extraction

Definition

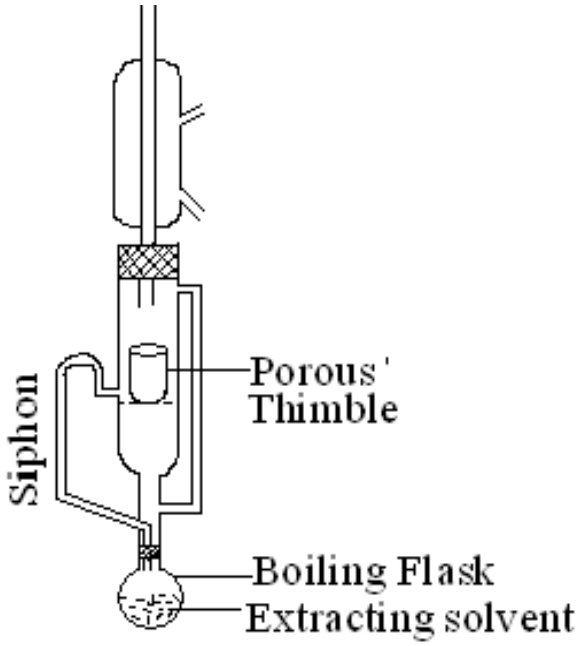
It is the extraction of a solid with an organic solvent, in which the impurities are insoluble, using the Soxhlet extractor.

Principle

The substance to be extracted should be soluble in a particular solvent while all the other constituents in the mixture should be insoluble. After extraction the solvent should be easily separable.

Procedure

Soxhlet extractor consists of a glass cylinder C with a side tube T and a siphon S. A water condenser is attached at the top of C. its bottom is fitted to a boiling flask in figure.



The sample from which the solid to be extracted is powdered. It is placed in a thimble made of a thick filter paper. The thimble is placed at the bottom of the water condenser. The extracting solvent is taken in the boiling flask. The solvent is boiled. Its vapour rise through T and enter the water condenser. There, the vapours condense. The condensed solvent falls on the

sample in the thimble. The substance to be extracted dissolves while the impurities don't. The solution is filtered by the thimble and collects at the cylinder C. When C becomes full, the solution reaches the flask through S. The solvent becomes a vapour again and rises up through T. The extracted solid remains in the flask. Thus the solvent extracts the solid continuously from the sample and brings it to the flask. Finally the solution from the flask is distilled. The solvent distills off, leaving behind the organic substance.

Superiority over conventional solvent extraction techniques

1. This method provides a through contact and heating with the solvent.
2. A small quantity of the solvent is enough to extract a maximum amount of solid as the solvent is recycled. In conventional methods it is not possible.
3. This is a continuous process. So the efficiency of extraction is more in this method.

Uses

This method is used for the extraction of oils and fats from flowers and seeds and alkaloids from plants.

Crystallization

This is another method of purification of solids.

Principle

This process is commonly employed for the purification of solid compounds. The impure solid is dissolved in the minimum volume of a suitable solvent. The soluble substances pass in to solution while the insoluble ones are left behind. The hot solution is then filtered and allowed to cool undisturbed till crystallization is complete. The crystals are then separated from the mother liquor by filtration and dried.

The efficiency of the process of crystallization depends on

1. Choice of solvent
2. Preparation of solution
3. Filtration of the solution
4. Crystallization
5. Separation and drying of crystals.

Technique

In this method, the impure solid is dissolved in enough hot solvent so as to get a super saturated solution. The solution is hot filtered to remove insoluble articles and dust. The filtrate is cooled. Pure crystals of the solid separate out.

The impurities remain in solution. Some solvents used for this purpose are; benzene, petroleum ether, methyl alcohol, acetone etc.

Fractional crystallization

This is a process used to separate two or more substances from a solution. When a hot solution containing two or more solids is cooled, the solid with lesser solubility crystallizes out first. The other solids crystallize out in the order of their increasing solubilities.

This technique involves the following steps whether the quantity being purified is 100 g or 1 g

1. Selection of the solvent
2. Preparation of a nearly saturated hot solution
3. Cooling of the solution to induce the formation of crystals
4. Separation of mother liquor and washing
5. Drying of the crystals.

The most appropriate vessel for crystallization of 50 – 200 mg of substance is 3 or 4 – inch tubes. Small beakers and flasks are not recommended as loss of solvent by evaporation in these becomes considerable. When the solvent is volatile and the rate of solution is slow, a finger condenser is used. Heating is done either over a micro or semimicro burner or an adjustable micro hot plate.

The filtration apparatus should be capable of filtering hot solutions rapidly without clogging from crystallization. Let us consider the inside of the funnel is ground to provide a firm seat for a perforated porcelain disc which fits inside the funnel. A disc of filter paper is placed on top of the porcelain disc which fits inside the funnel. A disc of filter paper is placed on top of the porcelain disc and moistened with water. The funnel fits into an 8- inch tube having a side arm. The solution is allowed to cool slowly. The funnel described above is also used for filtration of crystals. The crystallization is repeated till the melting points of the crystals from two successive crystallizations agree with in 0.50.

Separation of crystals from the mother liquor can also be accomplished by centrifuging. The centrifuge tube consists of two ordinary glass tubes with the rim of the inner tube resting on the rim of the outer. The inner tube has a small opening at the bottom. The mother liquor drains through this opening.

Sublimation

Sublimation is a process in which a solid, when heated passes directly in to the vapour state without melting and the vapours when cooled, become solid directly without condensing in to a liquid.

Principle

Certain substances when heated pass directly from solid to vapour state without melting. The vapours when cooled, give back the solid substance. This process is known as sublimation. The process of sublimation is used in the purification of volatile solids like naphthalene, camphor etc., from non volatile solids.

(i) Sublimation under ordinary pressure

The solid to be purified by sublimation is placed on a sand bath in figure a. The china dish is covered with a perforated filter paper. A funnel is inverted over the china dish. The china dish is gently heated. The sublimable solid evaporates, passes through the pores of the filter paper and is deposited as a pure solid on the walls of the funnel. The filter paper prevents the sublimed solid from falling back in to the china dish and prevents the funnel from getting the heat and thus keeps it cool.

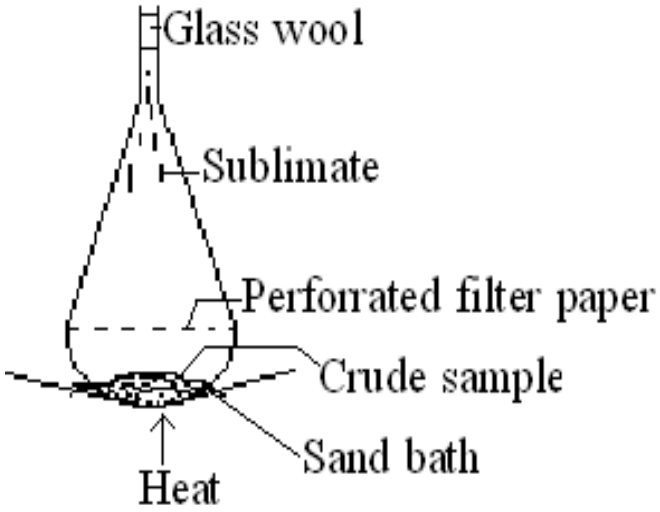


Figure a

(ii) Sublimation under reduced pressure

Substances having low vapour pressure and substances which decompose on heating cannot be purified by direct sublimation. For such substances, sublimation under reduced pressure is effected. The apparatus for this is shown in figure b. In this apparatus there is a small space between a large heating surface and a cooling surface. On heating the solid sublimes and condenses as a solid at the bottom of the surface cooled by water.

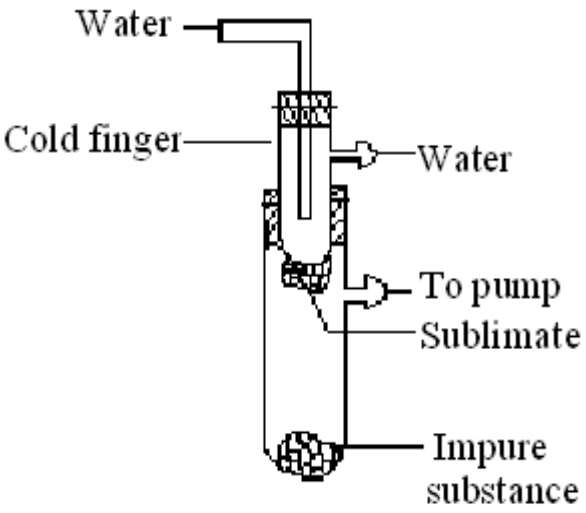


Figure b. Sublimation under reduced pressure

Uses

The technique is used for the purification of sublimable solids like camphor, naphthalene, benzoic acid etc.

PURIFICATION OF LIQUIDS

Liquids are purified by

(i) Distillation

(ii) Fractional distillation

(iii) Distillation under reduced pressure

(iv) Steam distillation

EXPERIMENTAL TECHNIQUES OF DISTILLATION

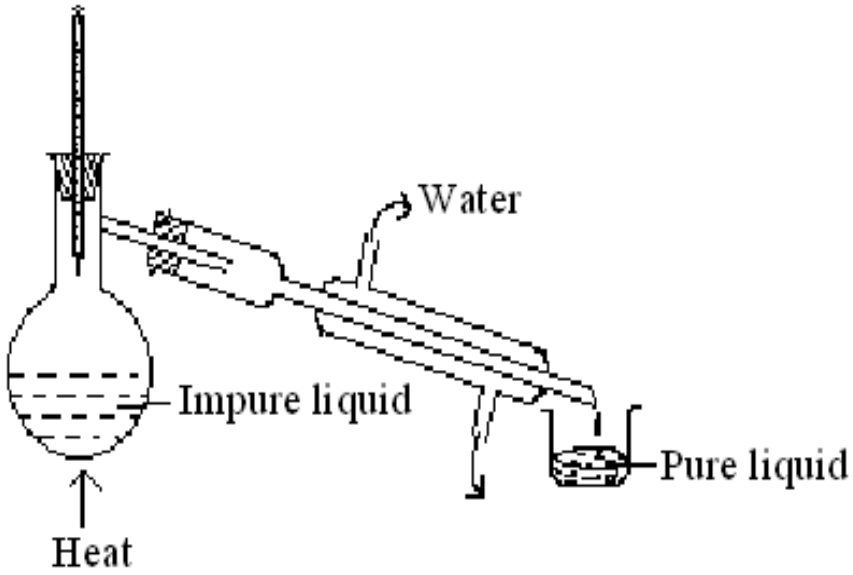
Principle

On heating under constant pressure, say under atmospheric pressure a liquid boils at a temperature at which its total vapour pressure becomes equal to the atmospheric pressure. The temperature of the liquid remains constant till all the liquid distills over. The constant temperature is termed as the boiling point. The distillation process is used for the purification of liquids from non volatile impurities.

Procedure

The distillation apparatus is shown in below figure. It consists of a round bottomed flask with a side tube. The flask is fitted with a one holed bark cork. A thermometer is inserted through the hole so that its bulb is near the opening of the side tube. A water condenser is attached to the tube. The impure liquid is taken in the flask. A few porcelain bits are added to avoid bumping of the liquid. The liquid is heated. When the liquid starts boiling, the thermometer

reading attains constant value equal to the boiling point of the liquid. A receiver is now placed near the end of the condenser and the distillate is collected. The collection of liquid is stopped when the thermometer reading starts to rise. The procedure is repeated with the distillate. We get the liquid in pure form.



FRACTIONAL DISTILLATION

Principle

A mixture of two or more volatile liquids can be separated by fractional distillation.

When their boiling points differ by more than 40^{o}C, they can be separated by fractional distillation. The more volatile liquid passes over first and is collected in a receiver. When the temperature begins to rise for the second time, the first receiver is disconnected; a new receiver is attached as soon as the temperature becomes constant once again. Thus the distillate is collected in fractions and the process is termed fractional distillation. When the liquids present in the mixture have their boiling points close to each other, the separation is best effected by fitting the distillation flask with a fractionating column.

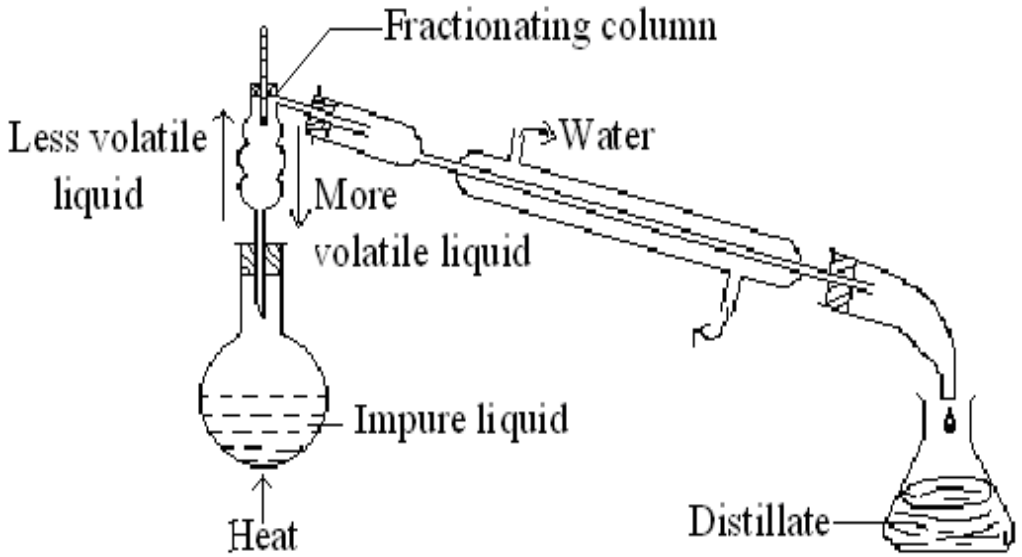
Procedure

The apparatus for fractional distillation is shown in below figure. It consists of a round bottomed flask. The flask is fitted with a one holed bark cork. A fractionating column consisting of a long glass tube blown in to bulbs is inserted through the hole. A thermometer is inserted so that its bulb is near the opening of the side tube of the fractionating column. A water condenser is attached to the side tube of the fractionating column. A receiver is placed near the other end of the condenser. The mixture of liquids to be separated by

fractional distillation is taken in the flask. A few porcelain bits are added to avoid bumping of the liquid mixture. The mixture is heated. The vapours of the liquid (A) with lower boiling point along with a little of the vapours of the liquid (B) with higher boiling point rise up and come in contact with the large cooling surface of the fractionating column. The vapours of B condense first and those of A pass on. The condensed B flows down the column. It meets the hot ascending vapour mixture. The condensed liquid removes more of B from the vapour mixture and gives up any A present in it. This process is repeated in every bulb of the fractionating column. Thus the vapour coming out at its top consists of nearly pure A. The liquid in the flask is nearly pure B.

Example

A mixture of benzene and toluene can be separated by this method.



Vaccum distillation

The boiling point of a liquid is that temperature at which the total vapour pressure is equal to the external pressure. From this it follows that when the external pressure, the pressure above the liquid is reduced, the boiling point will be decreased. When vaccum is applied (pressure = 0), the boiling point should be the minimum. At intermediate pressures the boiling points would be intermediate. Normally, distillation under diminished pressure is effected.

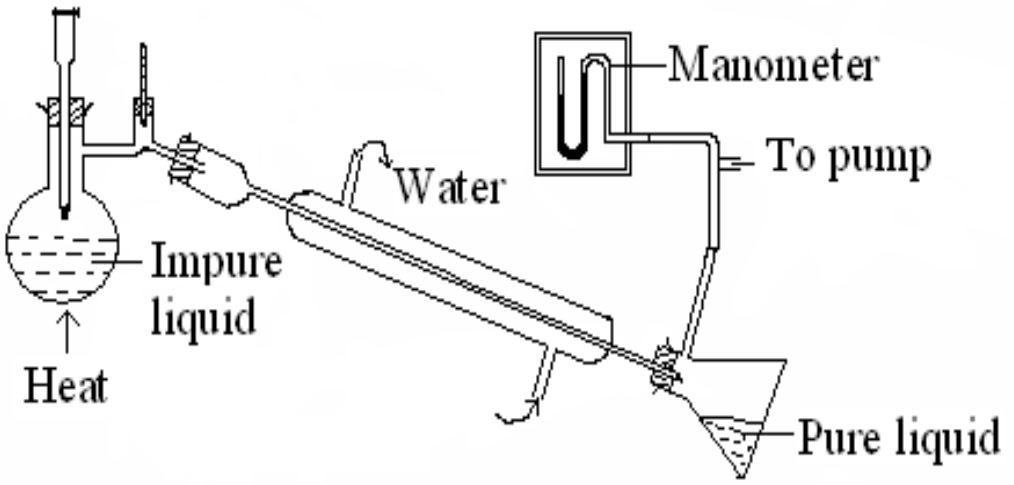
1. Certain liquids have very high boiling points and therefore distillation of these under vaccum is easy; distillation of these under atmospheric pressure would be slow and laborious.
2. In many cases, boiling temperatures at atmospheric pressure are too high to be desirable, because the compound is distilled may get oxidized, undergoing molecular rearrangement or decompose.

3. Sometimes, impurities present in the liquid may catalyze some reactions or may themselves react with the liquid at high temperatures.

TECHNIQUES

The apparatus for distillation under reduced pressure is shown in figure. It consists of a Claisen flask having two necks. Through one neck a long jet is inserted so that it dips in the liquid to be purified. During distillation the vapours of the liquid bubble up through the capillary of this jet. This prevents bumping. One end of a water condenser is connected to the Claisen flask. The other end of the condenser is connected to a filtration flask, which serves as the receiver. The receiver is connected to a pump and mercury manometer. The pressure in the apparatus is reduced using a water pump or a mercury pump. Now the distillation is carried out in reduced pressure.

This method is used to purify liquids which deompose near their boiling points. For example, glycerol from spent – lye in soap industries is recovered by this method. Glycerol decomposes at its boiling point (298^{o}C) but can be distilled unchanged at 12 mm pressure when it boils at 180^{o}C.



Steam distillation

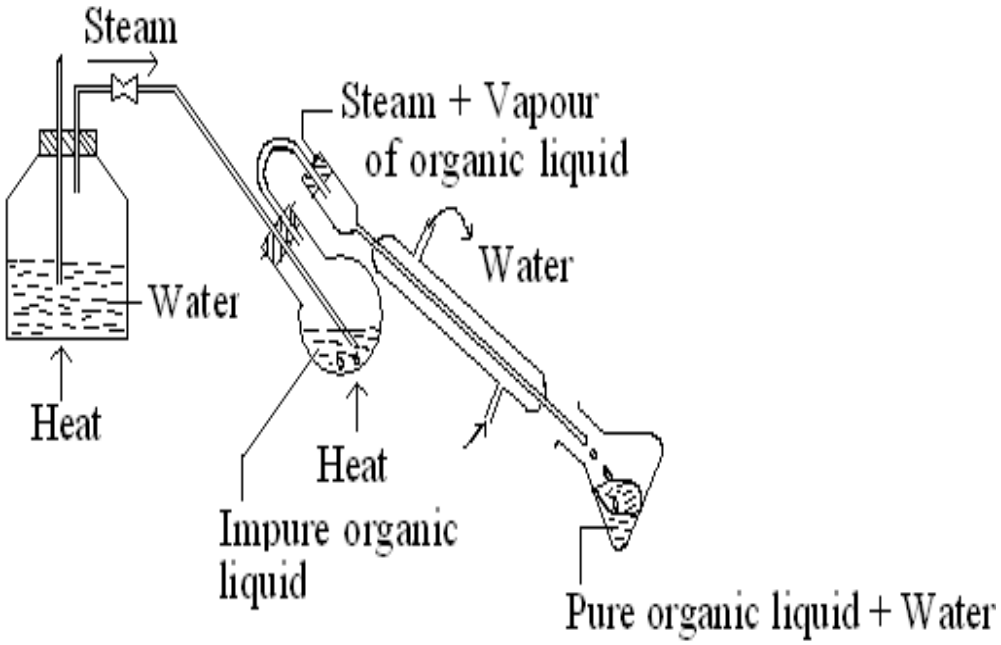
Principle

Many substances that are insoluble in water and are volatile in steam can be purified by distillation in a current of steam. The non – volatile impurities are left behind, in the distillation flask. A liquid boils when its vapour pressure is equal to the atmospheric pressure. In steam distillation, a mixture of water and an organic liquid is heated. The mixture boils when the combined vapour pressure of water (P_1) and that of the organic liquid (P_2) is equal to the atmospheric pressure (P) i.e., $P = P_1 + P_2$. Naturally, the boiling temperature of the mixture would be lower than the boiling temperature of the

organic liquid when the vapour pressure of this liquid alone would be equal to the atmospheric pressure.

Procedure

The apparatus for steam distillation is shown in figure. It consists of a round bottomed flask with a two holed bark cork. Through another hole a water condenser and receiver are attached. The flask is placed in a slanting position in such a way that the mixture which is steam distilled does not splash in to the condenser on bubbling steam through it. The mixture is taken in the flask and is gently heated to maintain the required temperature so that unnecessary steam condensation in the flask is avoided. Steam is passed through it.



The steam carries with it the steam volatile constituent in the mixture and enters the condenser where it condenses. The condensed water and the steam distilled liquid collect in the receiver as two immiscible layers. The distillate is then treated to recover the organic substance by a suitable method. In case it is a solid, the substance may be separated by simple filtration. If it is a liquid, it can be removed by means of a separating funnel. The aqueous layer in both cases may be extracted with a solvent.

Thus in steam distillation, the liquid is distilled at a lower temperature than its boiling point, when it might decompose. It serves the same purpose as distillation under reduced pressure. Example, Aniline is purified by this method.

Applications

Steam distillation is employed in industry for the recovery of various essential oils from plants and flowers. It is also used in the manufacture of aniline and turpentine oil.

Azeotropic distillation

An azeotrope is a mixture of two or more miscible liquids having constant boiling point and constant composition. For an azeotrope, the composition of the liquid phase is identical with that of the vapour phase. For example, a mixture containing 80% by weight of chloroform and 20% by weight of acetone boils at 64.7^0C. This is an azeotropic mixture. Several other liquids form azeotropes, water – ethyl alcohol, water – pyridine and phenol – aniline. The boiling point of an azeotrope may be lower than that of its components and such an azeotrope is called azeotrope of minimum boiling point. Water (100^oC) and ethanol (78.3^oC) form a minimum boiling (78.15^oC) azeotrope. The boiling point of an azeotrope may be higher than that of its components. Such an azeotrope is called maximum boiling azeotrope. For example, water (100^oC) and formic acid (100.8^oC) form an azeotrope of maximum boiling point (107.1^oC).

Technique of azeotropic distillation

Azeotrope formation is sometimes used for separating a liquid component from a mixture of liquids by distillation. For example, absolute ethyl alcohol is obtained in some industries by azeotropic distillation. To the alcohol contaminated with water, a calculated amount of benzene is added and the mixture is distilled through a fractionating apparatus. Benzene removes water from alcohol by forming a ternary azeotropic mixture of water, alcohol and benzene of minimum boiling point (64.85^oC) and containing 7.4% of water, 18.5% of alcohol and 74.1% of benzene. Once this azeotrope is distilled off, a second azeotrope, benzene-alcohol boiling at 68.25^oC, containing 32.4% of benzene distills off. At the end of this, absolute ethyl alcohol distills over. In this method a part of the alcohol is lost in forming the two azeotropes. An azeotrope may be broken in to its components by a chemical method, by a physical method, preferential adsorption, fractional extraction or fractional freezing.

Benzene can be freed from moisture by azeotropic distillation. Adding a calculated amount of ethanol and distilling the mixture will remove all the water in the form of a ternary azeotrope. Solids can sometimes be dried by azeotropic distillation. The wet solid is dissolved in a non – aqueous solvent which can form an azeotrope with water. Then water is removed by azeotropic

distillation. The solid would remain in the non – aqueous solvent free from moisture. It can be recovered in a dry form by evaporating the solvent.

CRITERIA AND TESTS FOR PURITY

A substance purified by any of the techniques described in the previous sections of this chapter has to be checked for its purity. The following physical properties are used for checking the purity of substances.

1. Melting point (mp)
2. Boiling point (bp)
3. Refractive index (η)
4. Density (d)

These properties have long been utilized in identification and charcterisation of organic compounds.

1. Melting point (mp)

The melting point of a substance is the temperature at which the liquid and solid phases exist in equilibrium with each other. The melting point of a pure substance is a constant and not affected by experimental conditions. It is an intensive property, i.e., not dependent on the quantity of the substance used.

In dealing with an unknown compound, it is usual to crystallize it repeatedly until its melting point is no longer affected by subsequent recrystallizations. As a rule, the compound will then have a sharp, definite meltin g point and is considered pure. A few substances have indefinite melting points. They begin to soften at a certain temperature and get completely molten only at a much higher temperature. As a rule, a melting point which is not sharp is due to the presence of an impurity.

(i) Decomposition and Melting point

The rate of heating may affect the melting point of a substance and therefore very different melting points may be obtained for the same substance owing to the different rates of heating.

For example

The melting point of phthalic acid has been given in the books at various temperatures ranging from 180^{o}C to 200^{o}C. This variation is especially possible when a substance decomposes on heating.

(ii) Isomerization and melting point

In some rare cases, two melting points are observed for the same sample. The substance melts first, solidifies and then melts once again at another temperature. This is due to the conversion of the substance at its melting point in to an isomer which then solidifies and then melts. In such a

case the lower melting point is that of the starting material and the higher melting point that of the isomer.

Precautions in melting point determination

1. Only a small amount of the substance should be used. The common mistake is to use too much material: then the temperature of the bath may rise through several degrees whilst the whole of the substance is melting unless the temperature is carefully regulated.
2. The temperature of the bath must be raised slowly by means of a microburner; rapid heating may lead to inaccurate melting points.

2. Boiling point (bp)

The boiling point of a pure substance under a particular is a constant and therefore this property can be used to check the purity of a liquid.

The boiling point may be determined to test the purity of a liquid. A sharp boiling point indicates that the substance is pure. Liquids which do not have sharp boiling points are impure.

Precautions in determination of Boiling point

(i) The whole of the thermometer bulb should be exposed to the hot vapour during distillation. The thermometer is arranged such that the bulb is just below the side tube. It should not be immersed in the liquid.

(ii) Superheating of the vapour must be avoided. When a low boiling liquid is boiled on a water bath, the steam issuing may surround the flask and superheat the vapour. As a rule, a small direct flame from a microburner is the best. The flame should be directed in such a way that it comes in to contact with only the portion of the flask filled with liquid.

(iii) The bulb of the thermometer should always be covered with a film of pure liquid produced by the condensation of vapour over the thermometer. This thin film of liquid will be in equilibrium with the pure vapour and hence the correct boiling point is registered.

(iv) Bumping should be avoided and regular boiling is facilitated by adding one or two pieces of porous bits.

3. Refractive index (η)

The velocity with which light travels in different media is not a constant. This leads to bending of a beam of light as it passes through the interface of two different media. The change in the ‘speed’ of light causes this bend. This is called refraction. The angle of refraction is dependent on the density, type of molecule and the temperature of medium and also on the

wavelength of light. Refractive indices are measured with an instrument called the refractometer. The refractive index is given by the equation.

$$\eta = \sin i / \sin p$$

where i is the angle which incident light in air makes with the perpendicular to the interface and p is the angle which the refracted light.

A pure liquid has a constant refractive index at a particular temperature. Even traces of impurities alter this value and therefore measuring the refractive index of a sample will indicate whether the sample is pure or not. The measured index may be compared with that obtained with a pure sample of the same compound or with the reported value.

4. Density (d)

The density of a substance is occasionally used to check the purity of the substance. Density is a measure of concentration of matter. This is reported in units of grams per milliliter at 20^{o}C. The density of a liquid is measured using a small container called a pyknometer whose volume is accurately known.

The pyknometer is weighed accurately and then filled with the liquid whose density is to be determined. The temperature of this is brought to 20^{o}C, the volume is readjusted, if necessary, and then it is weighed. The density may be calculated by dividing the weight in grams by the volume in milliliters.

Important questions

1. Write an essay on column chromatography
2. Write the various types of adsorbents used in column chromatography
3. How will you prepare a TLC plate?
4. What are the various adsorbents and solvent used in thin layer chromatography
5. What are the factors affecting R_f values
6. What are the significance of R_f values
7. Write an essay on paper chromatography
8. How will you prepare the paper chromatogram?
9. What are the ascending, descending and radial paper chromatography
10. Write notes on paper electrophoresis
11. How will you separate amino acids by paper electrophoresis?
12. What is the principle of ion exchange chromatography?
13. What are the resins used in ion exchange chromatography?

14. How will you separate Ca and Mg by ion exchange chromatography?
15. Write notes on analysis of milk by ion exchange chromatography?
16. How will you separate chloride and bromide by ion exchange chromatography?
17. Discuss the theory and applications of gas chromatography
18. Write the principle and applications of HPLC
19. How will you purify an organic compound soxhlet extraction?
20. Write notes on fractional crystallization
21. Write the principle and advantages of sublimation
22. Write notes on vacuum distillation and steam distillation
23. Write notes on fractional distillation and azeotropic distillation
24. Write an essay on purification of liquids

NOTES

UNIT – III

ELECTRO ANALYTICAL METHOD

3.1. POLAROGRAPHY

It is an electroanalytical technique to study the composition of a dilute solution of an electrolyte by plotting current – voltage curves. It is also known as voltammetry.

Polarisable electrodes

These electrodes have low exchange current density for a given reaction. There is little flow of charge in to the solution if the applied potential differences across the electrode are increased. The charges remain in the double layer and increase the potential difference across it.

Example

Mercury electrode in KCl solution.

Non – Polarisable electrodes

These electrodes have a high exchange current density for a given reaction. There is an increased flow of charge in to the solution if the applied potential differences across the electrode are increased. The potential difference across the double layer is not altered (i.e.,) the charges moves rapidly to and from the electrode.

Example

Calomel electrode

Principle

During electrolysis of a solution of an electrolyte the products of electrolysis collect at the electrodes and set up an e.m.f. opposite to the applied e.m.f. This is known as Polarisation.

For instance when a small e.m.f. is applied to the cell consisting of Pt electrodes dipped in dilute H_2SO_4 electrolysis sets in and a small current flows through the circuit. But soon the electrodes are surrounded by the bubbles of the gas which acts as gas electrode and sets e.m.f. in the reverse direction, consequently, the electrolysis stops. In the case of electrolysis of $CuSO_4$ solution using Cu electrodes, to begin with a very small e.m.f. is sufficient to start electrolysis but after a very short time it is observed that concentration of $CuSO_4$ near the cathode the anode is different. This results in the setting up of a concentration cell with e.m.f. opposite to the applied e.m.f. Soon the electrolysis stops, so to bring about the process of electrolysis continuously, a minimum e.m.f. has to be applied.

The minimum e.m.f. which must be applied to bring about electrolysis of solution of a particular electrolyte continuously is known as the decomposition potential or discharge potential. A graph is drawn between the applied voltage and the current passing through the solution. The resulting graph is known as polarogram. From the graph the composition of the solution can be determined.

Concentration polarisation

As the electrolysis proceeds, the ions get deposited at the electrodes. Due to this, the concentration of ions around the electrode becomes less. This is more pronounced if the solution is not properly stirred. This results in a difference in concentration of ions. The difference in concentration may set up a concentration cell working in the opposite direction. This will end up in polarization. This type of polarization is known as concentration polarization.

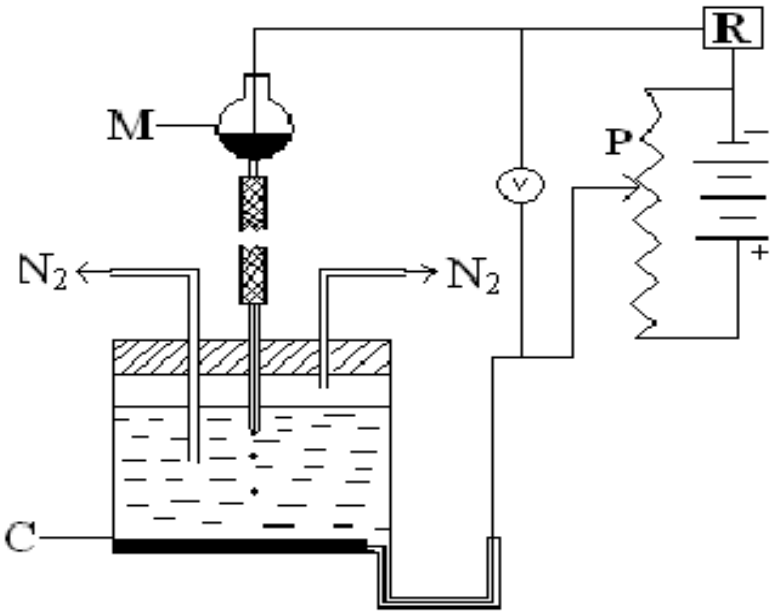
Significance

Concentration polarization causes over voltage. So to cause electrolysis we will have to apply more potential than the theoretical deposition potential.

Methods to minimize concentration over voltage

1. Electrodes with large surfaces areas are to be used.
2. Currents for electrolysis must be kept small.
3. The solution must be stirred well during electrolysis
4. Electrolysis must be conducted at higher temperatures.

Experimental assembly



C – Cell; M – Mercury Reservoir; P – Potentiometer; R – Recorder; V – Voltammeter

Dropping Mercury Electrode (DME)

A schematic diagram of a polarograph is shown in figure. It consists of a mercury reservoir from which mercury trickles down as small drops through a capillary. This acts as the cathode. It is also called an indicator electrode or micro electrode. The anode consists of a pool of mercury. This is called reference electrode. The concentration polarization at the anode is negligible because of the large area. The anodic potential may therefore be regarded as constant. Hence, the polarization of the whole cell is governed by reactions occurring at the DME (micro electrode).

DME has the following advantages

1. The surface area is reproducible with any given capillary.
2. Constant renewal of the electrode surface eliminates passivity.
3. The hydrogen over voltage on mercury is extremely high so that there is no interference for the discharge of metal ions at mercury electrode.
4. The extent of electrolysis is so small owing to the small surface area.
5. Mercury forms amalgams with many metals and lower their reduction potential.
6. The diffusion current assumes a steady value immediately and is reproducible.

Disadvantages of DME

1. The cathodic versatility of mercury is not matched by its anodic behaviour.
2. Oxygen is reduced by a two stage process giving two polarographic waves occupying a considerable range of voltage which interfere with those of the sample. This difficulty is overcome by passing nitrogen through the solution just before the experiment.
3. Mercury is toxic

Capillary

The capillary should be of such length and internal diameter that drop rate is of the order of 2 to 6 seconds. Usually capillary with hole about 0.02 to 0.05 mm and length about 5 to 12 cm are used.

Solutions

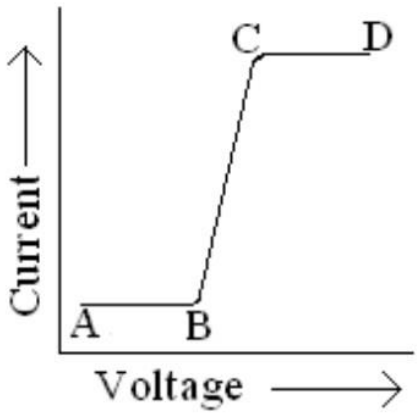
The solution of the substance under study should be above 10^{-3} M. In modern sophisticated instruments even solutions up to 0.2 to 2 X 10^{-6} M can be an analysed. 0.005 to 0.01% gelatin and 0.5 M or larger concentration of supporting electrolyte are added to the solution under study. Under these conditions the diffusion current will be directly proportional to the concentration of the substance under study.

The rate of discharge of ions at DME is controlled by two factors

1. Diffusion of the substance from the bulk to the vicinity of the electrode – called diffusion current (i_d).
2. Migration of charged particles in the electric field caused by the potential difference between the electrode and solution – called migration current (i_m).
3. The total current passing through the cell $i_{total} = i_d + i_m$.
4. The solution for the polarographic analysis always contains a large excess of an inert electrolyte called the supporting electrolyte (e.g., KNO_3 or KCl) having high decomposition potential so that most of the current is carried by the ions of the supporting electrolyte, eliminating the migration current (i_m) practically.

POLAROGRAM / CURRENT VOLTAGE CURVE

The essential features of the wave like current – voltage curve known as polarographic wave or polarogram are shown in the figure:



In the polarogram

(a) Rising portion of the curve AB represents the residual current which includes contributions from

(i) The condenser current (ie) the current required to charge the individual drop to the applied potential.

(ii) Electrolysis or Faradic current, resulting from the reduction of impurities such as dissolved oxygen, mercury ions etc.

(b) The potential represented by point B may be used to characterize a metal but decomposition potential (B) is concentration dependent and hence cannot serve the purpose.

(c) The portion of the curve BC represents the diffusion current (i_d). The height of this portion helps to determine concentration.

(d) The portion CD represents a state of extreme concentration polarization and is called limiting current. The factors that affect the limiting current are

(I) Residual or Condenser current

(ii) Migration or Electric transference current

(iii) Diffusion current

(iv) Kinetic current

Residual or Condenser current

The current flowing between the electrodes when the applied voltage is lower than the decomposition voltage is called residual current. The current is caused by the same electrode process as in the case of voltage higher than the decomposition voltage, but at a low rate due to high electrode polarization and also by deposition of impurities at the electrodes. At voltage lower than equilibrium potential differences the residual current is caused by the discharge of solution impurities.

At any potential, the magnitude of current indicates the energy required to set up the electrical double layer of cations and anions and to maintain this structure against the disordering effects of the growth of the drop and the Brownian motion in solution.

The electrical double layer is similar to an electrical condenser and the current needed to develop and maintain this layer is known as condenser current.

Migration current

There are two processes by which electro active material reach the surfaces of respective electrodes:

(a) One involving the movement of charged particles in the electric field caused by the potential differences exists between the electrode surfaces and the solution.

(b) The other involving diffusion of particles.

The current needed for both these processes is called migration current. This current can be totally eliminated by adding indifferent electrolytes to the solution. The concentration of indifferent electrolyte added should be so large that its ions carry essentially all the current. The indifferent electrolyte added is called supporting electrolyte. Generally, the concentration of supporting electrolyte added is 100 times that of the electro active material.

Diffusion current

The rate of electrode reaction is determined by the transport of the discharging particles to the electrodes, solution concentration in the layer near the electrode, outside the electrical double layer changes. The solution near the electrode is depleted with discharging particles and the concentration polarization of the electrode occurs.

Diffusion current is directly proportional to the concentration of the substance being reduced or oxidized at the dropping electrode. Thus

Diffusion current = Limiting current – Residual current.

Kinetic current

The limiting current is also affected by the rate of non – electrode reaction and is known as kinetic current. This type of current results when oxidized or reduced form of electro active species is involved in a chemical equilibrium with other species.

Ilkovic equation

The diffusion current is giving by Fick's Equation

$$i = i_{diff} = Z.F.D. (C_o - C_b) / \delta_{diff}$$

where Z is the charge of diffusing ion, F is the Faraday constant, D is the Diffusion coefficient, C_o is the bulk concentration outside diffusion layer, C_b is the concentration of the solution at the boundary with the electrical double layer, mol / m^3, i_{diff} is the thickness of diffusion layer.

The equation for polarographic diffusion current was given by Ilkovic in 1934 and is known as Ilkovic equation. According to

$$i = i_{diff} = 607nD^{1/2} Cm^{2/3} t^{1/6}$$

where,

i = i_{diff} = Average diffusion current in micro amperes during the life of a drop.

n = Number of Faraday required per mole of electrode reaction

D = Diffusion coefficient in cm^{-2} sec^{-1}

C = Concentration of substance in millimoles per litre.

m = Rate of flow of mercury from DME in mg / sec.

t = Drop time in sec.

Significance

1. From Ilkovic equation we find that $i_d \alpha C$ or $i_d = KC$ where K is a constant for a given set of conditions of temperature. That is, the observed diffusion current is directly proportional to the concentration of electro reducible substance. This fact forms the basis of quantitative polarographic analysis .
2. The diffusion current is proportional to the product of $m^{2/3}$ and $t^{1/6}$. This permits the comparison of results obtained when different capillaries are used, keeping the other conditions same.

Oxygen wave

Oxygen is capable of reduction first to H_2O_2 and finally to H_2O. So it gives polarograms. So when oxygen is present in the solution under observation we get one extra wave in the polarogram. This polarographic wave got is called oxygen wave. Thus polarography provides a way to determine the amount of dissolved oxygen in solution. But, at the same time when we are determining other reducible species in a solution, presence of oxygen in a solution is a nuisance. Hence it must be removed. This is done by passing an inert gas like nitrogen or hydrogen before doing the polarographic study.

Influence of temperature and agitation on diffusion layer

The standard temperature chosen for polarographic work is 25^{o}C. The diffusion co-efficient of many ions changes 1 to 2% per degree. Thus the influence of temperature on diffusion current is quite marked. Therefore the temperature must be controlled to within ± 0.5^{o}C around 25^{o}C.

Agitation or stirring of the solution causes a significant variation in the thickness of the diffusion layer. This produces an abnormally large current. A faster drop rate causes appreciable stirring or agitation. This is controlled by adding gelatin. Addition of gelatin decreases the drop time to the neighbour hood of 1.6 sec.

Half wave potential

Half wave potential is defined as “the electrode potential at a polarizing current equal to half of the limiting current” or “the potential corresponding to the mid point of the wave” is called half wave potential.

The potential at which a break will appear on the polarographic wave depends on the concentration of reducible substance. Hence the potential at the beginning of reduction is not a convenient characteristic of the substances. In the figure it may be noted that there is a shift of potential due to increasing

concentration. But all the curves in the figure are symmetrical with respect to a certain average potential called half wave potential.

It is used for the qualitative analysis of the substance. Quantitative measurement is based on the measurement of the height of the polarographic wave, i.e., of the diffusion current. For quantitative determination, a standard graph drawn between the heights of the polarographic waves corresponding to the different concentrations of the same ion can be used.

It has been found that the half-wave potential is a constant independent of the concentration of the reducible ion. It depends only on the nature of the reducible ion.

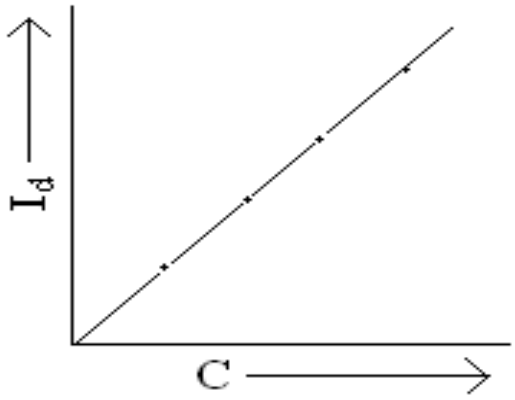
Polarography as an analytical tool in quantitative and qualitative analysis

1. Qualitative analysis

Each ion has got a characteristic half wave potential value. Thus from half wave potential values we can identify the ion qualitatively. Further from number of waves we can determine the number of ions present in a mixture.

2. Quantitative analysis

Diffusion current is directly proportional to the concentration of the ions undergoing reduction at the dropping mercury electrode. The wave height is a measure of diffusion current. Different solutions with different concentrations are analysed polarographically keeping the other conditions of the experiment unaltered, and the respective wave heights are determined. The wave heights are plotted against respective concentrations. We get a straight line. The solution whose concentration is to be determined is analysed polaragraphically and the wave height is determined. Interpolating this value in the graph, the concentration of the given solution can be found out. Thus we can estimate quantitatively the given solution.



I_d – Wave height of diffusion current; C - Concentration

Example: Determination of C_d by direct comparison method

Cadmium solutions of different concentrations in the concentration range of 10^{-2} to 10^{-3} moles per liter are prepared. Each solution is mixed with a known quantity of KCl (I mol) and gelatin (1%). Unknown solution of Cd is also mixed with KCl and gelatin. The polarograms for all the solution of constant volume are recorded. The wave height in each case is measured. A linear calibration curve between the concentration of Cd(II) in the standard solutions and their wave heights is drawn. The concentration of the unknown Cd(II) solution can be known from its wave height by interpolation method.

3.2. AMPEROMETRIC TITRATIONS

AMPEROMETRY

It is an electrometric titration based on polarographic method. The existence of a simple relationship between the concentration of a substance and its polarographic current was used for developing a sensitive and convenient analytical tool called amperometry.

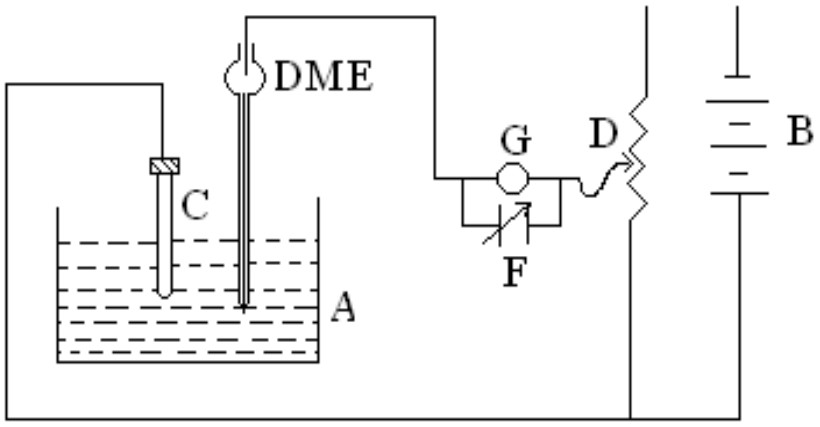
Basic principle

There are certain reactions in which the end point cannot be determined potentiometrically or visually and for reactions in which the product is some what soluble (as in precipitation titrations). For such reactions amperometric titrations are useful.

If some of the electroactive material is removed by the addition of another reagent, the diffusion current decreases. This forms the basis of amperometry. In amperometric titration the voltage applied across the indicator electrode and the reference electrode is kept constant, the current passing through the cell is measured and plotted against the volume of the reagent added.

Apparatus

Amperometric titrations are carried either with a dropping mercury electrode or a rotating platinum electrode. Before carrying out the titration, the potential at maximum diffusion current should be ascertained by polarographic measurement.



DME – Dropping Mercury electrode; A – Titration cell (Indicator Electrode); B – Battery; C – Calomel Electrode; D – Voltage Divider; F – Condenser (To reduce the fluctuation in the voltage) G - Galovanometer

The potential of the indicator electrode (DME) must be within 1 volt of the potential that corresponds to maximum diffusion current of the solution to be titrated. The galvanometer needle is set either at maximum or at minimum depending upon the nature of the titration. After each addition of the titrant, the galvanometer readings which are proportional to the current are noted and plotted against the volume of the titrant.

Advantages of amperometry

1. The titrations can be carried out rapidly.
2. Results obtained are independent of characteristics of capillary.
3. Range and sensitivity are higher than conductometry and potentiometry.
4. Accurate results are obtained.
5. Though temperature need not be known, it must be kept constant.
6. Presence of foreign substances causes no interference.
7. Highly dilute solutions can be titrated.

Disadvantages

1. Co – precipitation and post precipitation errors are introduced.
2. Relative changes in current are small in presence of excess supporting electrolyte.

Typical examples

1. Titration of reducible substances like lead (II) ions with a non – reducible reagent like sulphate ions. Pb^{2+} ion being reducible at cathode gives diffusion current. The concentration of Pb^{2+} ions and consequently the diffusion current is diminished as SO_4^- ions are

added. When all the Pb^{2+} ions are precipitated as lead sulphate the current value remains a constant at its residual value which is characteristic of supporting electrolyte. (fig. 1)

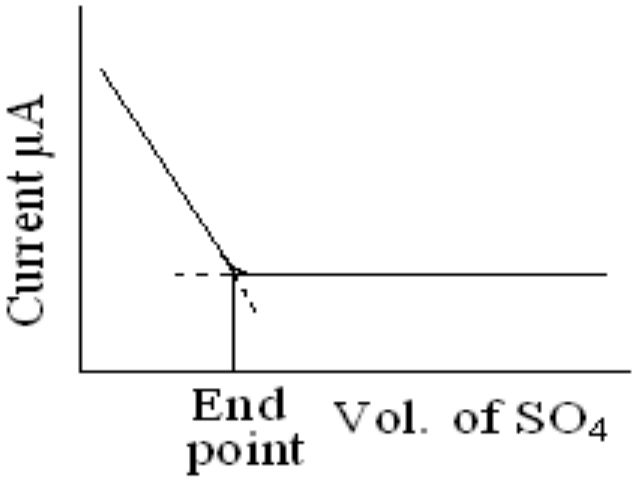


Figure. 1

2. If Pb^{2+} ions are added to a solution containing SO_4^- ions, the current remains constant until all the SO_4^- ions are precipitated as lead sulphate and then increases due to the addition of electro reducible Pb^{2+} ions. (fig. 2)

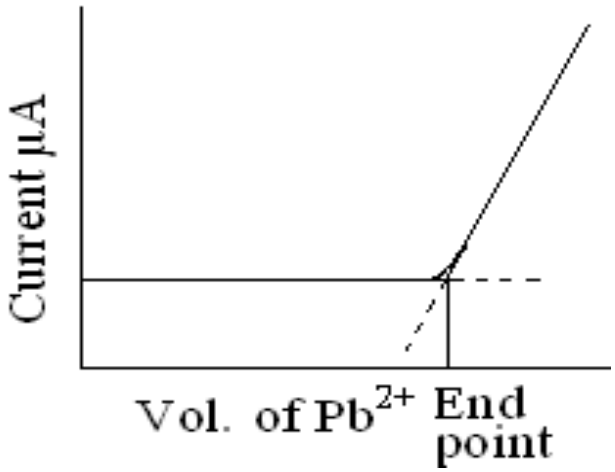


Figure. 2

3. In some cases the reaction product (I_2) is reducible at the cathode. In such cases the diffusion current goes on increasing with the addition of the titrant (arsenate ion) and becomes constant at the end point (e.g.,) Titration of arsenate ion Vs I^- ion. (fig. 3)

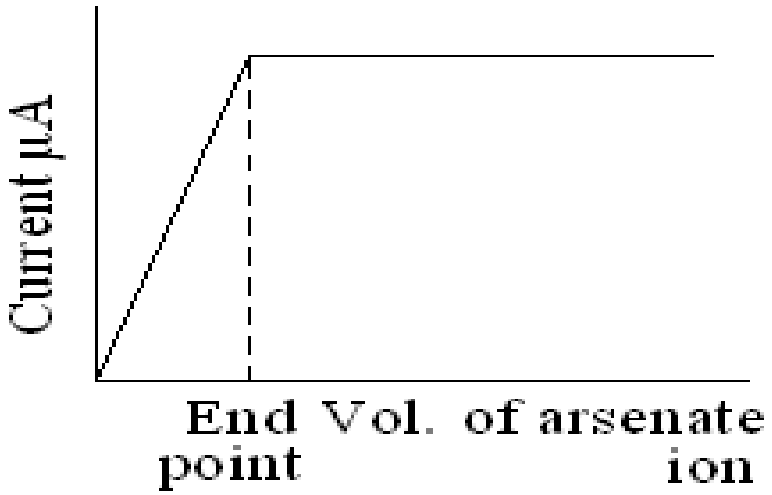


Figure. 3

4. When both the ions of the titrate and titrant are reducible at cathode, the current decreases first and then increases after the end point. (fig. 4)

   Example

   Molybdenum, Lead, Ni (II) salt solutions Vs diacetyl dioxime.

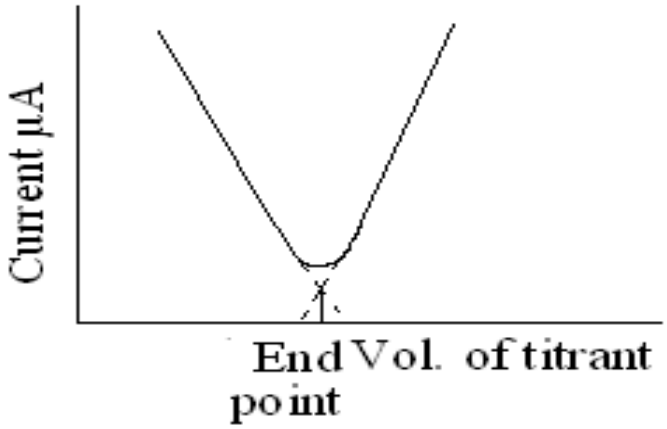


Figure. 4

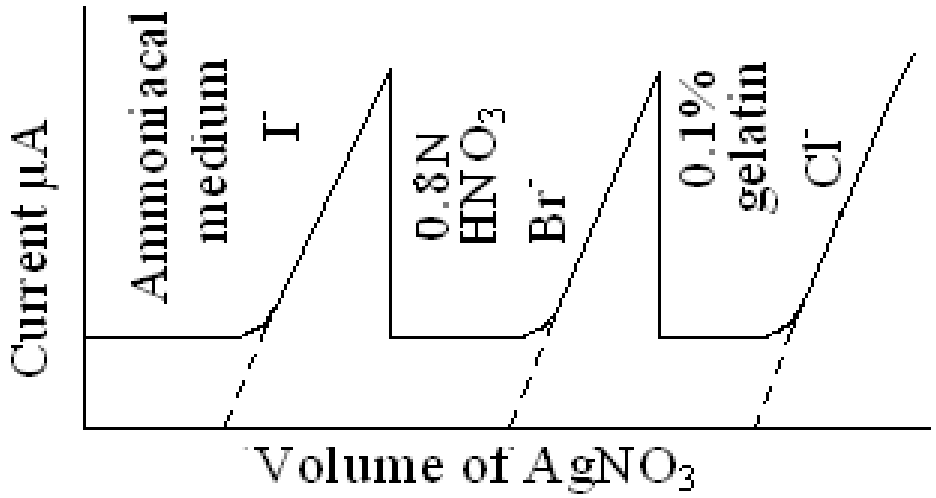
5. Another important type of amperometric titration is successive titration.

   Example

   Titration of a solution of chloride, bromide and iodide Vs $AgNO_3$.

   In ammoniacal medium, the current remains constant at about zero until all the iodide ions are removed as silver iodide and then rises. After three or four points in the rising part of the current, the solution is acidified with 0.8N nitric acid and the titration is continued. Immediately silver bromide gets precipitated and the

current drops to zero again and remains constant until Br- ions are removed as AgBr. After a few points in the rising part of the current, a 0.1% gelatin is added to the titration vessel while the chloride ions get precipitated as AgCl and the current again drops to zero and it remains constant until all the chloride ions are removed as AgCl. Finally the current rises again due to the addition of electro reducible Ag^+ ions. Thus there are three successive rises in the curve which indicate the corresponding end points.



APPLICATIONS

Cl^- can be estimated using standard $AgNO_3$ using this method. Pb^{2+} and Ba^{2+} can be estimated using standard K_2CrO_4. (Here, DME is used as the indicator electrode).

3.3. THERMOANALYTICAL METHODS

Thermal analysis

In thermal analysis substances are heated. They undergo physical or thermal changes. These changes are recorded as a function of temperature or time. The two important thermal analysis are

1. Thermogravimetric analysis (TGA)
2. Differential thermal analysis (DTA)

Principle of thermogravimetric analysis

The substance is heated or cooled in a given environment at a controlled rate. The weight of the substance is recorded as a function of time or temperature. A graph is plotted between weight change and temperature or time. We get a thermogravimetric curve or TG curve or thermogram.

On heating a substance, various physical and chemical bonds are formed or broken resulting in a change in weight. A weight loss indicates the formation and a subsequent escape of a volatile product or the formation of some other product.

Principle of differential thermal analysis

The substance to be analysed and an inert reference material like α – alumina are heated or cooled in a given environment and at a controlled rate. Whenever the substance to be analysed undergoes an endothermic change, like when it melts or when it is dehydrated, the temperature of the substance will be lower than that of the reference material. On the other hand, if the substance to be analysed undergoes an exothermic change its temperature will be higher than that of the reference material. When the substance under observation does not undergo any heat change, there will be no difference in temperature between the sample and the reference material. These temperature differences, ΔTs, are plotted against temperature or time. We get DTA – curves.

Instrumentation – discussion of various components with block diagram

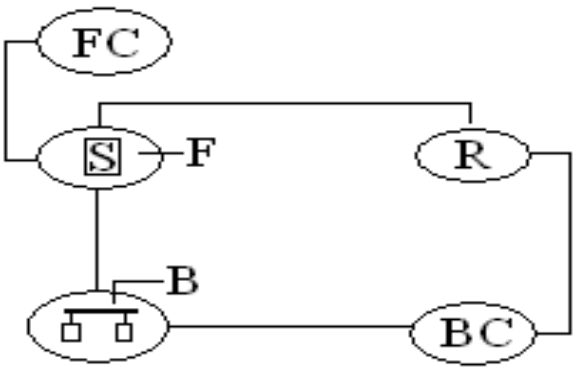
TGA instrumentation

The instrument used for TGA consists of

(i) A furnace

(ii) A precision thermo balance

(iii) A recorder



Block diagram for TGA

FC – Furnace control; S – Sample container; F – Furnace; R – Recorder;

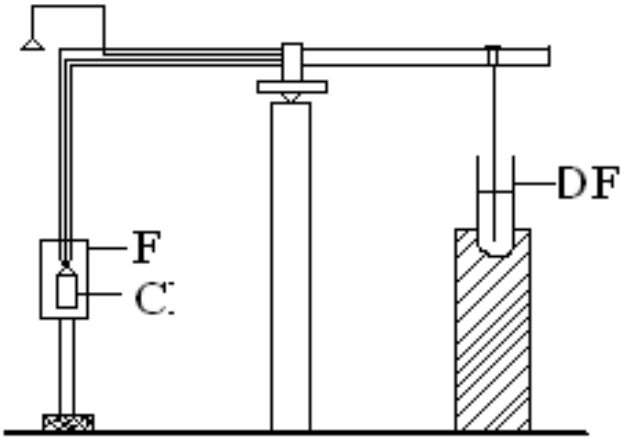
B – Balance; BC – Balance recorder

(i) Furnace

The furnace is controlled by a furnace control so that the furnace can be heated in such a way that the temperature rises linearly with time.

(ii) Thermobalance

A platinum crucible(C) is suspended from one arm of the thermobalance (figure). The other arm of the balance is prevented from oscillating by attaching it with a Dewer flask (DF) through a spiral, so that it acts as a damper. A known weight of the sample is taken in C. C is enclosed by a furnace F. The furnace temperature is raised at a slow and steady rate. The temperature of the sample and the corresponding weight are monitored as follows:



A platinum / platinum – rhodium thermocouple is used to measure the sample temperature. It monitors the furnace temperature continuously and sends signals to the recorder.

Changes in weight are recorded from the beam deflection. This is done with the help of detectors. They contain a sensing element which detects a deviation of the balance beam from its null position. Normally a detector consists of a pair of photocells, a slotted flag connected to the balance arm and a lamp. Any change in sample weight causes the balance to rotate. This moves the flag so that the light falling on each photocell is no longer equal. The resulting signal is amplified and fed as current to a motor to restore the balance to equilibrium. This current is proportional to the weight change which is recorded by the recorder.

In another type of detector, a helical spring is used to detect the weight change. The spring undergoes contraction or elongation with weight change. This movement is detected by the movement of an attached core in a linear variable differential transformer.

(iii) The recorder

A recorder is a device with a pen and graph sheets. It records the weight change in the Y – axis. It records the signals from the thermo couple, on the X – axis, we get a thermogram.

Precautions to be observed in using a thermo balance

1. The hot zone of the furnace should be uniform for sufficiently long time.
2. The crucible must always be inside the hot zone.
3. The heating rate should be linear and reproducible.
4. Conducting and magnetic samples must not interfere with the furnace electrical winding.
5. The weighing unit must not be affected by radiation and convection currents from the furnace.
6. Attack of the apparatus by volatile products from the sample must be prevented.

Characteristics of TGA curve

TG curves are characterized by breaks (A'B') and plateaus, (A'A and B'B) as shown in below figure. Each break corresponds to some loss in weight, due to evolution of H_2O, CO, CO_2 etc., and each plateau corresponds to the formation of stable compounds. These can be illustrated by taking the TG – curve of calcium oxalate monohydrate. The weight – change curve for calcium oxalate monohydrate is shown in figure a. heating rate is 6^oC / min.

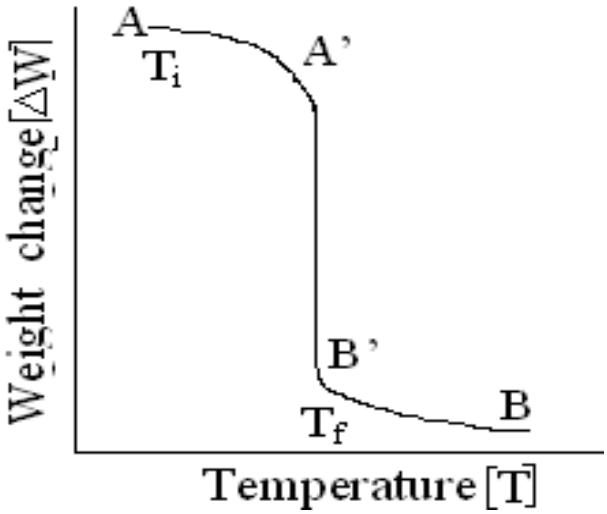
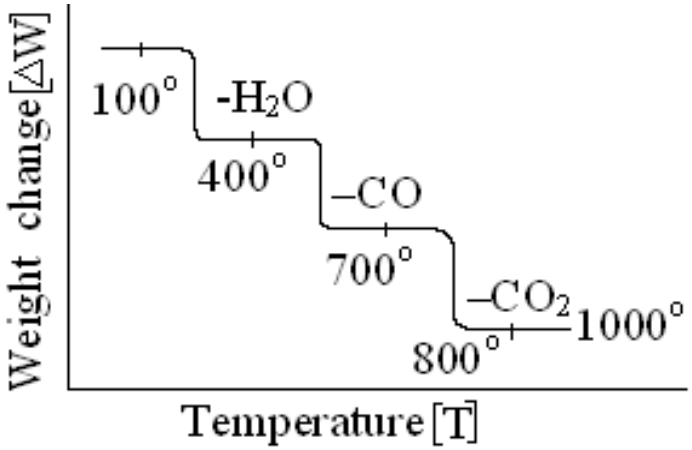


Figure a

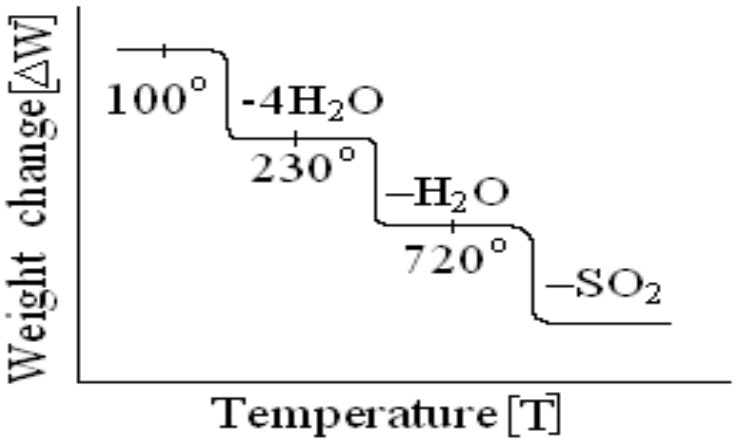
TGA curve for $CaC_2O_4.H_2O$



At about 100°C water is evolved. At about 250°C anhydrous salt is got. Between 500 – 600°C calcium carbonate is present. About 870°C calcium oxide is present.

$$CaC_2O_4H_2O \xrightarrow{\triangle} CaC_2O_4 \xrightarrow{\triangle} CaCO_3 \xrightarrow{\triangle} CaO$$

TGA curve of $CuSO_4.5H_2O$ is given in below figure.



At about 100°C it loses four molecules of water and becomes pale blue monohydrate, $CuSO_4.H_2O$. At about 230°C it becomes white anhydrous $CuSO_4$. Anhydrous $CuSO_4$ decomposes at about 720°C to give Co and SO_2.

$CuSO_4.H_2O \rightarrow CuSO_4.H_2O \rightarrow CuSO_4 \rightarrow CuO + SO_2$

Blue Pale blue

Uses / Application

1. The TG curve is quantitative. So stiochiometric calculations can be done at any given temperature.
2. The correct drying temperature of precipitates in gravimetric analysis can be determined.

3. Thermogravimetry is used to find out suitable analytical standard substances. E.g., EDTA, NaF etc.
4. Thermobalance is used to determine the purity of various substances.

Information from the TGA– curves / Merits of TGA

The following information can be drawn from the curves:

1. The thermal stability of the material can be known (from the horizontal portions for TGA curve of $CuSO_4.5H_2O$).
2. The weight loss, when the sample is heated to a given temperature is known. This helps to determine the composition of the compound.
3. The thermodynamics, kinetics of various chemical reactions, reaction – mechanisms, and the intermediate and final reaction products can be studied from TG curve.

Factors affecting TGA curve

These are two factors which affect TG curves. They are

(i) Sample characteristics

(ii) Instrumental factors

(i) Sample characteristics

The amount and nature of the sample, solubility of evolved gases in the sample, particle size and heat of the reaction which the sample undergoes, the nature of packing of the sample, the thermal conductivity of the sample are some of the sample characteristics that affect the TG curve.

For example, when large amounts of the sample are taken, a deviation from linearity of weight change with the change in temperature takes place, particularly when the reaction is fast and exothermic. When small amounts of the sample are used, the presence of intermediate compounds can be detected.

When samples of different particle sizes are used, the rate of reaction and the shape of TG curve are altered.

A compressed sample decomposes at a higher temperature while loosely packed sample decomposes at a lower temperature.

If the decomposition is exothermic, the sample temperature is more than that of the furnace while in the case of an endothermic decomposition the furnace temperature is more than that of the sample temperature.

(ii) Instrumental factors

Some of the instrumental factors which affect TG curve are: heating rate of the furnace, speed of the recording, furnace atmosphere, geometries of

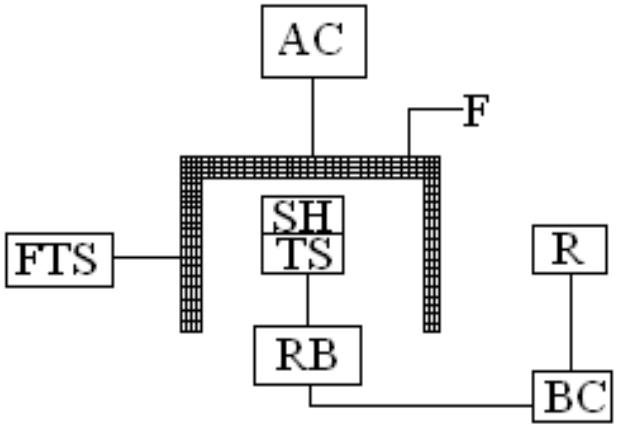
sample holder and furnace, sensitivities of the recorder and the thermobalance etc.

For example, when the rate of heating of the furnace is more, the decomposition temperature will be higher than that obtained when the rate of heating is less.

Furnace atmosphere also affects the TG curve. For example, if the decomposition of $CaCO_3$ is carried out in an atmosphere of CO_2, the decomposition takes place at a higher temperature than when it is conducted in an atmosphere of N_2.

Similarly, a shallow sample holder gives better results than a deep sample holder.

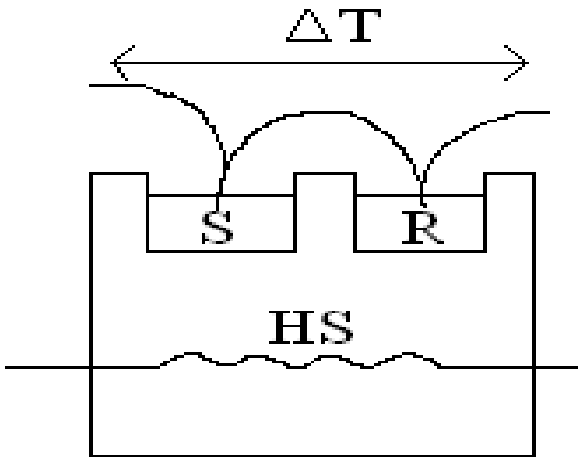
DTA Instrumentation (Block diagram)



DTA instrument consists of the following components:

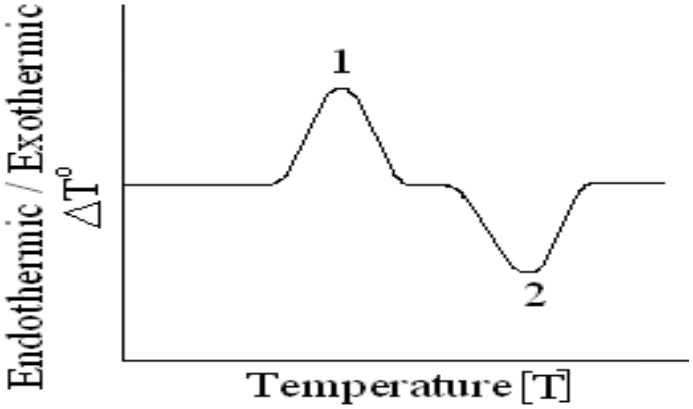
1. **A furnace (F)** to heat the sample.
2. **A holder (SH)** to hold the sample and the reference material.
3. **A furnace programmer and controller (FTS)** to increase the furnace and sample block temperature at a linear rate (5 – 12^{o}C per minute). This is done either by increasing the voltage through the heater element by a motor driven, variable transformer or by a thermocouple – actuated feed back type of controller.
4. **Detectors or Temperature sensors (TS):** These are thermocouple assemblies inserted separately in the sample and the reference material (below figure). These continuously measure the difference in temperature between the sample and the reference material. Microvolt level signals are got. These are sent to the amplifier.
5. **Recorder (R):** The signals got from the thermocouple are amplified by about 1000 times and the difference signal is recorded on the Y-

axis of mill volt recorder. The temperature of the furnace is measured by a separate thermocouple and is recorded on the X-axis.



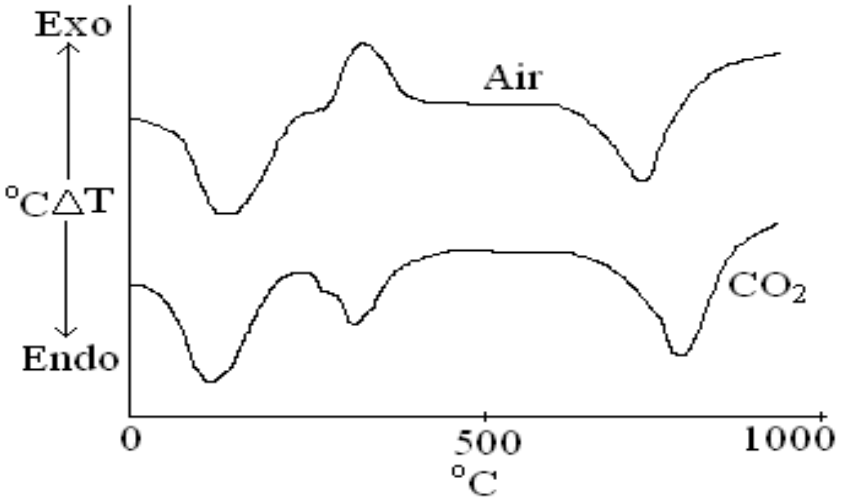
S – Sample; R – Reference; HS – Heat source

Characteristics of DTA curves



(1) Exothermic peak; (2) Endothermic peak

In DTA, a plot is made between ΔT and temperature or time. A typical DTA curve is shown in above figure. The shape and size of the peaks can give a large amount of information about the nature of the test sample. Sharp endothermic peaks signify changes in crystallinity or fusion processes. Board endothermic peaks indicate dehydration reaction. Physical changes give endothermic curves. Chemical reactions particularly, oxidation reaction, are exothermic. These can be illustrated by taking the DTA curve of calcium oxalate monohydrate. In two different atmospheres (air or CO_2), the DTA thermogram of $CaC_2O_4.H_2O$ is as shown in figure.



(i) In air

There are 3 – peaks: Two endothermic and one exothermic. The first endothermic peak indicates elimination of H_2O. The exothermic peak: This corresponds to the endothermic peak in CO_2 atmosphere. Here CO is eliminated and CO is oxidized to CO_2 which reaction is exothermic. The second endothermic peak corresponds to elimination of CO_2.

(ii) In CO_2

There are three peaks, all endothermic. These correspond to the successive elimination of H_2O, CO and CO_2. These elimination processes are endothermic as they require energy to break the bonds.

The DTA thermograms of $(CH_3COO)_2$ Ca. H_2O in three different atmospheres (O_2, CO and Ar) are shown in below figure.

(a) The first endothermic peak is unaffected by the change in atmosphere. The weight loss indicates formation of anhydrous salt.

(b) In Ar and CO_2 second endothermic peak is got; but exothermic in O_2. Here one mole of CO and CO_2 are lost. In O_2 atmosphere CO is oxidized to CO_2 which is exothermic.

(c) Finally $CaCO_3$ decomposes in to CaO and CO_2. This process is a function of partial pressure of CO_2. So the peak is shifted to higher temperature in CO_2 atmosphere.

Uses / Applications

1. DTA is very useful in the identification of dyes. Melting point can be easily determined.
2. DTA – technique is used for the quality control of a large number of substances like cement, glass, textiles, explosives etc.

3. DTA – technique is used to study the thermal stability of inorganic compounds and complexes.
4. It is used to ascertain the heating range of precipitates in which the precipitates remain stable. These data are used in gravimetric analysis.

Factors affecting DTA curves

These are two factors which affect TG curves. They are

(iii) Sample characteristics

(iv) Instrumental factors

(i) Sample characteristics

Particle size, thermal conductivity, heat capacity, packing density, amount, swelling ans shrinkage of the sample is some of the sample characteristics that affect DTA curves.

(ii) Instrumental factors

Furnace atmosphere, size and shape of the furnace, the material with the sample holder is made, the geometry of the sample holder, heating rate, speed and response of the recording device, the location of the thermocole in the sample chamber are some of the instrumental factors that affect the DTA curves.

THERMOMETRIC TITRATIONS

It is a titration in which the change in temperature of the reaction mixture is observed and plotted against the volume of the titrant added and thereby the analyte is estimated.

One of the methods to estimate an analyte is to titrate it against a standard or standardized solution and plot the change in temperature (ΔT) against the volume of the titrant added. A sharp break in the curve gives the end point.

Example

Titration of HCl against NaOH.

Principle

All chemical reactions are accompanied by heat changes. These will be accompanied by temperature changes. So the course of a titration may be followed by measuring the changes in T i.e., ΔT. At the equivalence point the temperature change will be maximum.

Example

Titration of HCl against NaOH.

Need for thermometric titrations

Other methods of titrations are based on a decrease in free energy, i.e., negative ΔG and hence on equilibrium constant, K. But there are reactions in which ΔG is very low, or even zero or in some cases positive. E.g., For the neutralization reaction of boric acid, $\Delta G = -6$. For such reactions ordinary titrations which depend on pH are not useful. For such reactions thermometric titrations are useful. Here ΔH is measured in terms of ΔT.

Conditions of thermometric titrations

1. The heat change accompanying the reaction to be studied by thermometric titrations should be sufficient enough to be measured.
2. The heat of dilution of the product must be negligible.
3. The reaction must be simple, one step reaction.
4. Heat exchange between the titration vessel and its surrounding must be avoided. i.e., The titration should be conducted under adiabatic conditions.

Apparatus

The apparatus for thermometric titration contains

(i) Motor driven burette

(ii) An adiabatic titration chamber, a small Dewer flask

(iii) Thermistor assembly

(iv) A recorder.

Method / Procedure

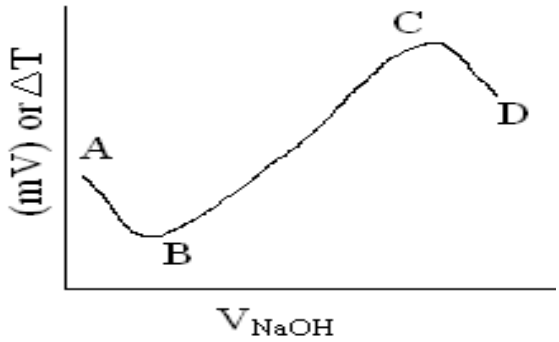
The titrant is added from a burette in to a thermally insulated vessel containing the analyte. The temperature change after every addition is recorder. The measured ΔT is plotted against the volume of the titrant added. A sharp break in the curve gives the end point.

Example

Titration of HCl against NaOH.

A known volume of HCl to be estimated is taken in an adiabatic chamber (Dewer flask). NaOH is added from a burette. The temperature change after each addition is recorded. The measured ΔT is plotted against the volume of NaOH added. A sharp break in the curve gives the end point.

The thermometric titration curve between HCl and NaOH is as shown below:



A – Before NaOH is added; ABC – NaOH is being added; C – The titration is complete (end point)

Applications

(i) Determination of the concentration of an unknown substance.

(ii) Determination of thermodynamic quantities like ΔH etc.

Important Questions

1. Discuss the principles and applications of polarography
2. Write notes on DME
3. What are the advantages and disadvantages of DME
4. Explain limiting and diffusion current
5. Explain migration and residual current
6. What is the effect of temperature and agitation on diffusion.
7. Explain half wave potential
8. Write an essay on theory and applications of amperometric titrations
9. Discuss the various components of TGA
10. Explain the TGA-DTA curves of $SrCO_3$
11. What are the factors affecting TGA and DTA curves
12. What are the principles and applications of thermometric titrations

NOTES

UNIT – IV

INFRARED AND RAMAN SPECTROSCOPY

4.1. INFRA RED SPECTROSCOPY

Infrared spectroscopy is based on the absorption of infrared radiations by molecules. The region of light having wave lengths between 35000 ^{o}A and 80000 ^{o}A is called the infrared region. These radiations can be measured with the help of thermopiles which register the heat generated by them. The optical parts are made of large crystals of NaCl or CaF_2.

Basic principles or theory of IR spectroscopy

Absorption in the IR region is due to the changes in the vibrational and rotational levels of molecules. When radiations with frequency range less than 100 cm-1 are absorbed, molecular rotation takes place in the substance. As this absorption is quantized discrete lines are formed in the spectrum due to molecular rotation.

The molecules interact with the absorbed radiation and bending, rotation and vibrational motions of atoms in the molecule occur simultaneously. These motions produce a highly complex absorption spectrum. This spectrum is uniquely characteristic of the functional groups in the molecule and also its overall configuration.

A molecule absorbs a radiation when

(i) The energy of the radiation corresponds to the energy difference between two electronic, vibrational or rotational levels in a molecule

(ii) There is strong coupling interaction between the radiation and the molecule which absorbs that radiation.

This coupling interaction takes place only if there is a change in dipole moment in the molecule during the adsorption process. If there is no change in dipole moment during vibration or rotation of a molecule, then there will be no coupling interaction between the radiation and the molecule which absorbs the radiation. So no absorption takes place even though the condition (i) is satisfied.

Band positions in IR spectra are presented either as wave number or wave lengths. The wave number unit (cm^{-1}) is used most often. cm^{-1} is directly proportional to the energy of the vibration and most modern instruments are linear in the cm^{-1} scale. Wave numbers are reciprocally related to wave length as follows:

$$Cm^{-1} = 1 / \mu m \text{ X } 10^4$$

Notice that the wave numbers ($\tilde{\upsilon}$) are often called “Frequencies”, it is not rigorously correct, since wave numbers are $1/\lambda$ and frequency is c/λ. Band

intensities are expressed either as transmittance (T) or absorbance (A). Transmittance is the radiant power of incident on the sample.

Thus, the condition for a molecule to be IR active is that, it should undergo a change in dipole moment during rotation or vibration. Thus molecules with permanent dipoles in them and which undergo a change in their dipole moments during rotation and vibration only will be IR active. Thus, homo nuclear diatomic molecules like H_2, N_2, Cl_2 are IR inactive, while hetero nuclear diatomic molecules like HD, HCl, NO etc., are IR active, i.e., they give IR spectrum.

In the case of tri atomic molecules, which have a permanent dipole moment like H_2O and NH_3 are IR active. In the case of CO_2 the molecule is having no dipole moment. But it undergoes some anti symmetric stretching and bending vibrations during which the molecule undergoes some change in dipole moment. So CO_2 is IR active.

Instrumentation

The essential part of an IR spectrometer is

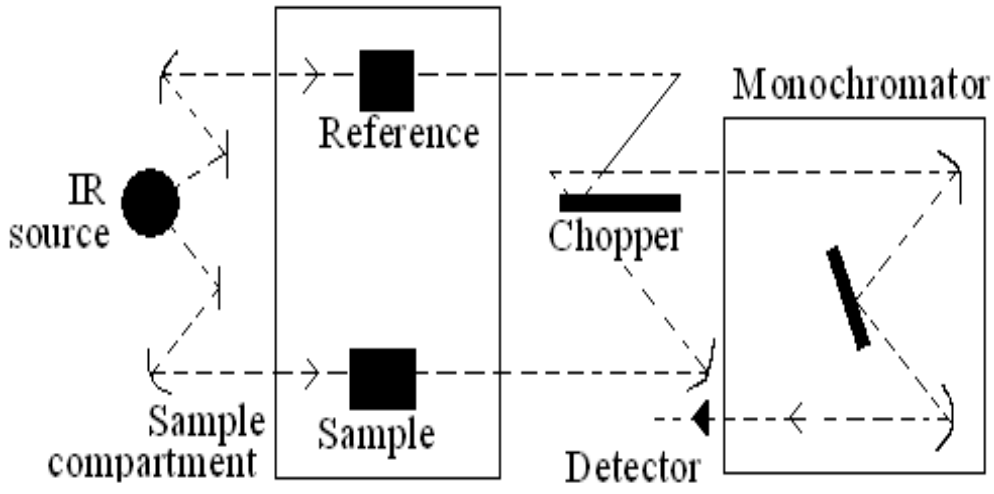
(i) The IR radiation source

(ii) The monochromators

(iii) The sample cells

(iv) Detectors

Block diagram



Mode of operation

Light from source S is split in to two beams. One beam passes through the sample (sample beam SB). The other is the reference beam. The different in intensities between the two beams at each wave length is measured in such a

double beam operation. The two beams are reflected to a chopper C. This reflects alternately the sample beam and the reference beam to the monochromator grating MG. From here individual frequencies reach the detector D. Here the infrared (thermal) energy is converted in to electrical energy.

When the sample has absorbed light of a particular frequency, the detector receives from the chopper, alternately, an intense reference beam and a weak sample beam. This leads to a pulsating or alternating current flowing from the detector to the amplifier A.

The amplifier, receiving the output of balance signal is coupled to a motor M. This drives an optical wedge W in to the reference beam until the detector receives light of equal intensity from sample and reference beams. This movement of the wedge (or attenuator) is coupled to a pen recorder R. We get absorption bands on the printed spectrum.

Now we shall see the descriptions of the individual components.

Source

The various IR radiation sources are

(a) Incandescent lamp

(b) Nernst glower (It is made up of rare earth oxides such as zirconia, yttria and thoria. It emits IR radiations over a wide wave length range).

(c) Globar source (It is made up of sintered silica carbide)

(d) Various ceramic materials

Monochromator

Both prisms and gratings are used. The common prism material is NaCl. NaCl is only transparent down to 625 cm-1. CsI or a mixture of ThBr and ThI is used for lower frequency works. Some other alkali metal halides like CsCl, KBr also used as prism material. Gratings give better resolution at higher frequency than do prisms. NaCl is hygroscopic (disadvantage). Therefore it must be protected from condensation of moisture. This done by maintaining the temperature at about 20^{o}C above the ambient temperature.

Glass or quartz can not be used as prism material since they absorb strongly though most of the IR region.

Cell

A wide range of cells is available for mounting the sample in the beam of the infrared spectrometer, depending upon whether the sample is a gas, a liquid or a solid. If the sample is a gas, a gas cell is used. Nowadays it has

infrared transparent glass windows made of NaCl. Multi pass gas cells are used for better sensitivity. They contain mirrors mounted inside, which permit the light beam to be reflected several times through the sample.

Rock salt (NaCl) flats are used for liquids. A solution cell or a variable path – length cell is used for solutions. It consists of transparent windows with a lead or poly tetra fluoroethylene spacer between them of known thickness. The thickness of the spacer determines the path length of the cell which is usually 0.1 to 1.0 mm.

Sampling techniques

Several techniques are adopted to mount the sample in the beam of the infrared spectrometer. These techniques depend on whether the sample is a gas, liquid or a solid. Intermolecular forces vary considerably in passing from solid to liquid to gas. The effect of these differences will be displayed in the form of frequency shifts, additional bands etc. Therefore it is necessary to record the sampling technique used, on a spectrum.

Gases

The gas sample is introduced in to a gas cell. The cell is directly mounted in the path of the sample beam.

Liquids

The liquid is made as a thin film between two infrared transparent windows. E.g., NaCl flats. The assembled pair of flats and liquid film is mounted in the sample beam. Liquid samples can also be examined in solution.

Solids

There are three common techniques to record spectra of solids.

1. KBr discs
2. Mulls
3. Deposited films

Solids can also be examined in solution. But solution spectra may have different appearances from solid spectra since intermolecular forces will be altered.

KBr discs

KBr discs are prepared by grinding the sample with KBr in the ratio 1:50 by weight. The whole mixture is compressed using a special die to a transparent water or disc having 13 mm diameter and 0.3 mm thickness.

Mulls or Pastes

Mulls or paste are prepared by grinding the sample with a drop of oil. The mull is then squeezed between transparent windows as for liquid samples.

Usually liquid paraffin (Nujol) is used as the mulling agent. The spectrum produced always shows the absorptions of the mulling agent superimposed on that of the sample. Nujol shows absorptions due to C-H stretching at 3030 – 2860 cm^{-1} and at 1460 - 1374 cm^{-1} for C-H bending.

Solid films

Solid films are deposited on to NaCl flats by allowing a solution in volatile solvents to evaporate drop by drop on the surface of the flat.

4. Solutions

The sample can be dissolved in a solvent (e.g., CCl_4, CS_2 or $CHCl_3$) and the spectrum of this solution is recorded. The solution (usually 1 to 5%) is placed in a solution cell. A second cell containing pure solvent is placed in the reference beam, so that solvent absorptions are cancelled out and the spectrum recorded is that of the solute alone.

Although, in principle, solvent cancellation is possible throughout the entire range, in those regions where the solvent too has strong absorption bands, little light passes to the detector. In such cases two spectra are run using solvents whose transparent regions are complementary.

Solution spectra give more valid results as complicated features introduced by intermolecular forces and polymorphism are absorbent in them. However the solvent must be specified on the spectrum since band frequencies change with the polarity of the solvent used.

Detectors

In IR spectroscopy thermal detectors like thermocouple, Bolometer etc., are used.

Thermocouple

It is the most widely used IR detector. It work on the principle that if two dissimilar metal wires are joined head to tail then a difference in temperature between head and tail causes a current to flow in the wires. **The current will be proportional to the intensity of radiation falling on the thermo couple.**

Two dissimilar metals, e.g., silver and platinum are placed together and the radiation to be measured is focused on a receiver plate connected to one of the junction. The receiver plate absorbs the incident radiation and converts it in to heat. The temperature difference between the hot and cold junctions sets up a small voltage difference between these two junctions. The resulting current is directly proportional to the intensity of the incident radiation. It is measured.

Advantages

1. A thermopile which is a highly sensitive device
2. It consists of several thermocouple junctions joined to a common receiver plate is used in modern IR spectrometers.

Bolometers

A bolometer is constructed from a metal of a semiconductor that exhibits a large change in electrical resistance as a function of temperature. When radiation falls on the bolometer, its temperature changes and hence its resistance also changes. The degree of change in resistance is a measure of the amount of radiation falling on the bolometer.

Recorders

The electrical energy got from the detectors is amplified with the help of an amplifier. The amplifier is coupled to a small motor which drives an optical wedge. The movement of the wedge is in turn coupled to a pen recorder which draws absorption bands on the calibrated chart. The movement of the wedge continues till the detector receives light of equal intensity from the sample and the reference beam. Calibration of the chart can be carried out using spectrum of poly styrene.

Types of stretching and bending vibrations

There are two types of molecular vibrations or fundamental vibrations.

(i) Stretching vibration

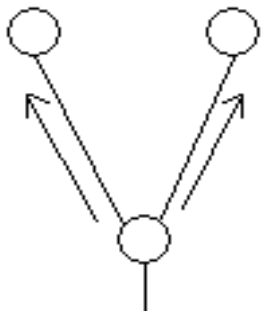
A stretching vibration is a rhythmical movement along the bond axis such that the inter atomic distance is increasing or decreasing.

Types of stretching vibrations

There are two types of stretching vibrations.

Symmetric stretching

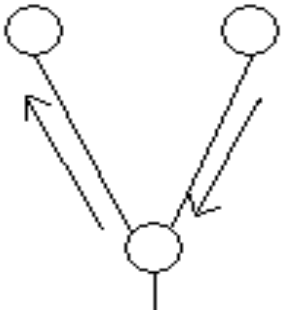
In this type, the movement of the atoms, with respect to a particular atom in a molecule is in the same direction.



Symmetric

Asymmetric stretching

In these vibrations, one atom approaches the central atom while the other departs from it.



Bending vibration

A bending vibration may consist of a change in bond angle between bonds with a common atom or the movement of a group of atoms with respect to the remainder of the molecule without movement of the atoms in the group with respect to one another.

Types of bending vibrations

Bending vibrations are of four types.

Scissoring

In this type two atoms approach each other.

Rocking

In this type, the movement of the atoms takes place in the same direction.

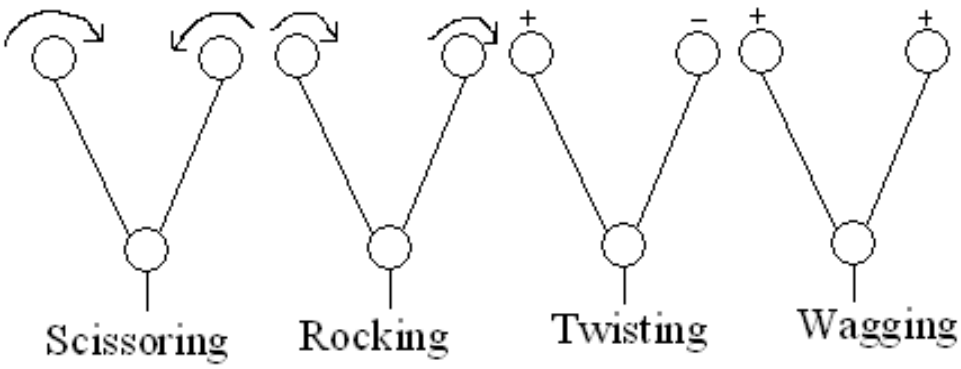
Wagging

Two atoms move up and below the plane with respect to the central atom.

Twisting

In this type, one of the atoms moves up the plane while the other moves down the plane with respect to the central atom.

Bending or deformation modes



Vibrational frequencies

We know that molecule vibrate. For this they need energy. They get the necessary energy by absorbing light of suitable energy in the IR region. The position of the absorbed light in the spectrum can be specified in units of frequency. Such frequencies are called vibrational frequency.

At ordinary temperature, organic molecules are constantly vibrating. Each bond absorbs light of specific frequency and performs stretching and bending vibrations. Thus each bond has its characteristic stretching and bending frequency. The stretching energy of a bond is greater than the bending energy. Thus stretching absorptions of a bond appear at higher frequencies in the IR spectrum than the bending absorptions of the same bond.

We can calculate the vibrational frequency of a bond using Hooke's law

$$\upsilon = \frac{1}{2\pi}\left[\frac{k}{(m_1 m_2 / m_1 + m_2)}\right]^{1/2}$$

Where ύ = frequency; k = force constant of the bond; m_1, m_2 = the masses of the two atoms connected by the bond.

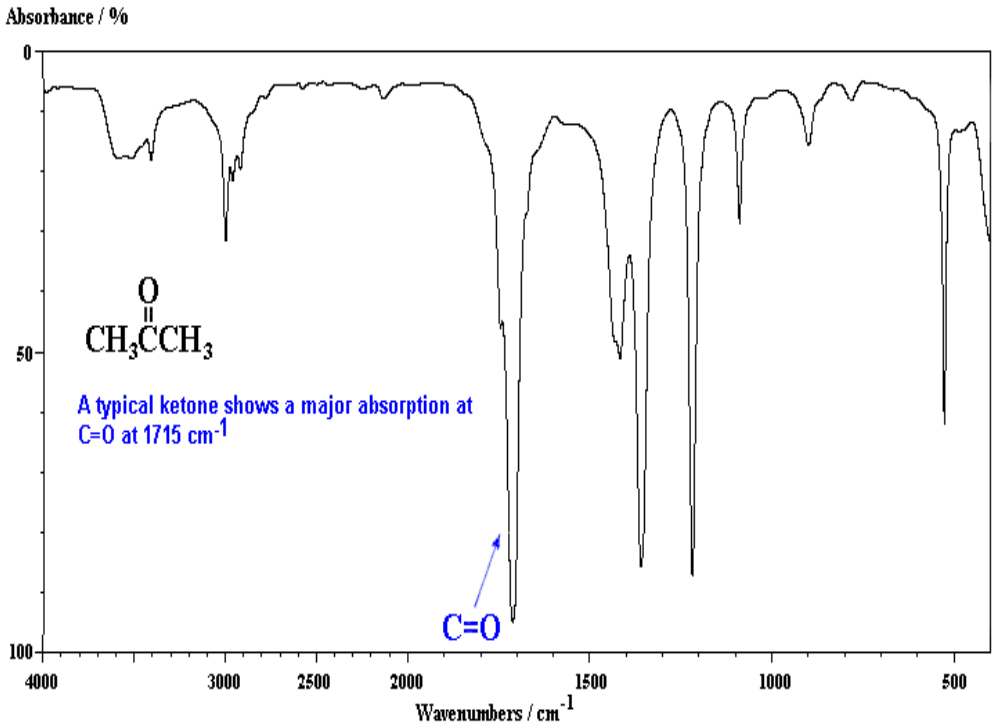
The quantity $m_1 m_2 / (m_1 + m_2)$ is expressed as μ the reduced mass of the system.

Interpretation of IR spectra

Acetone (2-propanone)

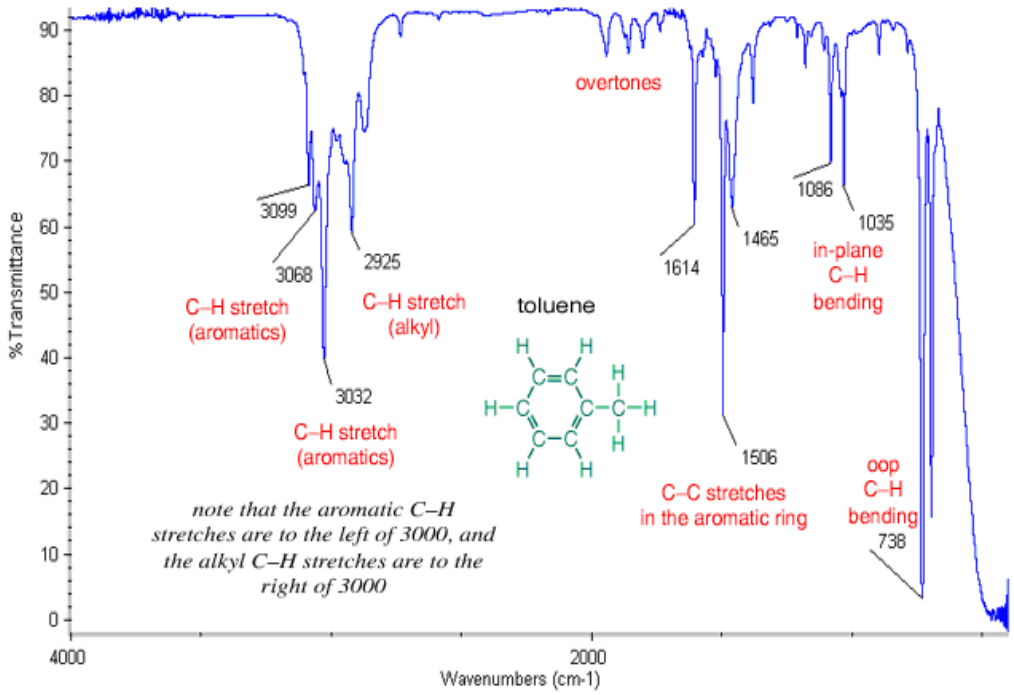
Acetone (2-propanone) is the "classic" carbonyl containing compound with the obvious C=O stretch in the middle of the spectra. Note that the peak is a very strong absorption. Compare it with the C=C in the previous case which are weaker and sharper.

- υC = O str ~ 1715 cm^{-1}
- υC – H str 2960 – 2850 cm^{-1}

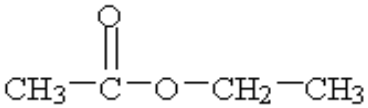


Toluene

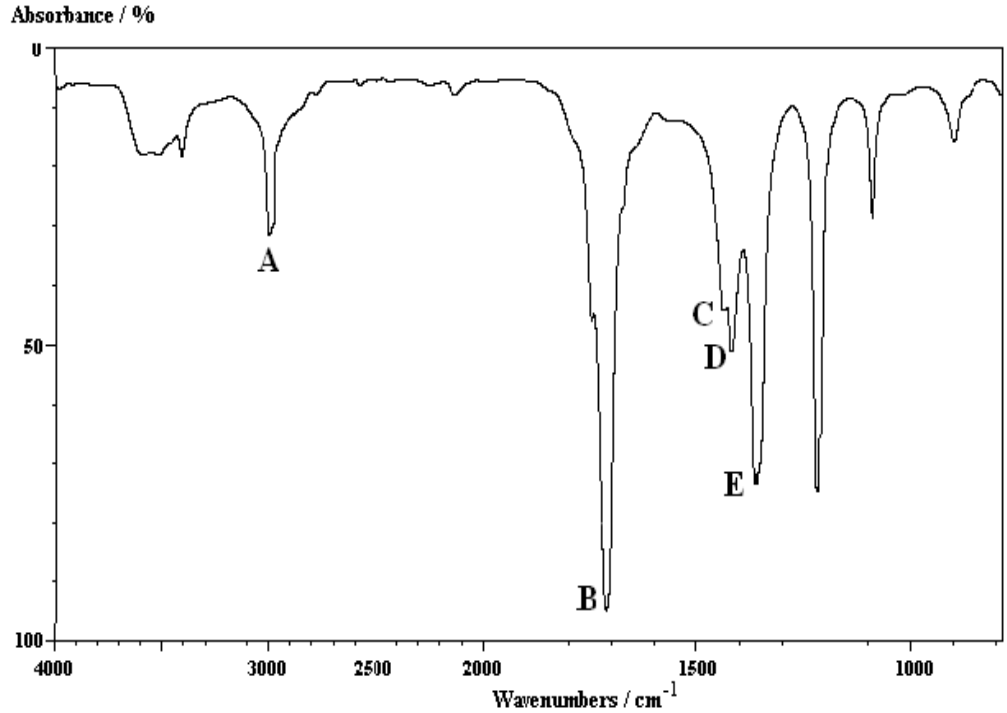
The spectrum of toluene is shown below. Note the =C–H stretches of aromatics (3099, 3068, 3032) and the –C–H stretches of the alkyl (methyl) group (2925 is the only one marked). The characteristic overtones are seen from about 2000-1665. Also note the carbon-carbon stretches in the aromatic ring (1614, 1506, 1465), the in-plane C–H bending (1086, 1035), and the C–H out of plane (738).



Ethyl acetate



Principal characteristic peak is υ C=O at 1710 cm^{-1}; also υ C-O at ~ 1240 cm^{-1}



Positions of some characteristics absorptions

A = 3002 cm^{-1} C – H str in methyl / methylene

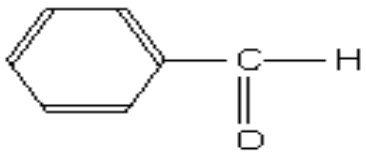
B = 1742 cm^{-1} C = O str (in esters)

C = 1450 cm^{-1} C – H def

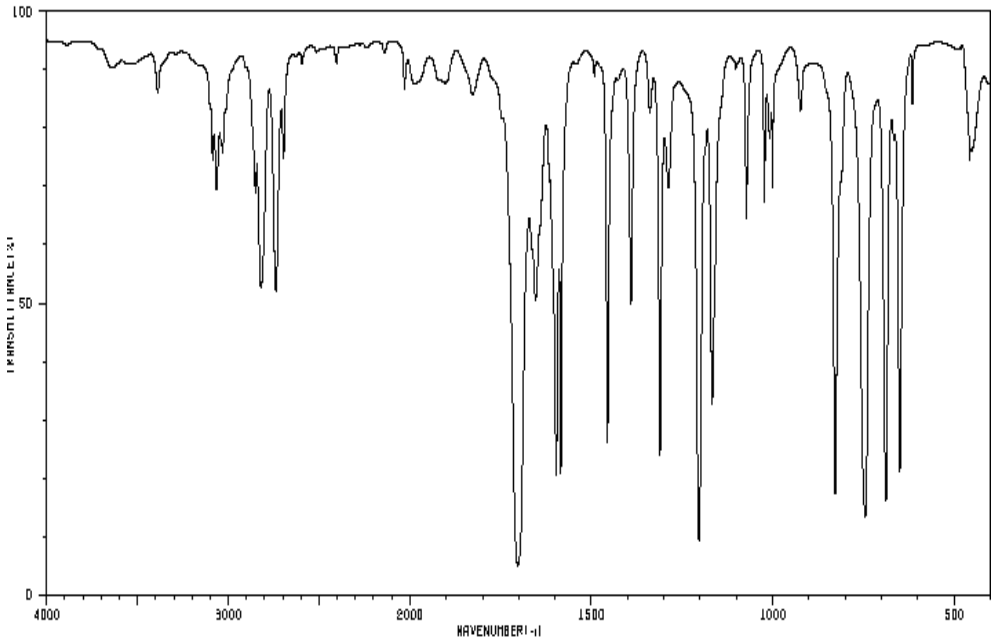
D = 1370 cm^{-1} C – H def

E = 1240 cm^{-1} C – O str in esters

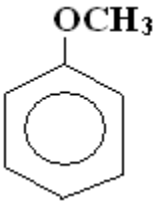
Benzaldehyde

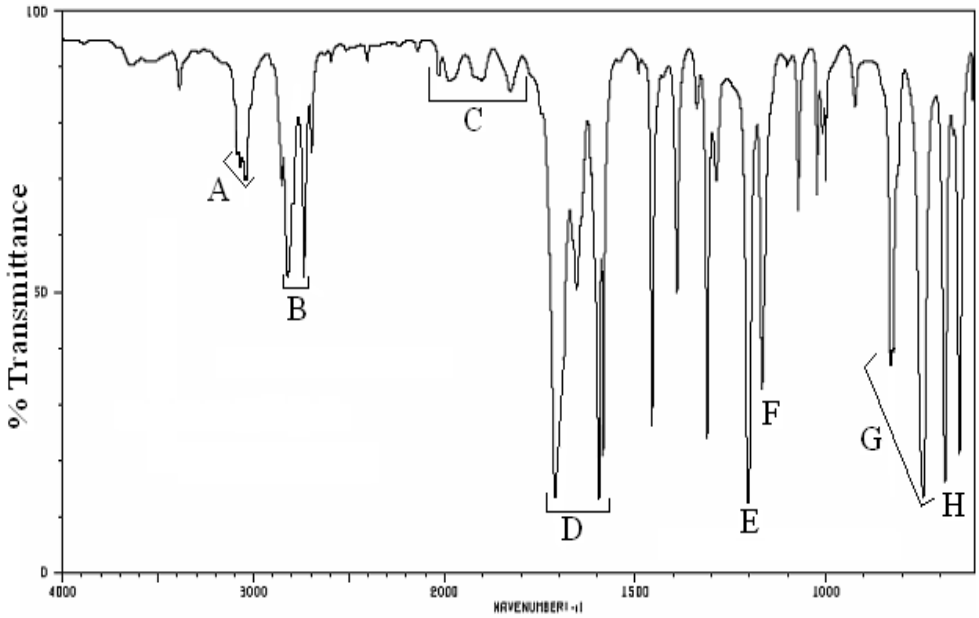


Infrared spectrum: Liquid film



Anisole





A = Aromatic C – H str 3060, 3030, 3000 cm^{-1}

B = Methyl C – H str 2950, 2835 cm^{-1}

C = Overtone – combination region, 2000 – 1650 cm^{-1}

D = 1600, 1498 cm^{-1}

E = Asymmetric C – O – C str 1247 cm^{-1}

F = Symmetric C – O – C str 1040 cm^{-1}

G = Out of plane C – H bend, 784, 754 cm^{-1}

H = Out of plane ring C = C bend, 692 cm^{-1}

4.2. RAMAN SPECTROSCOPY

When a beam of strong light is passed through a substance (solid, liquid or gas) and when the scattered light is observed in a direction perpendicular to the direction of the incident beam, the scattered light has a slightly different frequency than that of the incident light. This effect is called Raman Effect.

If the incident light is monochromatic, the scattered spectrum exhibits a number of lines displaced from the original frequency. This spectrum is called Raman spectrum.

If $\acute{\upsilon}_i$ is the frequency of the incident beam and $\acute{\upsilon}_s$ is that of the scattered light then the Raman frequency $\acute{\upsilon}_R$ is given by, Ramen Effect occurs because of

$$\acute{\upsilon}_R = \acute{\upsilon}_i - \acute{\upsilon}_s$$

and $\Delta E = h\acute{\upsilon}_R$

(i) The dipole moment induced in the molecule by the incident light. Therefore, it depends upon the polarizability of the molecule.

(ii) The rotational and vibrational oscillations of molecule of a compound.

If a vibration or rotation of a molecule is to be active in Raman its polarizability, must change during the vibration or rotation. This is the basic requirement or condition.

Rayleigh and Raman scattering (Origin of Raman spectra)

Let us assume a quantum of $h\acute{\upsilon}_i$ of incident light strikes a molecule. If it is scattered elastically, i.e., without energy change, then the frequency of the incident light and that of the scattered light are the same. Such a scattering is called Rayleigh scattering or Tyndall effect.

During Rayleigh scattering atoms and molecules are polarised in the electric field of the light. During this polarization if the distances between atoms in the molecules do not change then the frequencies of incident and scattered radiations will be same. This is what happens in Raleigh scattering. Sometimes the inter atomic distances in molecules may vary. Further the molecules are not spherically symmetrical. Thus a condition of anisotropy or polarisability takes place. The consequent vibrational frequencies of the

scattering molecules are superimposed on the fundamental frequency of incident radiation. Thus the light is scattered inelasticially, i.e., the frequency of the incident light will not be equal to the frequency of the scattered light. Such a scattering is called Raman Scattering.

Selection rules for Raman spectra

1. Raman spectra are exhibited when there is a change in the polarizability of molecule during rotation or vibration.
2. $\Delta J \pm 0$ or ± 2 only.
3. $\Delta V \pm 0 \pm 1 \pm 2$….(But we can ignore ± 2… since the probability of $\Delta V = \pm 2$…. Decreases rapidly and the bands are of low intensity and are weak).

The scattering of light by molecules can be explained by making use of electro – magnetic theory. The incident light wave represented by an electric field E induces a dipole moment in the molecules. The induced moment P is given by

$$P = \alpha E$$

Where α is the polarisability of the molecule. Raman Effect depends upon polarisability.

Stokes and antistokes lines

The difference between the frequency of the incident light and the frequency of the scattered light is called Raman frequency.

That is $\acute{\upsilon}_R = \acute{\upsilon}_i - \acute{\upsilon}_s$

The incident radiation may give up energy to the molecule on which it falls. Then the frequency of the incident light will be more than that of the scattered light. i.e., $\acute{\upsilon}_i > \acute{\upsilon}_s$. Then the Raman frequency $\acute{\upsilon}_R$ is given by

$$\acute{\upsilon}_R = \acute{\upsilon}_i - \acute{\upsilon}_s = \text{Positive}$$

We get lines on the lower frequency side of the incident light. These are called stokes lines.

On the other hand the incident light can also be taken up energy from the molecule. Then the frequency of the incident light will be less than that of the scattered light. i.e., $\acute{\upsilon}_i < \acute{\upsilon}_s$. Then the Raman frequency $\acute{\upsilon}_R$ is given by

$\acute{\upsilon}_R = \acute{\upsilon}_i - \acute{\upsilon}_s$ = Negative

We get lines on the higher frequency side. These are called anti stokes lines. These stokes and anti – stokes lines are generally called Raman lines.

Characteristic properties of Raman lines

1. The intensities of Raman lines are in the following decreasing order. Stokes lines > incident radiations > anti – stokes lines. This is because

the stokes lines result from the absorption of energy of the incident light by the molecules. In this case the molecular energy increases. Therefore stokes lines are more intense than the incident light.

On the other hand the anti – stokes lines result from the emission of energy to the incident light by the molecules. In this case, the molecular energy decreases. Therefore, anti – stokes lines are less intense than incident light. Totally stokes lines are more intense than the incident light.

2. The intensity of anti – stokes lines increases with an increase in temperature.
3. The intensity of Raman line changes with the frequency of incident radiation and has been found to be proportional to the fourth power of frequency. Hence a light of as short a wave length as possible should be used in order to get as intense a Raman line as possible.
4. Raman shift, $\Delta\acute{\upsilon}$, generally lies within the range of 100 – 3000 cm^{-1} which in turn, lies in the far and near IR region.
5. **Polarisation of Raman lines and depolarization factor:**

After scattering by any medium light is found to be polarised, i.e., the intensity of the ray with vibrations parallel to the direction of incident light I_h is not the same as the intensity of the ray with vibrations perpendicular to the incident light I_v.

Such a polarization of Raman line may be regarded as due to the differences in the symmetry of molecular vibration and each Raman line can have its own state of polarization independent of other simultaneously emitted lines. The state of polarization of a Raman line is measured by a quantity known as **depolarization factor P**.

$$P = \frac{I_h}{I_v} = \frac{\text{Intensity of horizontal light vibrations}}{\text{Intensity of vertical light vibrations}}$$

Important points about depolarization factor

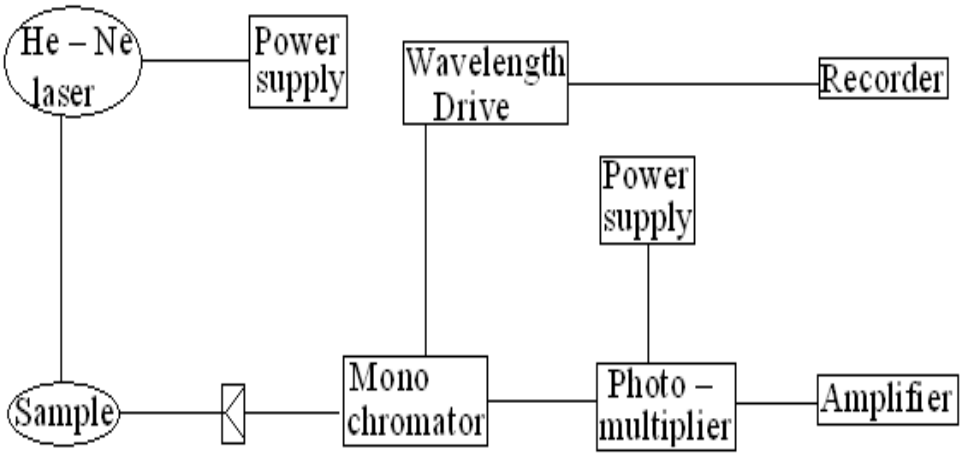
1. It varies from zero to 0.86 for the vibrational Raman lines while it has constant value of 0.86 for rotational lines.
2. Low depolarization factors result in sharp and strong lines. High depolarization factors result in diffuse and weak lines.
3. Molecules having similar structures give Raman lines which have the same depolarization factors.

Differences between Stokes and anti – stokes lines

Property	**Stokes lines**	**Anti – stokes lines**
Sign of Raman frequency $\acute{\upsilon}_R = \acute{\upsilon}_i = \acute{\upsilon}_s$	Positive	Negative
Intensity	More – intense	Less intense

Instrumentation

Block Diagram



Component description

Source (S)

Low pressure mercury arc (436 nm) was used as the source of radiation in the beginning. Now – a – days LASERS (Light Amplification by Stimulated Emission of Radiation) are used as the source of radiation.

Example

He – Ne laser (633 nm)

Filters (F) (Monochromators)

If mercury arc source is used, a saturated aqueous solution of sodium nitrate is used to absorb light below 436 nm. A rhodamine dye solution is used to absorb light above 436 nm. A double pass Littrow – mounted grating type monochromator is used for lasers.

Sample optics

The sample optics must be arranged so that as much scattered light as possible reaches the spectrometer. Commercial Raman spectrometer involves 90^o method, the scattered radiation is observed at 90^o to the incident light. It has been shown to be a versatile method and is applicable to gas, liquid and solid samples.

Monochromators

The monochromator must have excellent stray light characteristics because of the very weak nature of the Raman effect. Laser Raman spectrometer generally incorporates two or more grating monochromators. The stray light due to the exciting radiation can be drastically reduced using double monochromators. The effects of scattered light due to dust particles and precipitates can also be avoided using double or triple monochromators. The removal of dust particles from the sample solution is necessary since it would otherwise increase the background radiation near the exciting line. The use of double or triple monochromators increases the resolving power of the spectrometer and reduces the background caused by Raleigh scattering by the sample.

Detectors

Photographic plates were used previously to detect and record the spectrum. Long expression times were usually needed. Now – a – days high sensitivity photomultiplier is used as the detector in laser Raman spectroscopy. In a photomultiplier, the protons incident on the photocathode cause the emission of electrons, the number emitted being proportional to that of the protons. The spectrum is recorded on a chart paper.

6. Calibration

For outline calibration CCl_4 and Indene used. The usual method of calibrating the Raman spectrometer is to be record the emission line from the laser source that is being used, the standard frequencies for the emission lines of Ne, Ar and Kr are available.

Difference between Raman and IR spectra

S. No	Raman spectra	IR spectra
1.	It appears due to the scattering of radiation by the vibrating molecules.	It arises because of the absorption of radiation by the vibrating molecules.
2.	For Raman spectrum to be observed, there must be a change in the polarisability of the molecule.	For IR spectrum to occur, there must be a change in dipole moment of the molecule.

3.	Water can be used as a solvent.	Water is opaque to IR, and can not be used because cell materials will dissolve.
4.	Optical systems are made of glass or quartz.	Optical systems are made of alkali metal halides.
5.	Concentrated solutions are used to increase the intensity of Raman lines.	Dilute solutions are generally preferred.
6.	Substance under analysis must be pure and colourless.	Impure and coloured samples may be taken for inverting atom.
7.	The method is very accurate and relatively less sensitive.	The method is accurate as well as sensitive.
8.	Homopolar molecules respond to Raman scattering.	Homopolar molecule can not exhibit vibration spectra.
9.	Sometimes photo chemical reactions occur in the region of Raman lines.	Photo chemical reactions do not take place.
10.	In the Raman effect, double transitions involve three stationary levels. The third energy level may be above or below the normal ground level.	It involves only one transitions, the direct transition between the two energy levels.

Mutual exclusion principle

According to the law of mutual exclusion which states that, for molecules with a centre of symmetry, transitions that are allowed in the IR are forbidden in Raman and vice versa. This law also implies that transition may occur both in Raman and IR if the molecule has no centre of symmetry.

Example

CO_2 has one strong band in its Raman spectrum at 1389 cm^{-1} and two strong bands in its IR absorption spectrum at 668 and 2349 cm^{-1}. None of these occur both in IR and Raman spectra. This is because CO_2 has a centre of symmetry.

APPLICATIONS

Structural diagnosis

1. Molecular structure

(a) Raman spectra have been widely used in deciding the constitution of organic compounds since Raman frequencies for single, double and triple bonds are available. Hence these can be easily located.

(b) Similarly linear structures and non – linear structures can be identified.

2. Using Raman spectra we can study the strength and nature of forces which are present in the crystals.

3. Centres of vibrational bands which do not appear in IR spectra are produced in Raman spectra. Thus force constants, and moments of inertia can be calculated from Raman spectra.

4. In inorganic chemistry we can determine the chemical constitution and the nature of the valence bands. Isomers show different Raman lines. It is used in the study of complex compounds, mixed molecules and water of crystallization.

5. In organic chemistry aliphatic and aromatic compounds have different Raman spectra. Structure of organic compounds and isomerism can be studied with Raman spectra.

6. In physical chemistry amorphous states and crystalline states, electrolytic dissociation, hydrolysis etc., may be studied.

SPECTROPHOTOMETRIC AND COLORIMETRIC ANALYSIS

4.3. UV – VISIBLE SPECTROSCOPY

Introduction

Spectroscopy is the branch of science which deals with the study of interaction of electromagnetic radiation with matter. It is powerful technique for the study of atomic and molecular structure.

Electromagnetic radiation is a type of energy that is transmitted through space at enormous velocities.

When a beam of monochromatic light passes through a transparent medium, part of the light is absorbed and the transmitted beam has a lower intensity than the intensity of the incident beam.

Solutions are placed in containers (called "cuvettes") whose material is transparent: quartz is commonly used, but for visible light, one also uses disposable cuvettes made of polystyrene or polycarbonate.

The **transmittance, T** of the solution is defined as the ratio of the intensities of the transmitted beam, **I** to the intensity, $\mathbf{I_o}$ of the incident beam:

$\mathbf{T = I/I_o}$

The **absorbance, A** of a solution is defined as

$\mathbf{A = -\log_{10}T}$. Since **A** is a logarithmic function, it is dimensionless.

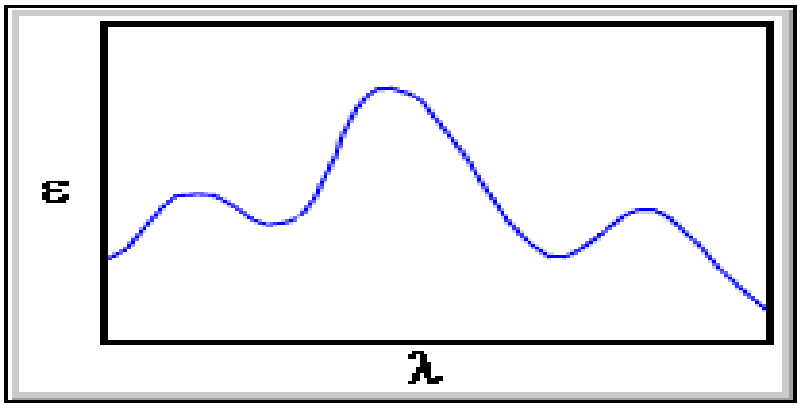
Both of the above quantities are readily measured using an instrument called a **spectrophotometer**, which delivers monochromatic light over the ultraviolet and visible ranges of the electromagnetic spectrum, and have suitable light detection and recording devices.

The Beer-Lambert law:

The Beer-Lambert law (also known as Beer's law) (as it applies to solutions of light-absorbing substances) states that the absorbance is directly proportional to the path length, **l** of the sample and its concentration, **c**:

$$\mathbf{A = \varepsilon cl}$$

where **ε** is the **molar extinction coefficient**(with dimensions of $dm^3.mol^{-1}cm^{-1}$) of the solute, **c** is the molar concentration (in $moles.dm^{-3}$), and **l**, the path length, is measured in centimeters.



The molar extinction coefficient is constant for a particular solute, and varies with the wavelength of the light. A plot of **e** against the wavelength, **l**, is called an **absorption spectrum**. Such spectra, an example of which is shown on the left, may be in the ultraviolet or visible ranges of the electromagnetic spectrum.

Instrumentation

When molecules absorb radiation in UV and visible region (200-800 nm) changes in electronic configuration and energy of molecules occur. In UV – visible spectroscopy the amount of adsorption of radiation is measured by determining the reduction in power suffered by a beam of radiation as a consequence of passing through the absorbing medium. The wave length at

which an absorbance maximum is found depends on the magnitude of the energy involved for a specific electronic transition.

Instruments designed for measuring the emission or absorption of radiant energy from substance is as follows:

1. Photometers

These give the ratio, or some function of the ratio of radiant power of two electromagnetic beams.

2. Spectrometers

These have an entrance slit, a dispersing device and one or more exit slits with which measurements are made at selected wave length within the spectral range, or by scanning over the range. The quantity detected is a function of radiant power.

3. Spectrophotometers

These have spectrometers with associated equipment so that it furnishes the ratio, or function of the ratio of the radiant power of two electromagnetic beams as function of spectral wave length. These two beams may be separated in time or space or both.

PHOTO COLORIMETER

This is an instrument used to estimate the amount of a substance present in a solution by comparing the colour of that solution with the colour of a standard solution.

Photo colorimeters or photoelectric colorimeters or absorptiometers are classified as single beam or double beam instruments according to whether there is one optical path from source to receptor or two. The double beam instrument may have two matched receptors or the radiation may be flashed alternately over the two paths to single receptor. The double beam instruments have the following advantages:

1. They permit a large degree of inherent compensation to fluctuations in the intensity of the source which might otherwise give spurious results.
2. Direct one step comparison of sample in one path with a standard or blank solution in the other path is possible in double beam instruments.

Block diagram (one cell photocolorimeter)

One cell filter photoelectric colorimeter consists of

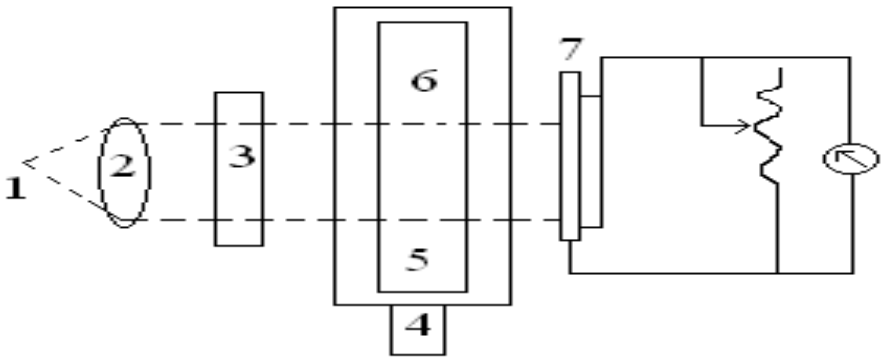
(i) A light source

(ii) A light filter

(iii) A container for the solution

(iv) A barrier layer photocell to receive the transmitted light

(v) A suitable arrangement the response of the photocell. This is illustrated in below figure.



1. Light source; 2. Collimating lens; 3. Filter; 4. Movable holder; 5. Solution; 6. Solvent; 7. Barrier type photo cell

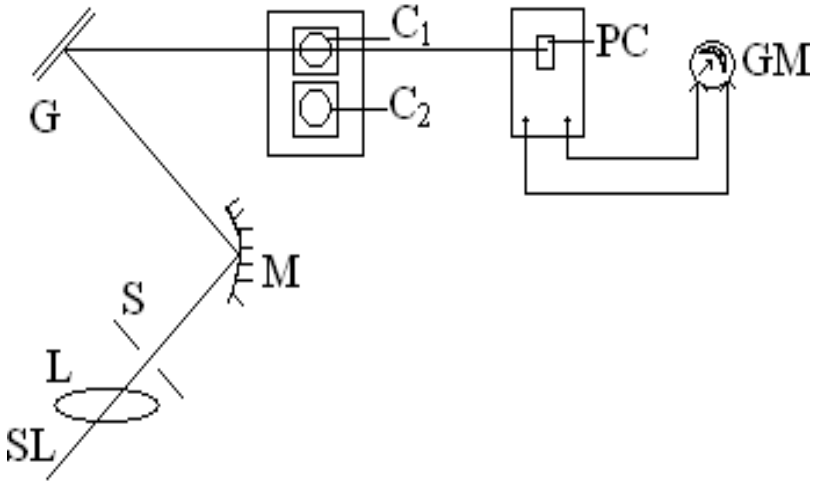
Spectrophotometers

Spectrometers are more precise than filter photometers.

Advantages of spectrometers over photometers:

1. True absorption curves can be determined.
2. Beer's law is followed. In photometers, absorptivity varies from photometer to photometer.
3. In spectrophotometers filters are replaced by monochromators.

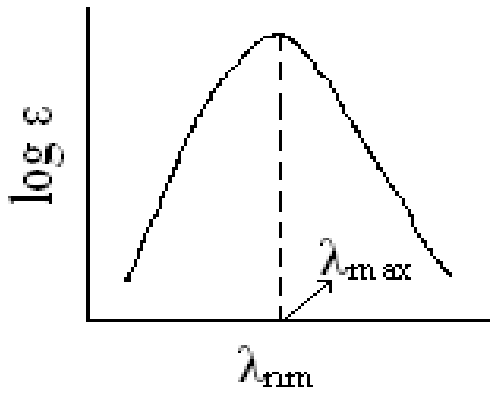
Block diagram



SL is a source of radiation of light. This may be hydrogen discharge tube in the UV region from 200-250 mμ and incandescent tungston lamps in the

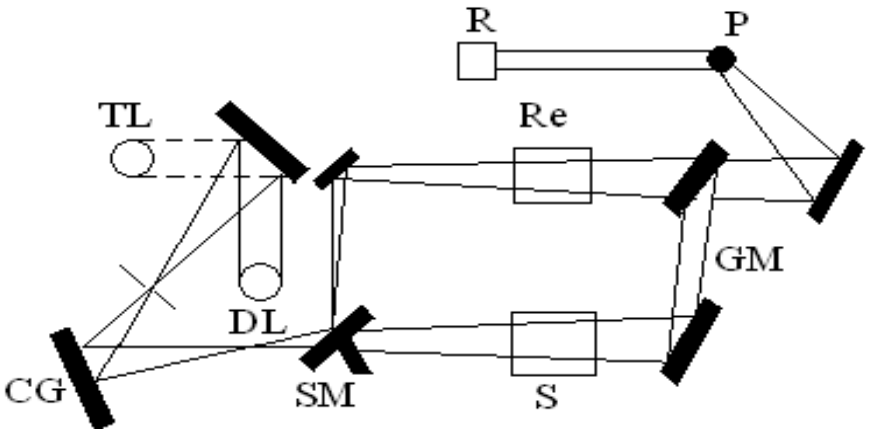
UV region 320-1100 mμ. The light from SL is allowed to pass through a lens L, and then through a slit S it falls on the mirror M. It is reflected to an optical grating G, which is provided with turning device. G divides the light in to narrow spectral regions corresponding to different wavelengths. The light of a desired wavelength emerging from the grating is allowed to pass through the cuvette C, containing the solution under examination and from there to a photo electric cell PC which is in contact with a galvanometer GM. The intensity of light which can be measured with the help of photoelectric cell PC is recorded. The cuvette C1 is now replaced by another cuvette C2 containing pure solvent and the same light is allowed to pass through it and then to photoelectric cell PC. The intensity of light is again measured by the deflection in the galvanometer as before.

A plot of molar absorbtivity (ε) or logarithm of molar absorptivity (log ε) versus wave length (λ) gives the absorption maximum. (Figure)



The main difference between photometers and spectrophotometer lies in the device used for selecting the desired spectral band width. Photometers use filters while spectro photometers use prisms or grating for this purpose.

Block diagram of a double beam spectrophotometer for UV – visible region



TL – Tungsten lamp; DL – Deuterium lamp; CG – Concave grating; SM – Sector mirror; Re – Reference; GM – Grid mirror; P – Photo multiplier; R - Recorder

Working

The light from source is dispersed by a concave grating. The dispersed beam is focused on a rotating sector mirror, which reflects the beam through the sample and reference substances. After absorption the beam reaches photomultiplier via a grid mirror. The multiplied electrical signals are recorded in a recorder.

Description of components

We shall give the description of the various components in photo colorimeter and spectrophotometer.

Light source

In order to be suitable for absorption measurements, the source of radiation must meet the following important requirements:

1. It must generate beam with sufficient power for ready detection and measurements.
2. It should be stable.
3. The source should provide continuous radiation.
4. The most common source of visible radiation is the incandescent tungsten filament lamp. Constant voltage transformers or electronic voltage regulators are used to control the voltage closely as the energy output of a tungsten lamp varies as the fourth power of the applied voltage. The source for UV is hydrogen gas lamp and deuterium lamp.

Filters

In spectroscopy we will have to use radiation of selected wavelengths. The following are the reasons for employing a radiation with in a limited wave length region.

1. The system under examination follows Beer's law when it absorbs with in a limited wave length region.
2. A greater selectivity is assured because the interference from other wavelength regions is not there.
3. A greater sensitivity is attained when wave lengths which are strongly absorbed only are employed. Only then a greater change in absorbance per increment of concentration will be observed.

Various devices are used to produce radiations of desired spectral regions. Among them the filters are the simplest and cheap. These are of two types:

Absorption filters

They absorb certain portions of the spectra there by giving radiations of desired spectral region. Thin films of gelatin containing different dyes sandwiched between two glass plates or coloured glasses are used as absorption filters.

Interference filters

They consist of a transparent, calcium or magnesium fluoride solution occupying the space between two semitransparent metallic films coated on the inside surface of two glass plates. They produce relatively narrow bands of radiation. They are based on optical interference.

The choice of the filter for any particular solution has to be done by laborious preliminary experiments. We have to select such a filter that would give maximum change in absorption for unit change in concentration.

The following table gives the choice in filters to be used for solution with various colours.

Colour of solution	Colour of filter
Orange	Blue green
Yellow	Blue
Purple	Green
Red	Blue
Violet	Yellow-green

Green	Purple
Blue green	Red orange
Blue	Yellow

Monochromator

It consists of entrance and exit slits and a dispersing device, either a prism or grating, so arranged that radiation of relatively narrow spectral band widths are obtained.

Glass prisms are used to work in visual region: quartz prisms in UV region and alkali halide prisms in IR region. Thus depending on the range in which experiments are done, we have to choose the prism.

In modern monochromators there are two dispersing elements e.g., two gratings or one prism and one grating. This arrangement reduces the amount of stray radiation to a greater extent, and also provides greater dispersion and spectral resolution.

(d) Slits

There are two slits.

1. Entrance slit
2. Exit slit

Both have same width. The light enters through the entrance slit passes through the prism or grating. The prism or grating is suitably rotated and the required wavelength alone is allowed to pass through the exit slit.

Gratings

Replica gratings are usually employed since they are cheaper than prism. One important drawback of gratings is that they give more than one order of diffraction thus causing several stray radiations. These may be eliminated either by using two gratings or by using filters in front of the entrance slit.

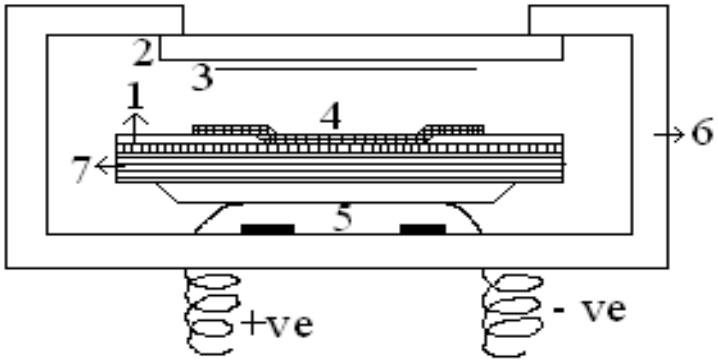
Cells / Cuvette

It must be made of such a material that it does not absorb in the range in which measurements are made. To work in UV region the cuvette should be made of quartz for the visual region they are made of colour corrected fused glass. The thickness of the cuvette will depend upon whether the absorption is weak or strong. They may be either rectangular or cylindrical in shape with flat ends.

Photocells (Detectors)

The most essential component in a photometer or spectrophotometer is the detector of radiant energy which produces a signal proportional to the radiant power falling upon it. There are several types of detectors. These are different photocells. The use of these has eliminated the errors due to the personal characteristics of individual observer.

1. Barrier – layer or photovoltaic cells



1. Selenium; 2. Glass; 3. Laquer; 4. Collector; 5. Spring contact; 6. Case; 7. Iron base

A plate of metal with a semi – conductor deposit on it such as selenium deposited on the iron base, is used in this. A very thin layer of a good conducting metal spread over the semiconductor acts as collector electrode. The metal base plate acts as cathode. Barrier cells have been found to be very sensitive and accurate in the visible region of the spectrum.

The 'EEL' selenium photocell is an example of a highly sensitive barrier layer cell. Barrier – layer cells are used where low cost and portability are required.

2. Photoemissive cell

Photoemissive cell (also called phototube) consists of a glass bulb coated internally with a thin sensitive layer such as cesium or potassium oxide and silver oxide which serves as cathode with a free space left to permit the entry of light. A metal ring inserted near the centre of the bulb forms the anode and this is maintained at a high voltage by means of a battery. The bulb is either evacuated or filled with an inert gas at low pressure (for example argon at about 0.2 mm). When light, penetrating the bulb, falls on the sensitive layer, electrons are emitted, there by causing a current flow through an out side circuit. This current is amplified and it is taken as a measure of the amount of light striking the photosensitive surface.

Preparation of samples

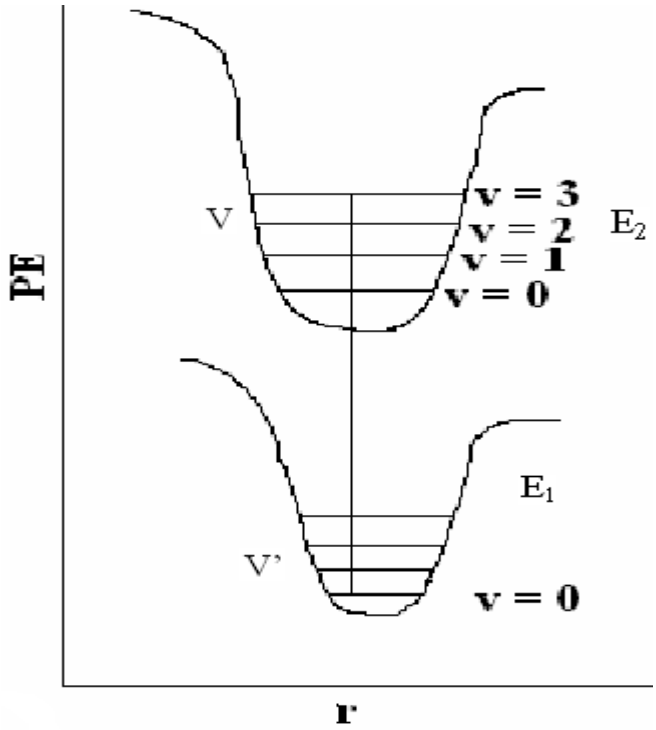
Samples for recording UV – visible spectra are prepared by dissolving the substance to be analysed in suitable solvents. For visible spectroscopy water is commonly used as solvent. The substance to be analysed may be coloured or colourless. If is it colourless, some colour forming substances are added to produce colour. The added substance should not absorb during recording.

In UV – spectroscopy also water is the common solvent for many inorganic compounds. Cyclohexane is suitable for organic compounds. When more polar solvents are required 95% ethanol is used.

Types of electronic transitions

Potential energy curves

The way in which the potential energy of a diatomic molecule varies with the internuclear distance in two different electronic levels is shown in figure.



E = Energy; r = Internuclear distance; E_1 = Ground state electronic level; E_2 = Excited state electronic level; V' = vibraional levels in the ground state electronic level; V = Vibrational level in the excited state electronic level; 0, 1, 2…….. = Vibrational quantum numbers.

Figure

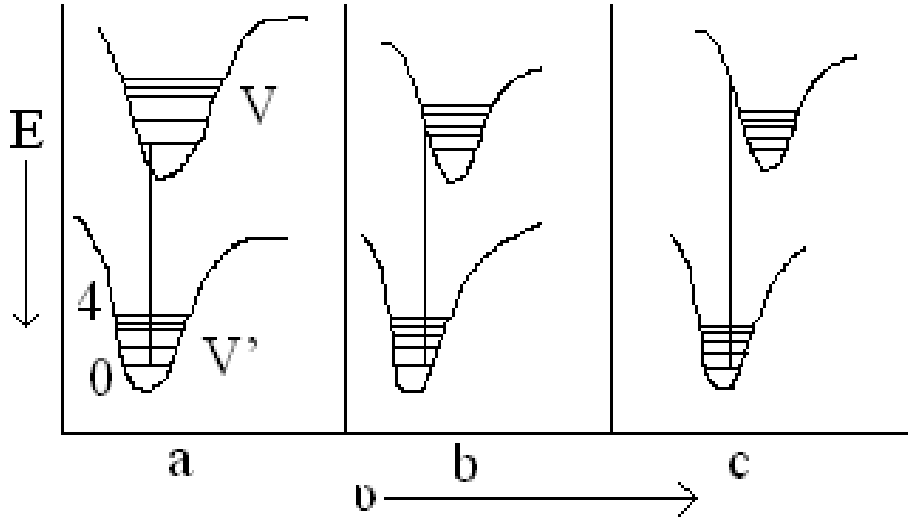
Considering transition from one electronic level (E_1) to the other (E_2) three factors decide which transition takes place and from where to where. **(Selection rules)**

(i) ΔV is not restricted to ± 1 as in vibrational transition it may be 0, $\pm$ 1, 2……

(ii) **Frank – Condon principle:** An electronic transition in a molecule takes place so rapidly compared to the vibrational motion of the nuclei, that the inter – nuclear distance can be regarded as fixed during the transition. Therefore the transition occurs vertically as shown in the diagram.

(iii) Electronic transitions are more probable if they begin or end at the middle of V = 0 or at either end of any of the higher vibrational levels.

Comparing the three factors given, the expected electronic transitions may be represented as in below figure.



Let us demonstrate the Frank – Condon principle for the electronic transition of a diatomic molecule. Consider the above figure b where two potential energy curves for the molecule in the ground state and in the first excited electronic state. Since the bonding in the excited state is weaker than in the ground state the minimum in the potential energy curve for the excited state occurs at a slightly greater inter nuclear distance than the corresponding minimum in the ground electronic state. Also according to quantum mechanics it is known that the molecule is in the centre of the ground vibrational level of the ground electronic state. Thus when a photon falls on the molecule the most probable electronic transition according to Frank – Condon principle takes place from V' = 0 to V = 2. Transition to other vibrational levels of the excited

electronic state occur with lower probabilities so that their relative intensities are smaller than the intensity of V' = 0 to V = 2 transitions as shown in above Figure.

The absorbing species may include transitions involving

(a) π, σ and n (non-bonding) electrons

(b) d and f electrons

(c) Charge transfer electrons.

(i) σ → σ* Transitions

Saturated hydrocarbons involve this type of transition. These occur in far UV region (150 nm).

(ii) n → π * Transitions

Unsaturated molecules containing oxygen, nitrogen and sulphur show this type of transition. These occur in the 250 – 270 nm region.

(iii) n → σ* Transitions

Saturated compounds with lone pair electrons show this type of transition. These occur just below 200 nm.

(iv) π → π * Transitions

This corresponds to promotion of an electron from a bonding π orbital to an antibonding π* orbital. These occur between 170 and 200 nm. The energies required for the transitions are in the order

$$\sigma \rightarrow \sigma^* > n \rightarrow \sigma^* > \pi \rightarrow \pi^* > n \rightarrow \pi^* \text{ (in below Figure)}$$

Chromophore

Definition

They are groups with multiple bonds. When these compounds are present in a molecule, it exhibits colour.

Examples

Nitroso (-N=O), Azo (-N=N-), carbonyl (>C=O), Thiocarbonyl (-C=S), Polyene $-(CH = CH)_n$, Nitro ($-NO_2$)

Many organic compounds are coloured. When they were analysed it was found that colour usually appeared in an organic compound when it contained groups with multiple bonds. Such groups are responsible for imparting colour to a molecule. Such groups are called chromophores. The compound containing a chromophoric group is called a chromogen.

If a chromogen contains only one chromophoric group the compound is usually yellow. As the number of chromophores present in a molecule increases the depth of the colour of the molecule also increases.

Auxochrome

Definition

They are groups which by themselves can not impart colour to a molecule. But when they are introduced in to a compound containing a chromophoric group then they deepen the colour of the chromogen.

Examples

Hydroxyl (-OH), alkoxy (-OR), amine (-NH_2), alkylated amino (-NHR, -NR_2) etc.

Auxochromes when they are present in a molecule they do not impart colour to the molecule. But if they are present along with a chromophore in the molecule then the molecule has deeper colour. If a substance has a chromophore along with an auxochrome then the substance will be usually a dye. Thus the presence of an auxochrome is necessary to make a chromogen, a dye.

Difference between chromophore and auxochrome

S. No	Chromophore	Auxochrome
1.	It is a group which causes colour in a molecule.	It is a group which can not cause colour to a molecule. But if it is present along with a chromophore in a molecule, then, it intensities the colour of the molecule.
2.	They are not salt forming groups.	They are salt forming groups.
3.	They have a multiple band.	They have a lone pair of electrons.
5.	**Examples** -NO, -NO_2, -NNO, -N_2-, >CO etc.	**Examples** -OH, -OR, -NH_2, -NHR, -NR_2 etc

λ_{max} and intensity of absorption bands

Compounds absorb light at various wavelengths. The position of the maximum point of absorption band is given by λ_{max}. The intensity of the band at this maximum is given by ε_{max}. The molar absorptivity at an absorption maximum ε is given by

$$\varepsilon = A / Cx = [\log (I_0 / I)] / Cx$$

Where $\log (I_0 / I)$ = Absorbance = A of the solution; C = Concentration of the solution in mol. dm^{-3}; x = The path length of the sample in cm; I_0 = The intensity of the incident light (or the light intensity transmitted through a

reference cell) and I = Intensity of the light transmitted through the sample solution.

Some simple organic chromophores and the approximate wavelengths at which they absorb, their molar absorptivity at the absorption band maximum are given in the following table.

Table

Chromophore	λ_{max} **nm**	ε_{max} **X 10^{-2} m^2 mol^{-1}**
>C = C <	175	14,000
-C ≡ C -	175 170 195 223	10,000 150
> C = O	160 185 280	18,000 5.000 15
RNO_2	200 274 165	5,000 15 5
Benzene	184 204 255	60,000 7,400 204

The positions of an absorption band (λ_{max}) may shift to shorter wave lengths or to longer wave lengths due to several factors. Similarly ε_{max} may increase or decrease. Some older established terms should be mentioned here, although the use should be progressively discouraged.

(i) Red shift or bathochromic shift

The shift of absorption maxima towards longer wavelength.

(ii) Blue shift or hypsochromic shift

It is a shift towards shorter wave length.

(iii) Hyperchromic effect

An effect increasing the intensity of absorption.

(iv) Hypochromic effect

An effect decreasing the intensity of absorption.

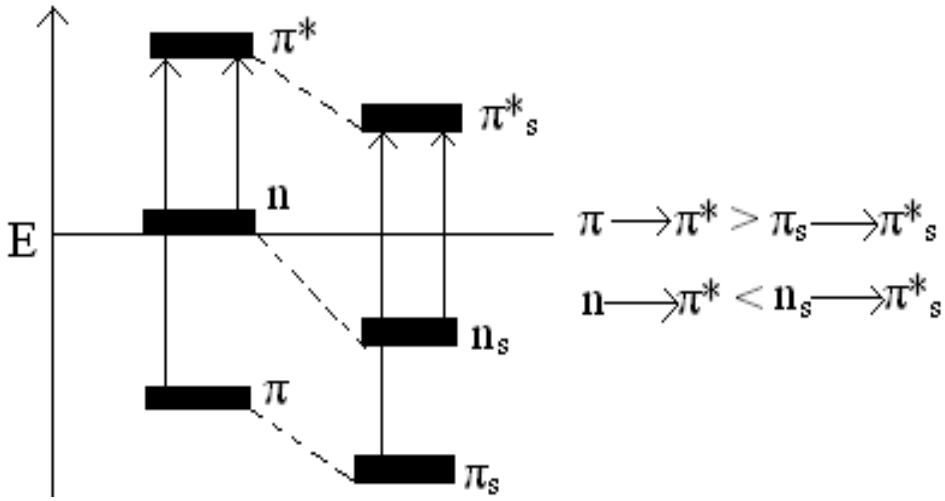
Factors affecting λ_{max} and ε

1. Solvent effects

The position and intensity of an absorption band may shift when spectrum is recorded in different solvents. For changes to solvents of increased polarity the shifts in the spectrum will be as follows:

1. Conjugated dienes and aromatic hydrocarbons experience very little shift.
2. α, β-unsaturated carbonyl compounds show two different shift
   a) The π → π* band moves to longer wave lengths (red shift).
   b) The n → π* band moves to shorter wave lengths (blue shift).

We can express this general picture in the from of an energy diagram as in figure.

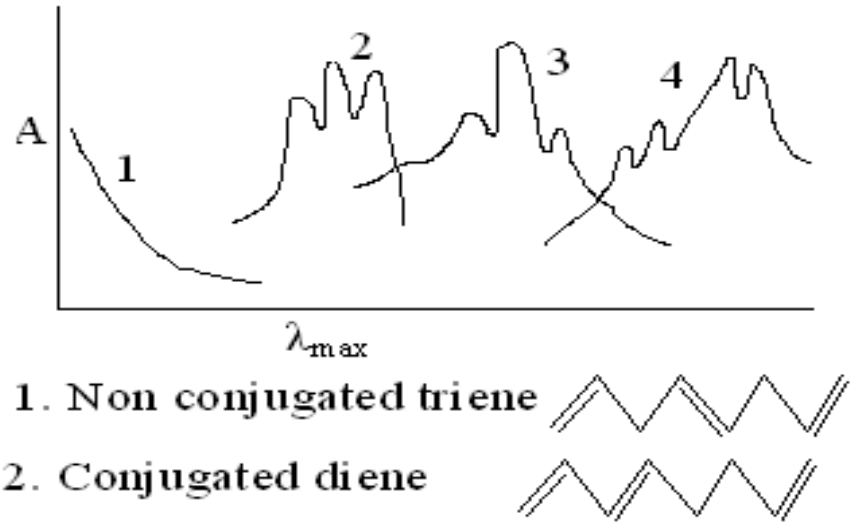


Solvation

Solvation by polar solvents stabilizes π, π* and n orbitals. The stabilization of n orbitals (non bonding orbitals) is particularly pronounced with hydrogen bonding solvents such as water or ethanol and π* orbitals are more stabilized by solvation than π orbitals, presumably because π* orbitals are more polar ones than π orbitals. The net result is the energy transition π → π* becomes less with salvation (red shift) while the energy of transition n → π* becomes greater (blue shift).

Conjugation

Increased conjugation increases λ_{max}. The following figure illustrates the point.



The λ_{max} values reach 450 nm for 11 or 12 double bonds in conjugation. Such polyenes are strongly yellow in colour. The red colour of tomatoes and carrots arises from conjugated molecules of this type.

Substituents present

Presence of substituents alters the position of λ_{max}. For example, simple alkyl substituents in benzene shift the absorptions slightly to longer wave lengths. Substituents with lone pair of electrons (like –OH, -OR, $-NH_2$ etc) shift the absorptions more substantially to longer wave lengths.

Altering the nonbonding pair availability also alters the position of λ_{max}.

E.g.

(i) When p – nitrophenol is converted by base to p – nitro phenolate ion there is a pronounced red shift.

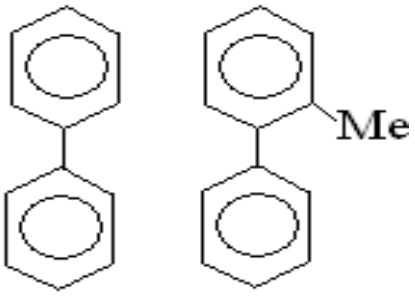
(ii) When amino groups are protonated, a blue shift results.

Stereochemical factors

Angular strain or steric over crowding distort the geometry of the chromophores. This leads to reduction of conjugation because of reduction in orbital over lapping. This also causes shifts in $\lambda_{max.}$

Example

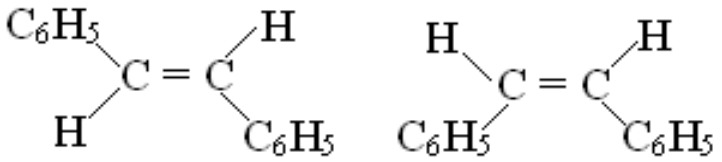
(i) Biphenyl (I) is not completely planar. The two benzene rings are at an angle of about 45^o. In 2 – substituted biphenyls (II), the two rings are pushed even further out of coplanarity. The result is that diminished π – orbital overlap in (II) leads to blue shift and diminished intensity in their electronic spectra.



	I	II
λ_{max}	250	237
ε	19,000	10,250

(ii) Where an alkene chromophore is capable of geometrical isomerism, it is normally found that the trans – isomer exhibits longer wavelength absorption (and higher intensity) than the cis – isomer. This is because there is more effective π – orbital overlap in the trans isomer.

Example



	Trans – stilbene	Cis – stilbene
λ_{max}	230 : 299 : 312	225 : 282
$\log \varepsilon_{max}$	4.2 4.5 4.5	4.34 4.1

It is generally not possible to predict ε_{max}. It may be stated that longer conjugation leads to higher intensity. In many cases ε is approximately proportional to the square of the length of chromophore.

Applications

UV/Vis spectroscopy is routinely used in the quantitative determination of solutions of transition metal ions and highly conjugated organic compounds.

- Solutions of transition metal ions can be coloured (i.e., absorb visible light) because d electrons within the metal atoms can be excited from one electronic state to another. The colour of metal ion solutions is strongly affected by the presence of other species, such as certain anions or ligands. For instance, the colour of a dilute solution of copper sulfate is a very light blue; adding ammonia intensifies the colour and changes the wavelength of maximum absorption (λ_{max}).

- Organic compounds, especially those with a high degree of conjugation, also absorb light in the UV or visible regions of the electromagnetic spectrum. The solvents for these determinations are often water for water soluble compounds, or ethanol for organic-soluble compounds. (Organic solvents may have significant UV absorption; not all solvents are suitable for use in UV spectroscopy. Ethanol absorbs very weakly at most wavelengths.) Solvent polarity and pH can effect the absorption spectrum of an organic compound. Tyrosine, for example, increases in absorption maxima and molar extinction coefficient when pH increases from 6 to 13 or when solvent polarity decreases.
- While charge transfer complexes also give rise to colours, the colours are often too intense to be used for quantitative measurement.

4.4. COLORIMETRY

Principle

The basic principle of colorimetric measurement consists of comparing the colour intensity produced by an unknown amount of the substance with the colour intensity produced by a known amount of the substance under the same conditions. For this several experimental methods are available.

Colorimetric titration

In this method a known volume of the 'test' solution is taken in a Nessler tube, the colour is developed by the addition of the reagent and the solution is made up to a definite volume, say, 50 ml. In another tube almost an equivalent quantity of water (slightly less) is taken together with the reagent. A solution containing the known amount of the substance being determined is run in to this Nessler tube from a burette till the colours produced in the two tubes match. From the volumes of the 'test' solution taken and the standard solution added, the concentration of the 'test;' solution can be calculated. Since the volumes of the solutions in the two Nessler tubes will be different, several determinations should be carried out by adjusting the volume of water taken initially in the blank. This method can be used only when the colour is independent of the order of adding various components to produce the colour. The method can not be used where the substance to be determined requires a preliminary treatment before the colour is developed. For example, in the estimation of Ni^{2+} using dimethyl glyoxime, the nickel solution has to be pretreated with bromine water and therefore the titrimetric method can not be used.

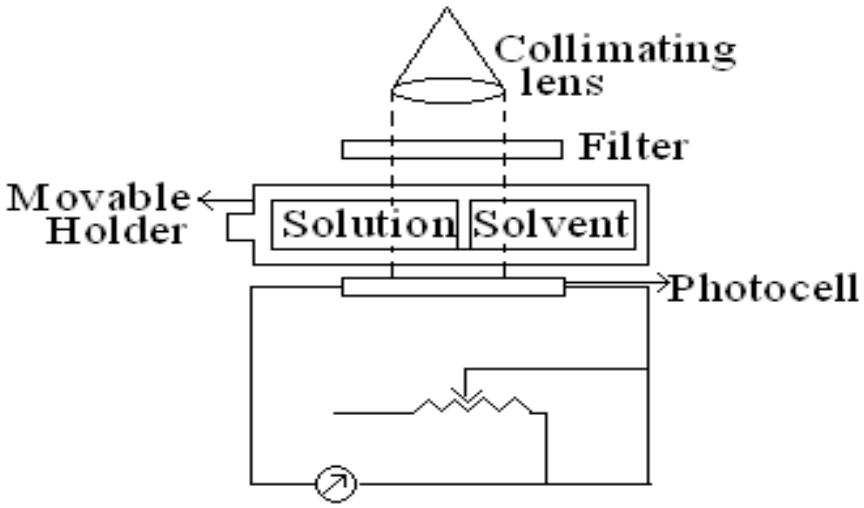
Advantages of colorimetric analysis

1. The operations involved in the estimation are simple, especially when a photoelectric colorimeter is available.
2. This method is not time – consuming therefore, it is ideally suited for routine work.
3. Determination of minute quantities of substances is possible.
4. The instruments / apparatus required are expensive.

Photoelectric Colorimeter

Progress in the development of colorimetric methods has resulted largely from application of the photoelectric cell to the problem of colour measurement. This improvement has eliminated the difficulties which generally complicate visual comparisons. A photoelectric colorimeter makes use of a light – sensitive device like photoemissive cell. A photoemissive cell (also called phototube) makes use of cathode which emits electrons when illuminated. The emission of electrons causes a current to flow through an outside circuit. This current may be amplified and taken as a measure of the intensity of light striking the photosensitive substance.

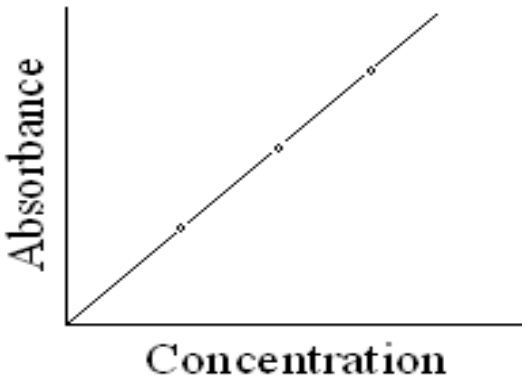
A number of commercial photoelectric colorimeters are available. These essentially consist of a light source, a light filter, a container for the solution, a photocell to receive the transmitted light and a device to measure the current produced in the photocell. A schematic diagram of the set – up is shown in Fig.



Filter consists of coloured glass or glass coated with coloured gelatin. These transmit light from a narrow region of the spectrum, absorbing at the same time other components. Use of an appropriate filter increases the sensitivity of measurements.

Using a sliding sample carriage either the reference solution or the pure solvent taken in small cells can be brought in to position, i.e., across the path of the light. The current measurements are made using a sensitive micro ammeter calibrated in percentage transmission and optical density. After adjusting the percentage transmittance of pure solvent to a value of 100 using a rheostat, readings are taken on the scale for the solution.

Standard solutions of different concentrations, preferably in the range of the unknown, are prepared and for each one of these solutions the absorbance is measured. A plot of absorbance versus concentration is drawn. From the graph the concentration of the 'test' solution can be found, using the value of absorbance in below figure.



Absorbance Vs Concentration

Estimation of copper

Principle

Cupric iron reacts with dithiocarbomate ion to give a series of soluble green coloured compounds. The colorimetric titration method can be used.

Procedure

(i) Preparation of Standard solution

0.0864 g of AR copper sulphate is dissolved in about 5 ml of dilute HCl and made up to 100 ml. One ml of this solution contains 0.1 mg of Cu.

(ii) Preparation of the solution for Estimation

A known quantity of the sample in which the amount of copper is to be determined is dissolved in a suitable acid and the excess acid is expelled by heating.

(iii) Estimation

10 ml of the solution for estimation are prepared out in to a Nessler tube. 2 ml of concentrated HCl and 5 ml of a 2 M solution of diethylammonium dithiocarbomate are added. The solution is made up to 50 ml and stirred. In

another Nessler tube about 40 ml of water are taken and 2 ml of concentrated HCl and 5 ml of diethylammonium dithiocarbomate are added. The standard solution is run in to Nessler tube till the colours match. The volumes of the solution in the two Nessler tube will be different. The procedures is repeated by adjusting the volume of water and that of the standard solution taken in the second Nessler tube till the volumes in the two tubes are same and the colours also match.

Estimation of iron

Principle

Ferric iron reacts with thiocyanate ion to give a series of soluble red – coloured compounds. These complexes have the general formula $[Fe(CNS)_n]^{3-n}$ where n = 1,2,3…..6. At very high thiocyanate ion concentrations the species present is $[Fe(CNS)_6]^{3-}$. This complex is not only stable but has an intense red colour. Therefore, a large excess of thiocyanate ion is used in the estimation. The colorimetric titration method can be used.

Procedure

(i) Preparation of Standard solution

0.0864 g of AR ferric ammonium sulphate is dissolved in about 5 ml of dilute HCl and made up to 100 ml. One ml of this solution contains 0.1 mg of Fe.

(ii) Preparation of the solution for Estimation

A known quantity of the sample in which the amount of iron is to be determined is dissolved in a suitable acid and the excess acid is expelled by heating. If total iron present in the sample is to be estimated, any ferrous iron present has to be oxidized with dilute $KMnO_4$, and the solution made up to a known volume. If one is interested in the amount of only ferric iron, the oxidation step is omitted.

(iii) Estimation

10 ml of the solution for estimation are prepared out in to a Nessler tube. 2 ml of concentrated HCl and 5 ml of a 2 M solution of ammonium thiocyanate are added. The solution is made up to 50 ml and stirred. In another Nessler tube about 40 ml of water are taken and 2 ml of concentrated HCl and 5 ml of ammonium thiocyanate are added. The standard solution is run in to Nessler tube till the colours match. The volumes of the solution in the two Nessler tube will be different. The procedures is repeated by adjusting the volume of water and that of the standard solution taken in the second Nessler tube till the volumes in the two tubes are same and the colours also match.

Estimation of nickel

Principle

Nickel is estimated gravimetrically by precipitating it as its nickelous complex with dimethylglyoxime. However, in the presence of an oxidizing agent like bromine water, a soluble, red complex is formed which contains nickel in the higher valence state (III and IV). This reaction is the basis of the colorimetric estimation of nickel.

Procedure

(i) Preparation of Standard solution

A standard solution containing 0.1 mg of Ni in 1 ml is prepared by dissolving 0.1683 g of AR nickel ammonium sulphate in 250 ml of water. The solution may be further diluted, if necessary.

(ii) Estimation

Different volumes of the standard solution of nickel are pipetted in to Nessler tubes kept in a rack. The colour is developed by adding 1 ml of saturated bromine water, 2 ml of ammonia and 1 ml of 1% dimethylglyoxime in alcohol to each Nessler tube.

10 ml of the solution of nickel salt in which nickel has to be estimated are pipetted out in to another Nessler tube and the colour is developed by adding 1 ml of bromine water, 2 ml of ammonia and 1 ml of 1% dimethylglyoxime. The colour produced in this tube is compared with the colours produced with standard solutions.

Important Questions

1. Explain theory of IR spectroscopy
2. Discuss the stretching and bending modes of vibrations
3. Discuss the instrumentation of IR spectroscopy
4. Explain the vibrational modes of CO_2
5. How will you study hydrogen bonding by using IR spectroscopy?
6. Interpret the IR spectra of anisole, benzaldehyde and toluene
7. What are Raman and Rayleigh scattering?
8. What are stokes and antistokes lines?
9. Write the instrumentation of Raman spectroscopy
10. What are the differences between IR and Ramn spectroscopy
11. Explain mutual exclusion principle

12. Write the applications of Raman spectroscopy?
13. Define and explain Beer-Lambert's law
14. Describe the various components of UV spectrophotometer
15. Explain various types of electronic transitions
16. Write notes on chromophore and auxochromes
17. What are the factors affecting wavelength maxima and intensity of radiation?
18. How will you estimate nickel by colorimetry?
19. How will you estimate iron by colorimetry?
20. What is the principle and application of colorimetry?
21. Discuss the theory of UV-Visible spectroscopy
22. Discuss the instrumentation of UV-Visible spectroscopy

NOTES

UNIT – V

^{1}HNMR SPECTROSCOPY

5.1. NMR SPECTROSCOPY

Nuclear magnetic resonance (NMR) is a branch of spectroscopy in which radiations of radio wave frequency is absorbed by spinning nucleus. This involves the magnetic energy of nuclei when they are placed in a magnetic field and the transitions occur in the region of the spectrum. In general, the study of the absorption of radio frequency radiation by nuclei is called nuclear magnetic resonance spectroscopy. The magnetic spectra that arise from proton alone are called Proton Magnetic Resonance (PMR) spectra.

Principle

We know that electrons have spin. In a similar manner the nuclei of atoms also have spins. Because the nuclei spin, they have spin angular momentum, given by

$$\sqrt{I\,(I+1)}\ \frac{h}{2\pi}$$

Where 'I' is the spin quantum number of the nucleus. I may have values of 0, ½, 3/2, 2……. depending on the particular nucleus. The numerical value of the nuclear spins quantum number I is related to the mass number and the atomic number as given in below table.

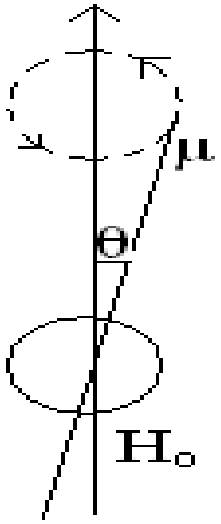
Mass number	**Atomic number**	**Spin quantum number**	**Examples**
Odd	Even or Odd	½, 3/2, 5/2…..	^{1}H, ^{19}F, ^{31}P etc
Even	Even	0	^{12}C, ^{16}O, ^{32}S etc
Even	Odd	1, 2, 3…..	^{14}N etc

The nuclei with I = 0 are non – magnetic. Hence ^{12}C, ^{16}O etc., cannot be observed by NMR spectroscopy. H^{1} nucleus has I = ½ and hence can be studied.

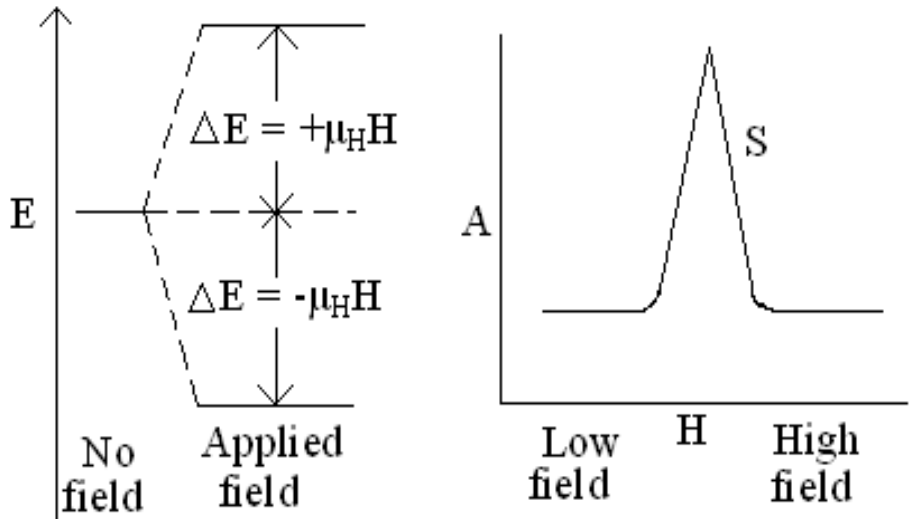
Any spinning has a magnetic moment. It is impossible to calculate the magnetic moment of a nucleus because the exact shape for the given nucleus is uncertain. But the magnetic moment values can be determined experimentally. If a magnetic nucleus is placed in an external magnetic field of strength H, the nuclear spin axis will lie at an angle ɵ to the field and the nucleus will process about the axis of the field. The frequency of precession of the given nucleus is directly proportional to the strength of the applied field H and is given by the Larmor equation,

$$\omega = 2\pi v = \gamma H$$

Where ω = angular velocity, v = frequency of precession and γ = magnetogyric ratio, a constant for the given nucleus.



Most of these NMR studies have been hydrogen nucleus for which I is equal to ½. If a proton placed in an external magnetic field its magnetic moment can be aligned with the field. This alignment is more stable a situation. On the other hand the magnetic moment of the proton can be aligned against the external field. This alignment is less stable. If H is the external field strength the magnetic moment of the proton is μ_H then the energy of the proton is given by $\mu_H H$. The energy required to promote (flip) a proton from the lower stable level to the higher unstable level is given by $2\mu_H H$ in below figure.



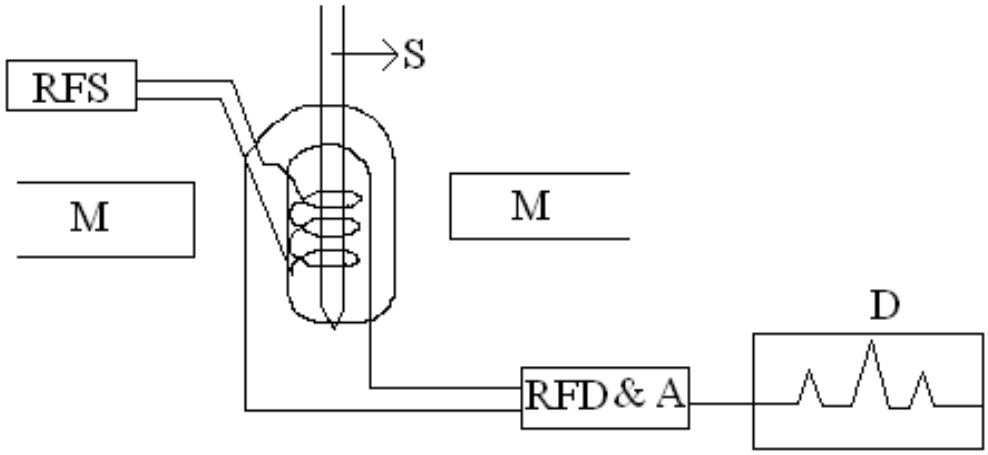
Now if a fixed magnetic field is applied on the sample and the frequency of the radiation is varied, the radiation is absorbed when

$$2\mu_H H = h\nu$$

Alternately the frequency of radiation may be fixed and H may be varied. At some value of the field strength, the energy required flipping the proton matches the energy of the radiation, absorption occurs and a single peak is observed. Such a spectrum is called NMR spectrum.

Basic instrumentation

Block diagram



Component description

There are three main components in an NMR instrument. They are

1. A radiation source
2. A dispersion device
3. A detector

1. Sample source (S)

The sample is taken in a glass tube of uniform dimension. The sample tube is spinning about an axis perpendicular to the field axis. The spinning minimizes the effects of in homogeneities in the field, so as to get sharper lines and better resolution.

2. Magnet (M)

Permanent electromagnet is used in an NMR instrument. The magnetic field should be homogeneous. The field strength is 1.4 Tesla. At this field strength the oscillator frequency needed is 60 MHz.

3. Field sweep generator

A pair of coils on the face of magnets alters the applied field over a small range.

4. Radio frequency source (RFS)

The signal radio frequency oscillator is fed in to the pair of coils. They are 90^o to the field path. A fixed 60, 90 or 100 MHz is used.

5. Radio frequency detector (RFD)

The radio frequency signal produced by resonating nuclei is detected by means of a coil. This coil is at right angle to the source coil.

6. Amplifier (A)

The signal got by RFD will be weak. So an amplifier is attached to RFD to amplify the signal.

7. Display (D)

The signal is recorded in pre – printed chart. Peak areas show the relative number of absorbing nuclei in a given chemical environment.

Sample preparation

About 30 mg of the sample, if it is a solid or a viscous liquid, is dissolved in about 0.4 ml of the solvent. A substance free of proton should be used as a solvent, so that it does not give its own NMR spectrum. It should be capable of dissolving at least 10 % of the substance under investigation. Some commonly used solvents in NMR spectroscopy are: CCl_4 (Carbon tetra chloride), CS_2 (Carbon disulphide), $CDCl_3$ (Deutro chloroform), $(CCl_3)_2CO$ (Hexachloroacetone) etc.

Operation

Now – a – days the radio frequency source is kept at 60, 90 or 100 MHz. The field strength is varied to make the precessional frequency of the nuclei to be 60, 90 or 100 MHz. It is called field sweep.

The sample is held in a glass tube in the form of a solution, with a trace of reference (TMS). The radio frequency oscillator sends radiation of 60 MHz by means of the coil placed around the sample. The magnetic field is slowly raised. At resonance condition the nucleus absorbs energy from the oscillator. When it comes to ground state the emitted energy is collected by the detector and passed on to the recorder.

At a given radio frequency all protons in a molecule may give NMR signals at different applied field strengths. This applied field strength is measured. It is plotted against the absorption. We get a spectrum with many

absorption peaks. From the relative positions we can get the detailed information about the molecular structure.

We know that the resonance condition in NMR is $2\mu_H H = h\nu$. This energy corresponds to the energy difference between the lower stable energy level and the higher unstable energy level of a proton. This value of hν is very small. So the NMR spectrum is taken at low temperatures. Because at higher temperatures, both the energy levels become equally populated and so flipping of proton, when the resonance condition is reached, will not take place.

Number of signals

In a given molecule protons with same environments absorb at the same applied field strength. A set of protons with the same environments are said to be equivalent. Protons with different environment absorb at different applied field strengths. The number of signals in an NMR spectrum tells us how many sets of equivalent protons are there in a molecule.

Example

(i) $CH_3 - CH_3$

a a

Ethane

1 NMR signal

(ii) $CH_3 - CH_2 - Cl$

a b

Ethyl chloride

2 NMR signal

(iii) $CH_3 - CHCl - CH_3$

a b a

Isopropyl chloride

2 NMR signals

(iv) $CH_3 - CH_2 - CH_2 - Cl$

a b c

n – Propyl chloride

3 N MR signals

Peak area and proton counting

The area under an NMR signal is directly proportional to the number of protons giving rise to the signals.

Example

CH_3OH	$CH_3 - CH_2 - OH$	$CH_3 - CH_2 - CH_2 - OH$
a b	a b c	a b c d
3 : 1	3 : 2 : 1	3 : 2 : 2 : 1
2 NMR signals	3 NMR signals	4 NMR signals

Areas under NMR signals are found by integration using electronic integrators. From the ratio of areas we can get the number of equivalent protons present.

Chemical shift

The difference between the magnitudes of the magnetic field at which free nuclei and molecular nuclei resonate is called chemical shift or the shift in positions is known as chemical shift.

The separation in the position of the spectral signal of hydrogen atoms in different chemical environment from a standard is known as chemical shift δ. Chemical shift is defined as

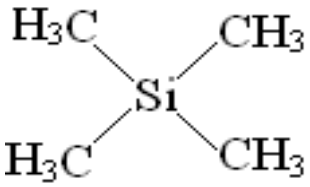
$$\delta = \frac{H_{sample} - H_{reference}}{H_{reference}} X\ 10^6$$

The reference chosen is usually tetramethyl silane (TMS), $(CH_3)_4Si$ 10^6 is introduced because the absorption takes place at fields that differ by the order of milligauss. Chemical shift is also represented by T (Tau) where

$$T = 10 - \delta$$

Reasons for using TMS as the reference

1. It has 12 equivalent protons. Since all the protons are having the same environments TMS gives a single intense peak in its NMR spectrum.



2. It has a low boiling point (27°C). So it can be easily recovered after the spectrum is recorded.
3. It is chemically inert. So it will not react with substances under study.
4. The methyl groups in TMS are highly shielded by their electrons from the external magnetic field. So TMS shows NMR signals at a very high magnetic field strength compared to other protons.

Significance

Chemical shifts indicate the type of environment that a particular proton has. The following table gives the chemical shifts for hydrogen in different environments.

Type of hydrogen	δ (ppm)
Methyl hydrogen	
$(CH_3)_4Si$	0.00
$CH_3CH_2CH_3$	0.85
CH_3CH_2Cl	1.40
CH_3Cl	3.05
Methylene hydrogen	
Cyclopropane	0.30
$CH_3CH_2CH_3$	1.25
Cyclohexane	1.5
CH_3CH_2OH	3.6
Aromatic proton	
In benzene	7.27
In toluene	7.10
In naphthalene	7.23

Thus we find that different protons in different environments exhibit different chemical shifts. So we get different NMR signals. Thus chemical shifts help us in interpreting the NMR spectra.

Factors affecting chemical shifts

1. Electronegativity

Table shows the chemical – shift positions for CH_3 protons when a methyl group is attached to functions of increasing electronegativity. As the electronegativity of the function is increased, CH_3 protons come to resonance at higher δ values.

Electronegativity groups, like fluorine in CH_3F, withdraw electron density from the methyl group (inductive effect) and this deshielding effect means that a lower value of the applied magnetic field is needed to bring the CH_3 protons to resonance. Fluorine is more electro – negative than chlorine, so the protons in CH_3F appear at higher δ values than those in CH_3Cl.

$$CH_3F > CH_3Cl > CH_3Br$$

Compound	Chemical shift δ
$CH_3 - \overset{\vert}{\underset{\vert}{Si}} -$	0.0
CH_3I	2.16
CH_3Br	2.65
CH_3Cl	3.10
CH_3F	4.26

2. Vander waals deshielding

In an rigid molecule it is possible for a proton to occupy a sterically hindered position and in consequence the electron cloud of the hindering group will tend to repel, by electrostatic repulsion, the electron cloud surrounding the proton. The proton will be deshielded and appear at higher δ values than would be predicted in the absence of the effect. Although this influence is small (usually less than 1 ppm) it must be borne in mind when predicting the chemical shift positions in overcrowded molecules such as highly substituted steroids or alkaloids.

3. Anisotropic effects (Space effect)

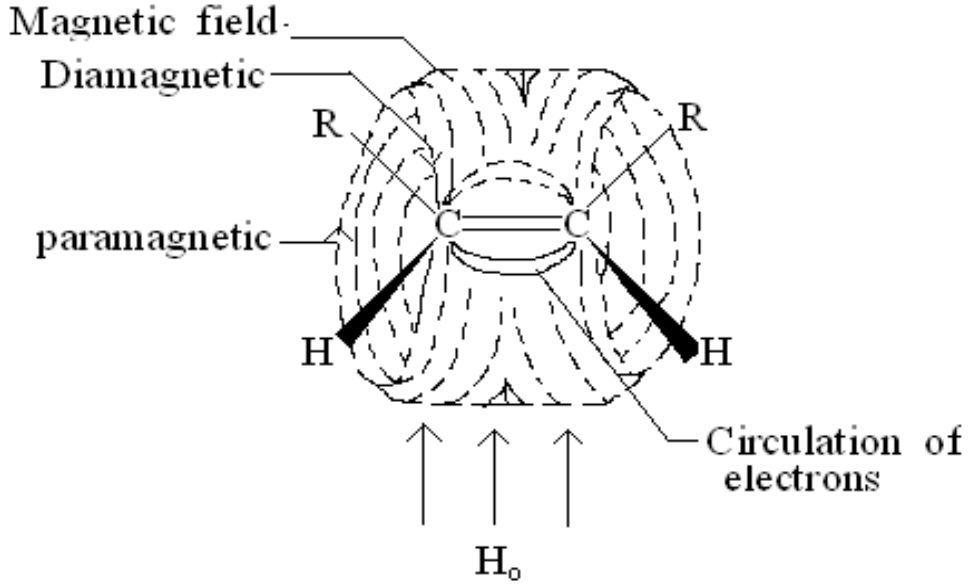
Circulation of electrons especially the π – electrons about nearby nuclei generates an induced field which can either oppose (causing shielding) or reinforce (causing deshielding) the applied field at the proton, depending upon the localization of the proton or the space occupied by the proton. In the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

The chemical shift positions (δ) for protons attached to C = C in alkenes is higher than can be accounted for by electro negativity effects alone. The same is true of aldehydic protons and aromatic protons. Alkene protons appear at relatively low δ.

(a) Alkenes

When an alkene group is so orientated that the plane of the double bond is at 90^o to the direction of the applied field (as in figure) induced circulation of the π – electrons generates a secondary magnetic field, which is diamagnetic

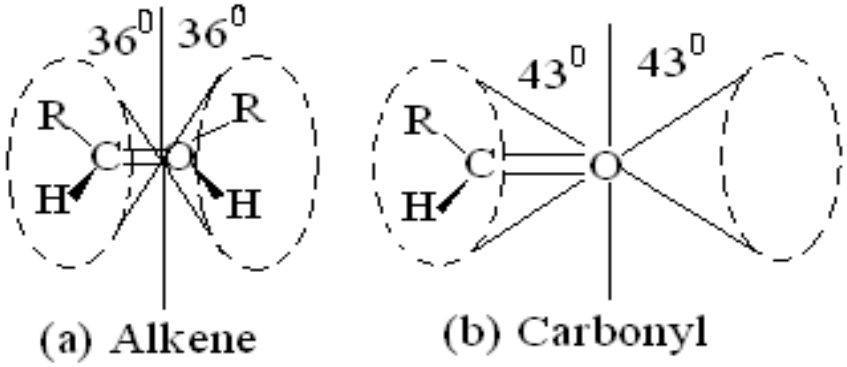
around the carbon atoms, but paramagnetic (that is, it augments B_0) in the region of the alkene protons.



Where the direction of the induced magnetic field is parallel to the applied field B_0, the net field is greater than B_0. Protons in these zones require a lower value of B_0 to come to resonance and therefore appear at lower field (higher δ values) than expected.

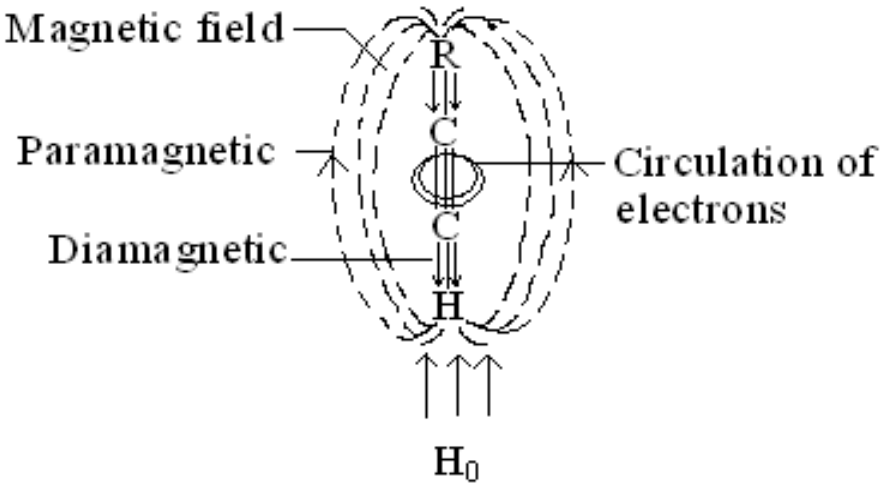
(b) Carbonyl compounds

For the carbonyl group, a similar situation arises, although the best representation of shielding and deshielding zones is slightly different from the alkene pattern. Two cone shaped volumes, centred on the oxygen atom, lie parallel to the axis of the C = O bond; protons within these cones experienced deshielding, so that aldehydic protons and the formyl protons of formate esters appear at high δ values. Protons held above or below these cones will come to resonance at lower δ values.



(c) Alkynes

Whereas alkene and aldehydic protons appear at high δ values, alkyne protons appear around δ 1.5 – 3.5. Electron circulation around the triple bond occurs in such a way that the protons experience a diamagnetic shielding effect. Figure shows how this arises, when the axis of the alkyne group lies parallel to the direction of B_0. The cylindrical sheath of π – electrons is induced to circulate around the axis and the resultant annulus – shaped magnetic field acts in a direction that opposes B_0 in the vicinity of the protons. Higher B_0 values are needed to bring the protons to resonance; therefore acetylenic protons appear at low δ values in the spectrum.



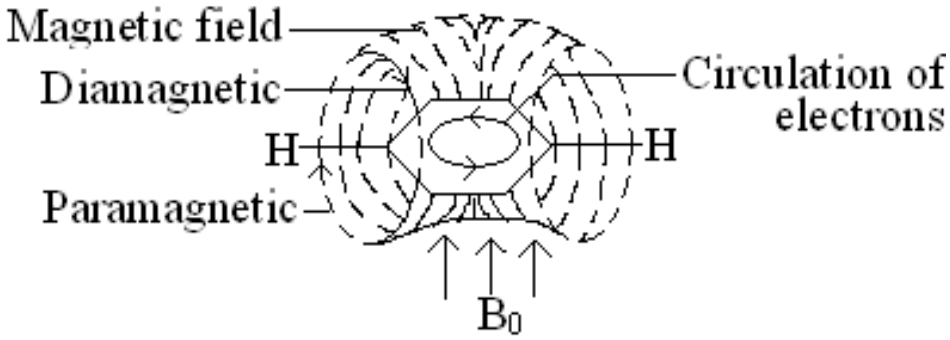
(d) Aromatic compounds

In the molecule of benzene (and aromatic compounds in general) π – electrons are delocalized cyclically over the aromatic ring. These loops of electrons are induced to circulate in the presence of the applied field B_0, producing a substantial electric current called ring current.

The induced field is diamagnetic (opposing B_0) in the centre of the ring, but the returning flux outside the ring is paramagnetic (augmenting B_0).

Protons around the periphery of the ring experience a magnetic field greater than B_0 and consequently come to resonance at lower values of B_0 (higher δ values).

In the molecule of toluene, the methyl protons resonate at δ 2.34, whereas a methyl group attached to an acyclic conjugated alkene appears at δ 1.95. This is some measure of the greater deshielding influence of the ring current in aromatic compounds (cyclically delocalized π – electrons) compared to the deshielding of conjugated alkene groups (having no cyclic delocalisation).



4. Hydrogen bonding

The hydrogen bonded proton, being attached to a higher electro – negative atom, will have a smaller electron density around it. So it is deshielded. So its δ value would be more than a proton which is not involved in hydrogen bonding.

Shielding and deshielding

Nuclei are essential parts of molecules held together by the electrons of the chemical bonds. The magnetic field at which a free or bare nucleus resonates is quite different from the field at which the same nucleus in a molecule resonates. It is thus clear that the electrons in a molecule affect the NMR frequency of a given nucleus in the molecule. The applied external magnetic field affects the motion of the electrons surrounding the nuclei there by, inducing local magnetic fields. This may slightly increase or decrease the magnetic field around the nucleus. So the field experienced by the nucleus is different from that applied to it. This effect of the extranuclear electrons on the strength of the applied magnetic field is known as magnetic shielding. The magnetic field acting on the nucleus is given by

$$H = H_0 (1 - \sigma)$$

Where H = The magnetic field experienced by the nucleus, H_0 = Applied magnetic field and σ = Shielding constant.

The shielding effect in such high electron density cases will therefore be larger, and a higher external field (B_0) will be needed for the if energy to excite the nuclear spin. Since silicon is less electronegative than carbon, the electron density about the methyl hydrogen in tetramethylsilane is expected to be greater than the electron density about the methyl hydrogen in neopentane (2,2 - dimethylpropane) , and the characteristic resonance signal from the silane derivatives indeed lie at a higher magnetic field. Such nuclei are said to be shielded.

Elements that are more electronegative than carbon should exert an opposite effect (reduce the electron density); such elements display lower field signals (they are deshielded). The deshielding effect of electron withdrawing group is roughly proportional to their electronegativity.

Spin – spin coupling

The interaction of the spin of a proton with that of another proton or protons attached to an adjoining carbon is called spin – spin coupling.

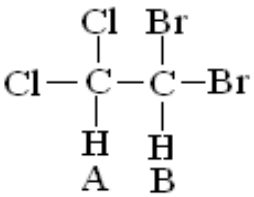
Rules governing splitting of NMR signals

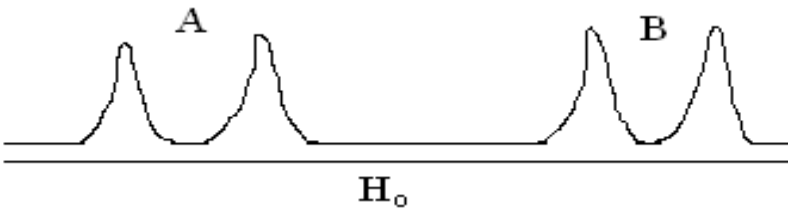
1. Equivalent nuclei do not interact with one another. So no splitting takes place because of them. For example, in ethanol the three protons in the methyl group give rise to splitting of the adjacent methylene protons but not to splitting among themselves.
2. If there are 'n' protons in the adjacent carbon atom, then, multiplicity is given by (n + 1). The multiplicity of NMR signal by adjacent methylene group protons is equal to 2 + 1 = 3. Similarly the mulitiplicity caused by methyl group protons is 3 + 1 = 4.
3. If the protons of atoms B are affected by protons on atoms A and C that are not equivalent the multiplicity of B is equal to $(n_A + 1)$ $(n_C + 1)$ where n_A and n_C are the numbers of equivalent protons on A and C respectively.
4. The relativite intensities of the triplet caused by methylene protons will be given by the coefficients of the terms of $(r + 1)^2$. i.e., $r^2 + 2r + 1$ i.e., 1: 2: 1. In general the relative intensities of the multiplets is given by the coefficients of the terms of $(r + 1)^n$ where n is the number of neighbouring protons, i.e., the intensities of these lines will be given by the $(n + 1)^{th}$ line of Pascal's triangle.

Example

1,1 – dibromo – 2, 2 – dichloroethane

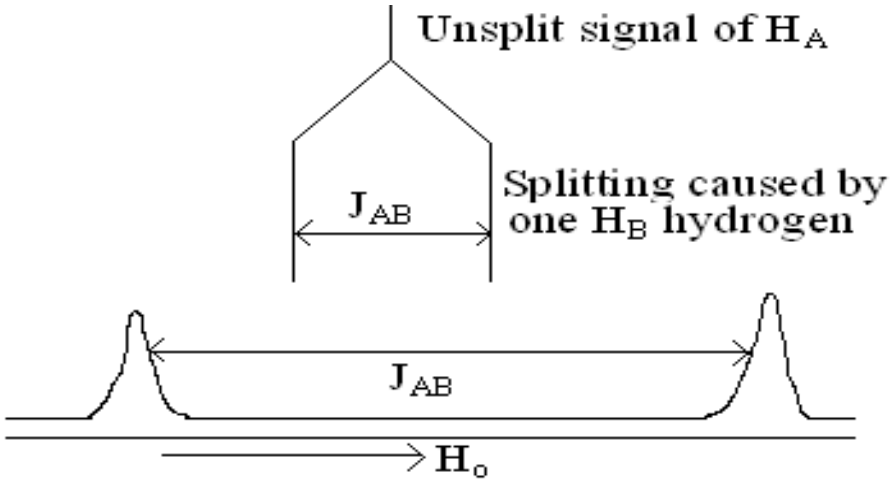
It is expected to exhibit two absorption lines due to the presence of two non – equivalent protons labeled A and B (in figure). But the actual spectrum consists of two well – defined doublets. The molecule belongs to AB system. For such a system the values for the hydrogens are of the same order.





The NMR Spectrum of 1, 1 – dibromo – 2, 2 - dichloroethane

The appearance of doublets can be rationalized: the hydrogen nucleus H_B has a spin of either +1/2 or – ½ which sets up a small magnetic field. The interaction of this small magnetic field with the external field can occur in two different ways. The magnetic field of H_B can be aligned with or opposed to the external field. If it is aligned with the external field, the adjacent non – equivalent hydrogen H_A is deshielded and the signal is shifted slightly downfield, resulting in a higher δ value. If it is opposed, the non – equivalent hydrogen H_A on the adjacent carbon atom is shielded from the effect of the external magnetic field and hence the signal is shifted slightly upfield, resulting in a lower δ value. Therefore, the single signal of hydrogen H_A is now split in to two separate signals of almost equal intensity. This is due to the fact that in half the molecules at a given applied magnetic field, the nuclear spin of H_B hydrogen is aligned with the applied magnetic field, and in the other half, the nuclear spin of H_B hydrogen is aligned against the applied magnetic field. This is pictorially represented in below figure.



The splitting of an AB system

Similarly, the hydrogen H_A splits the signal of H_B in to two signals. The distance between two split signals is proportional to the effectiveness of the interaction between the spins of these two hydrogen and is independent of the external field H_o. This separation between multiplet peaks is called coupling constant J and is always measured in Hz.

Coupling constant (J)

Definition

It is the distance between the centre of two adjacent peaks in a multiplet.

Unit

It is expressed in Hz or cps (cycles per second).

Explanation

We have seen that when high resolution NMR spectrometers are used, the NMR signals for a set of equivalent protons split in to several signals and that this splitting is due to spin – spin coupling. The distance between the centres of two such split, adjacent peaks is called coupling constant. It is denoted by J.

Characteristics

1. J gives a measure of the effectiveness of spin – spin coupling.
2. The value of J is independent of external field, i.e., for a particular compound at different radio frequencies, the distance between two adjacent peaks in a multiplet remains always a constant.
3. The J value of peaks in each multiplet is constant.

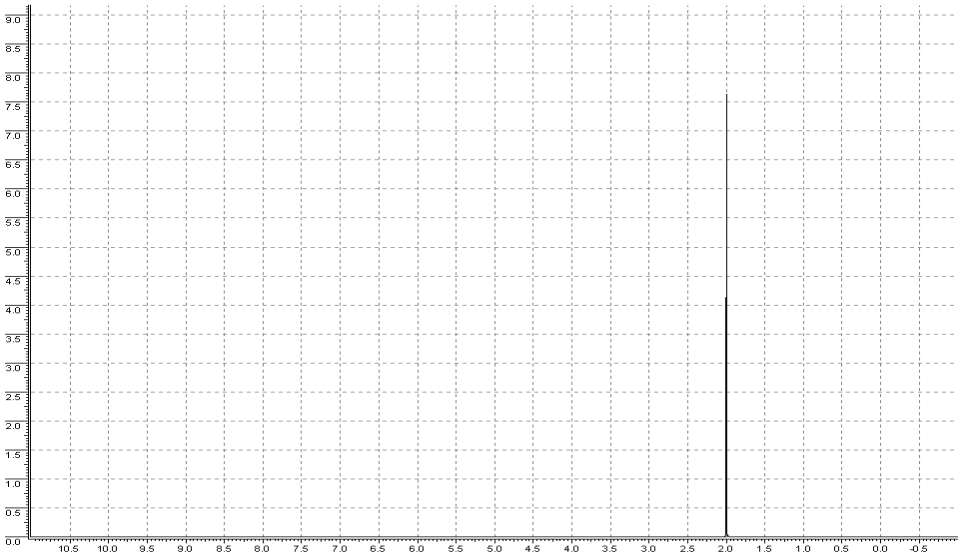
Example

Butanone – 2, has a J value of 7.3 cps.

Interpretation of NMR spectra of simple organic compounds

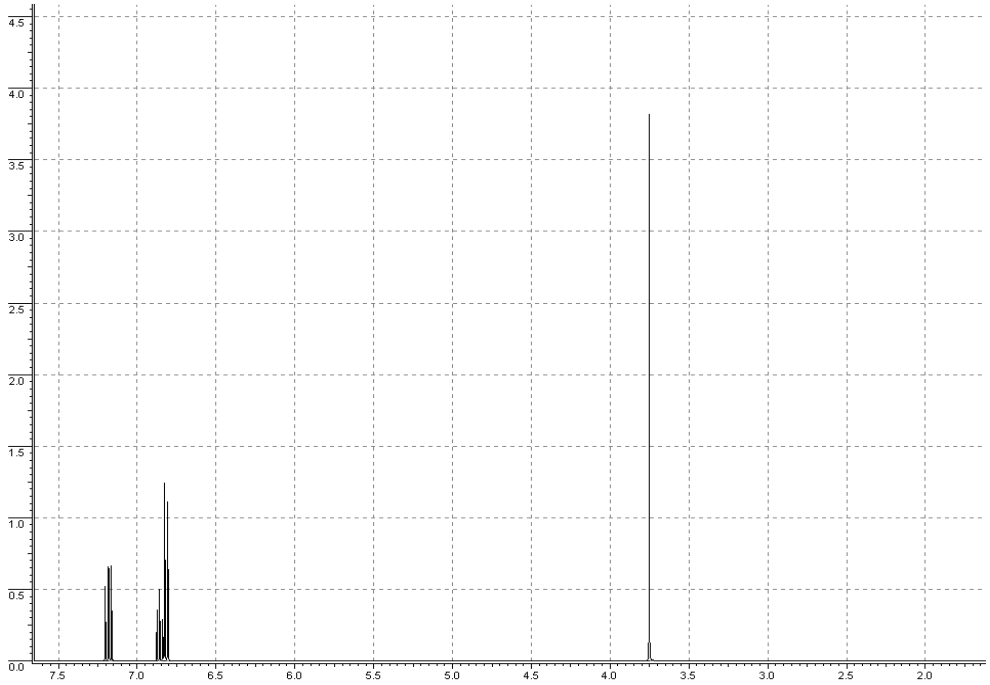
(i)

ACETONE



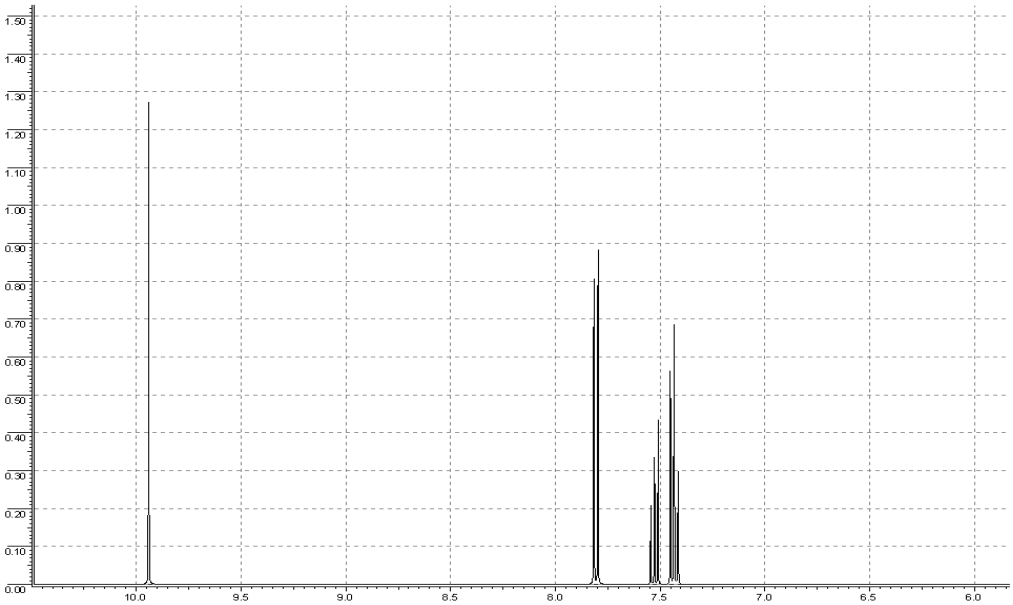
Acetone has two sets of chemically equivalent methyl protons (CH_3) and no spin- spin coupling. So it exhibit a sharp singlet peak at 2 ppm.

(ii) ANISOLE



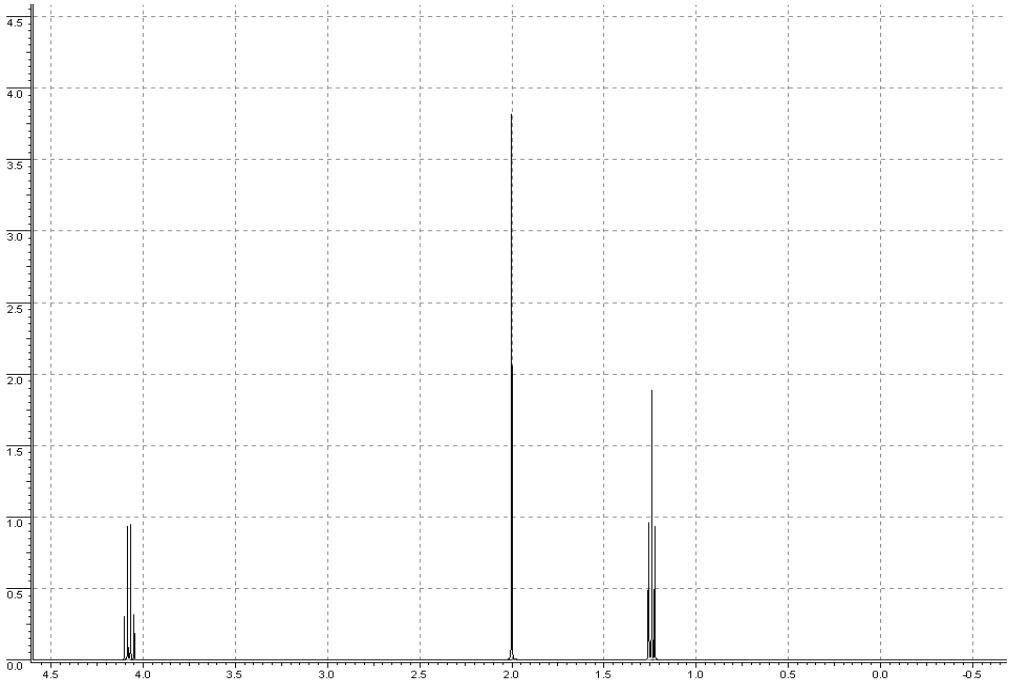
Anisole has three methoxy protons and six benzene ring protons. The methoxy protons have no coupling with neighbouring group. So they have a strong singlet sharp peak at 3.75 ppm. The benzene ring protons undergo coupling with neighbouring protons and exhibit two multiplets one at 6.75 and another at 7.25 ppm.

(iii) BENZALDEHYDE



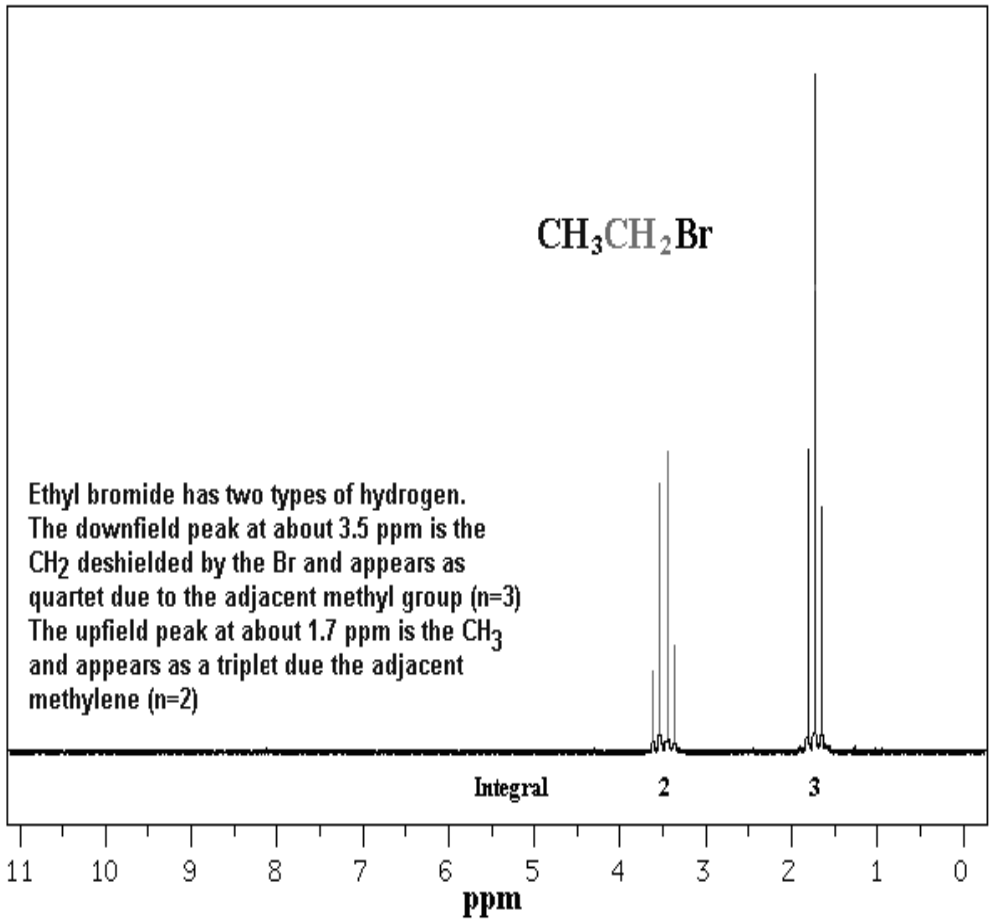
Benzaldehyde possess one aldehydic –CHO proton with no coupling which exhibits a strong sharp singlet peak at deep downfield region, 10 ppm. The benzene ring protons. The benzene ring protons undergo coupling with neighbouring protons and exhibit two multiplets one at 7.5 and another at 7.75 ppm.

(iv) ETHYL ACETATE



- Triplet at ~ 1.3 ppm ; relative intensity 3; assigned to CH_3 part of ethyl group, triplet splitting due to neighbouring CH_2.
- Singlet at ~ 1.9 ppm ; relative intensity 3; assigned to CH_3 part of acetyl (ethanoyl) group, no splitting as no hydrogens attached to adjacent carbon
- Quartet at ~ 4.1 ppm ; relative intensity 2; assigned to CH_2 part of ethyl group, quartet splitting due to neighbouring CH_3.

(v) ETHYL BROMIDE



(vi) ETHYLAMINE

c b a

$CH_3 – CH_2 – NH_2$

3 NMR signals

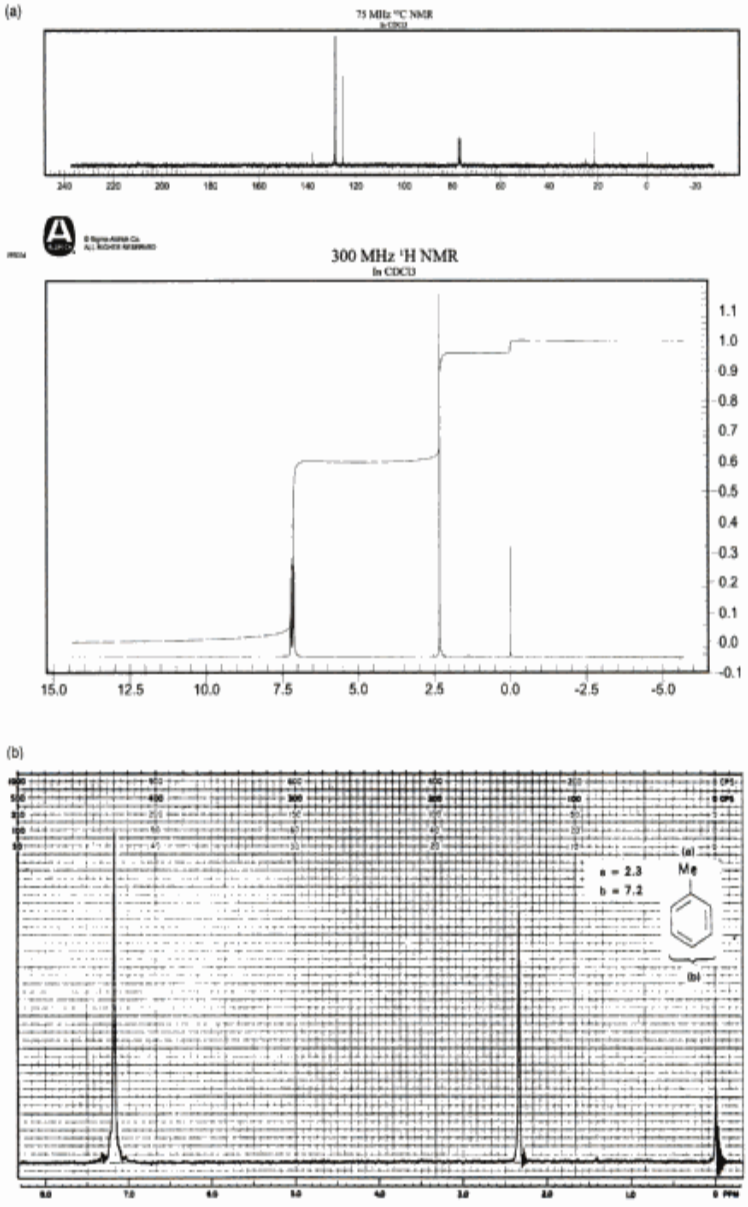


Figure 3.4 (a) The 300 MHz proton NMR spectrum of toluene (bottom) and the ^{13}C NMR spectrum of toluene (top). Reprinted with permission of Aldrich Chemical Company, Inc. (b) The 60 MHz proton NMR spectrum of toluene. The structure of toluene is shown, with Me indicating a methyl group, $-CH_3$. Two absorption peaks are seen, one due to the protons of the methyl group, and the other to the aromatic ring protons. The spectrum is discussed in Section 3.4. (From Bhacca et al., courtesy of Varian Associates, Palo Alto, CA, www.varianinc.com.)

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5.2. MASS SPECTROSCOPY

It is a technique for the identification of molecules by analyzing the positive ions produced by bombarding the molecules with electrons. The masses of the positive ions are recorded in the mass spectrometer.

Basic principles of mass spectrum

When a molecule is bombarded with rapidly moving electrons, positively charged ions are produced. These are separated on the basis of their m/e (mass/charge) ratios and then are identified. The separation of these ions is based on the fact that different ions with different m/e ratios move in different circular paths in a combined magnetic and electric field.

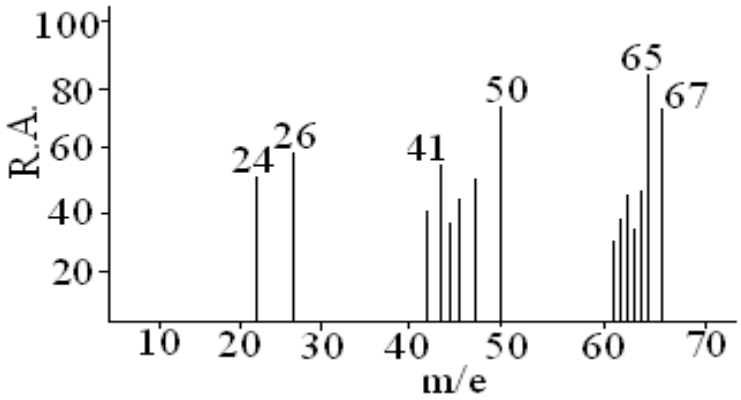
If a particle with mass m and with charge e is moving in a magnetic field with field strength H and in an electric field with a potential difference of V volts then

$$\frac{m}{e}=\frac{H^2r^2}{2V}$$

Where r is the radius of the circular path of the moving particle. Thus r depends on m as H, V and e are constants. So ions with different masses will move in different circular paths and therefore can be separated and identified.

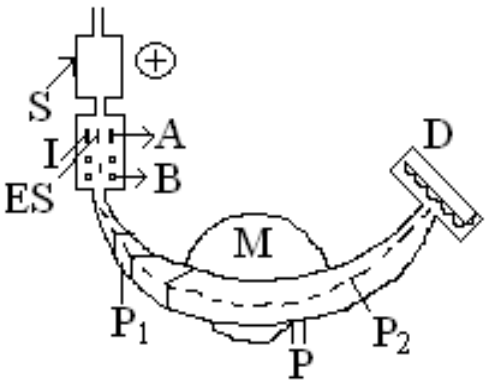
The mass spectrum is a record of the numbers of different kinds of ions. The relative abundances of ions are characteristics for every compound including isomers. It provides information regarding the molecular structure of compounds.

The mass spectrum is generally represented by a bar graph. Here the intensity on the ordinate is plotted against m/e in the abscissa. Recently the relative abundance (R.A.) is plotted against rate – m/s as shown in figure.



Instrumentation

A simple mass spectrometer is as shown in the figure.



S – Sample; I – Ionisation of molecules; ES – Electrons source; A – Anode to attract electrons; B – Acceleration of molecules; M – Magnet; P – To pump; D – Detectors; P_1 - Particles hitting side; P_2 – Particles reaching detectors

1. The inlet system

Here the sample is converted to the gaseous state. For this purpose the system is heated up to 400^{o}C.

2. The ion source (or ionization chamber)

Here a beam of electrons is sent across the sample molecules. The molecules become ionized. These are further broken in to fragments. Ionization in vapourised material may also be produced by irradiation with ultraviolet light.

3. Accelerating cell

The ionized particles from the ionization chamber are accelerated by the accelerating voltage V. The voltage is varied in such a way to allow the fragments in to a magnetic field at a range of velocities.

4. The ion separator (Analyser)

Here the charged particles describe a circular path under the influence of magnetic field.

5. Detector (The ion collector)

Generally the detector used is a photographic plate. Electron multipliers may be used as detectors.

6. Vaccum system

A high vaccum is to be maintained in the instrument with the help of pump.

Molecular ion peak

When the sample is bombarded with electrons of energies of 9 to 15 eV, the molecular ion is produced with the loss of an electron. The peak due to this ion is called molecular ion peak or parent peak.

$$M + e^- \rightarrow M^+ + 2e^-$$

(Molecule) (Molecular ion)

Usually the molecular ion peak is the peak of highest mass number. Highly reactive compounds do not form a stable parent peak. Even if it is present, its intensity is weak. This is because the bonds in the parent ion are readily broken forming fragmented ions. However thermodynamically stable molecules like benzene give an intense parent peak. On the other hand unstable molecules like t – butyl alcohol do not give intense parent peak. thus the ions of highest mass may not be the parent peak.

So, one must be careful enough to identify the molecular ion peak in a mass spectrum.

Uses

This molecular ion is important since its m/e value gives the molecular weight from which the molecular formula can be inferred.

Base peak

The largest peak in the mass spectrum, corresponding to the most abundant ion is known as the base peak. The base peak may be a parent peak or some other fragmented ion peak depending upon the nature of the compound.

Uses

Base peak gives us an idea about which ion is abundant when the molecule under study is bombarded with electrons. This helps us to identify the molecular structure.

Meta – stable peak

In a mass spectrum we can see some broad peaks at non integer m/e. These peaks are called Meta – stable peaks. The ions producing these peaks are called Meta – stable ions. These arise from fragmentation that take place during the flow down the ion tube rather than in the transition chamber.

A meta – stable ion is formed as follows:

Original ion → Daughter ion + neutral fragment.

The meta – stable peak will appear as a weak, diffuse peak usually at a non – integral mass given by

$$m^* = \frac{(\text{Mass of daughter ion})^2}{\text{Mass of original ion}}$$

Uses

They indicate one – step decomposition process. i.e., the parent ion undergoes decomposition in one step to the daughter. Thus investigation of meta – stable ions gives us a mechanistic picture of fragmentation process. From the position of these peaks we can identify the parent and normal daughter ions.

Isotopic peak

The molecular ion peak is always accompanied by satellite peaks at (M + 1) and at (M + 2), where M is the molecular weight of the substance under study. These peaks are due to the presence of isotopes of elements present in the molecule. These peaks are called isotopic peaks.

Each of C, N and H has a principal heavier isotope with one mass unit greater than the most common isotope. Therefore the presence of these elements in a compound will give rise to a small isotopic peak at $(M+1)^+$. i.e., one mass unit greater than the molecular ion. Thus the ion CH_3^+ for example will show a peak at m/e 15 corresponding to molecular ion peak and another at m/e 16 which is due to $^{13}CH_3^+$.

The elements showing (M + 1) peaks are C, H and N. The element showing (M + 2) peaks are O, S, Cl, Br etc.

Uses

Isotopic peaks are useful for determining the molecular formula of a compound. From knowledge of the relative natural abundance of isotopes, we can theoretically calculate the relative intensity of an isotopic peak expected for a formula. If this agrees with that of the experimental value, the assumed molecular formula can be taken as the correct one.

Example

Element	Isotopes	Relative abundance of isotopes	Relative intensity of isotopic peaks
Cl	Cl^{35}, Cl^{37}	77 : 23	3 :1
Br	Br^{79}, Br^{81}	50.5 : 49.5	1 :1
S	S^{32}, S^{34}	96 : 4	24 : 1

Nitrogen rule

Rule

All organic compounds with even molecular weights will have either no nitrogen atoms or even number of nitrogen atoms. All organic compounds with odd molecular weights will have an odd number of nitrogen atoms.

This rule is applicable to all covalent compounds containing carbon, hydrogen, oxygen, sulphur, halogens, phosphorous and boron, besides nitrogen.

Uses

This rule provides information concerning possible formulae of compounds whose molecular weights have been determined.

Ring rule

Rule

The sum total of the number of rings and double bonds present in a molecule can be determined by the formula.

$$R = ½ (2w – x + y + 2)$$

Uses

After deciding the exact molecular formula we can use the above rule to fix up the number of rings and double bonds in the molecule.

If the molecular formula is $C_wH_xO_zN_y$

Then $R = ½ (2w – x + y + 2)$

In benzene the molecular formula is C_6H_6

w = 6: x = 6

$$R = ½ (12 – 6 + 2) = ½ (8) = 4$$

R is sometimes referred to as double bond equivalent (DBE). If DBE = R = 1 then it means that either one double bond or 1 ring is present in the molecule.

R = DBE = 4 suggests the presence of an aromatic ring (one ring with three double bonds).

Fragmentation

Definition

When molecules are bombarded with a beam of energetic electrons, they ionize and are broken up in to many fragments. This is called fragmentation.

The relative abundance of fragmented ions formed depends upon

(i) The stability of the ion

(ii) The stability of the radical lost.

Factors like resonance, inductive effect, polarisability etc., contribute to the stability of the fragmented ions.

Important modes of fragmentation

This mode of fragmentation involves hemolytic or heterolytic cleavage of a single bond.

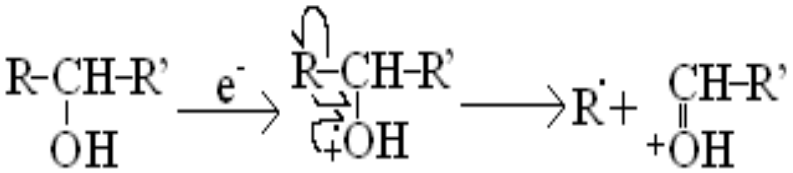
1 Homolytic cleavage

It is initiated by a radical site. These are very common. They are classified as follows:

(i) Mode I

Compounds containing a hetero – atom bonded to a carbon atom with a single bond undergo such cleavage. The parent ion is formed by the removal of one electron from the hetero – atom. A new bond is formed with the adjacent atom. A radical is loat.

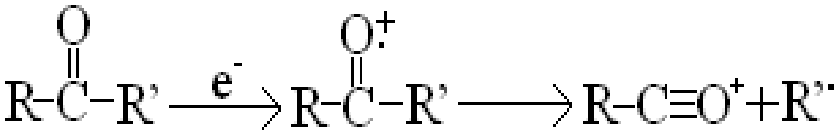
Example



(ii) Mode II

When a hetero atom is attached to a carbon atom by a double bond, α – cleavage takes place.

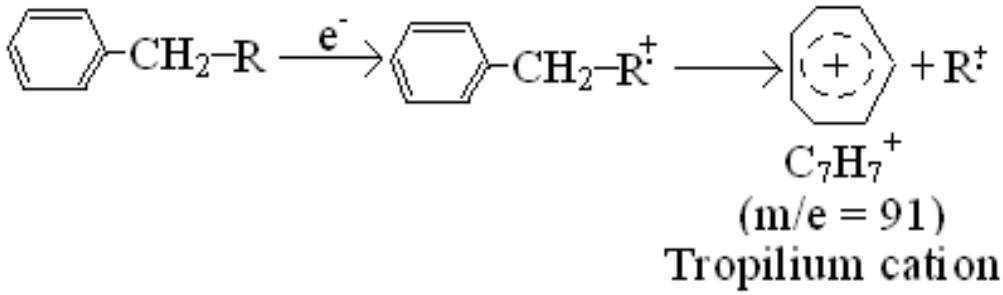
Example



(iii) Mode III

Benzylic cleavage is another mode of fragmentation. This is energetically preferred. Here a C – C bond β to an aromatic ring is cleaved.

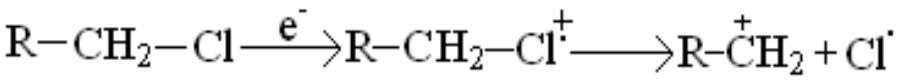
Example



2. Heterolytic cleavage

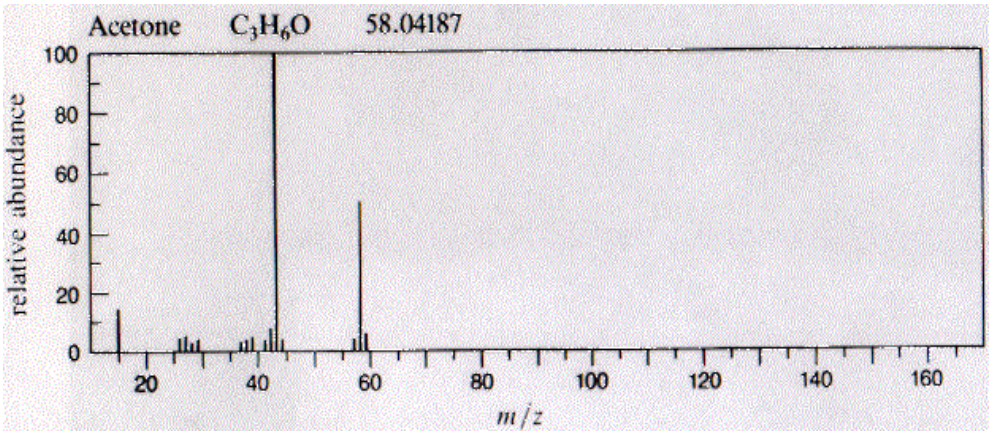
Cleavage of C – O, C – N, C – S and C – Cl bonds is more difficult than that of C – C bonds. In such cleavage the positive charge is carried by the carbon atom.

Example

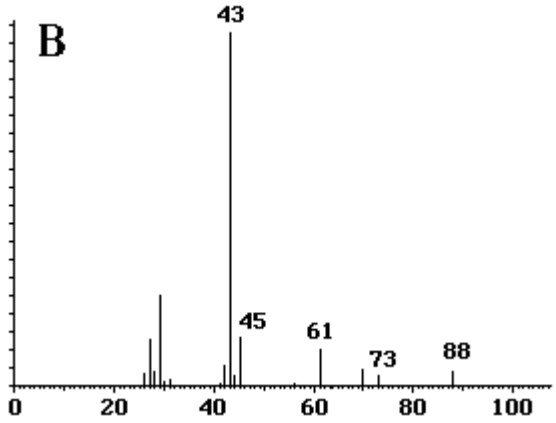


Interpretation of mass specta of simple organic compounds

(i) Acetone

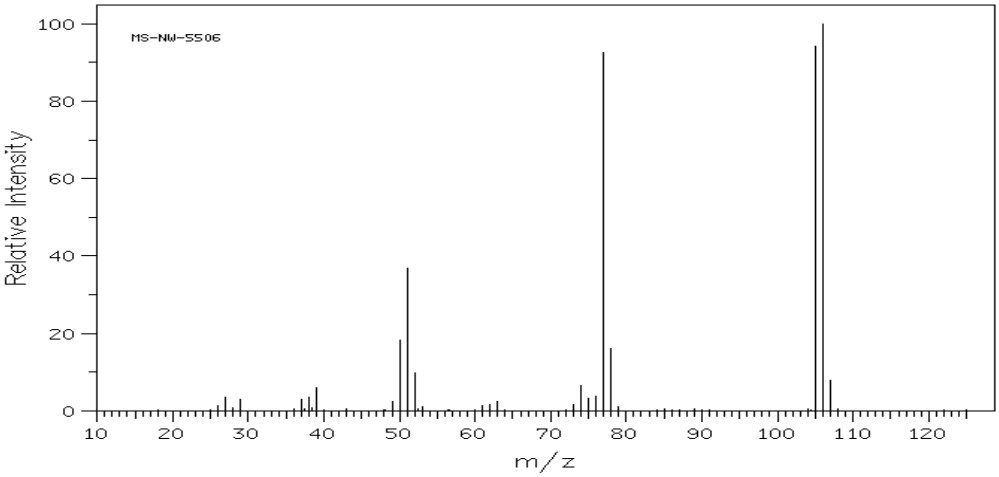


(ii) Ethyl acetate



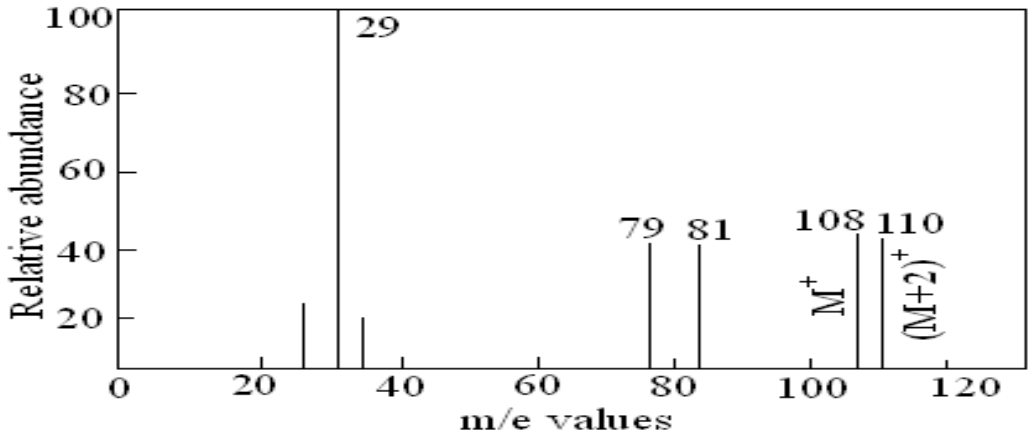
Peaks at *m/e* 88 (M^+); 73 ($(M\text{-}CH_3)^+$); 43 ($(M\text{-}OCH_2CH_3)^+$);

(iii) Benzaldehyde

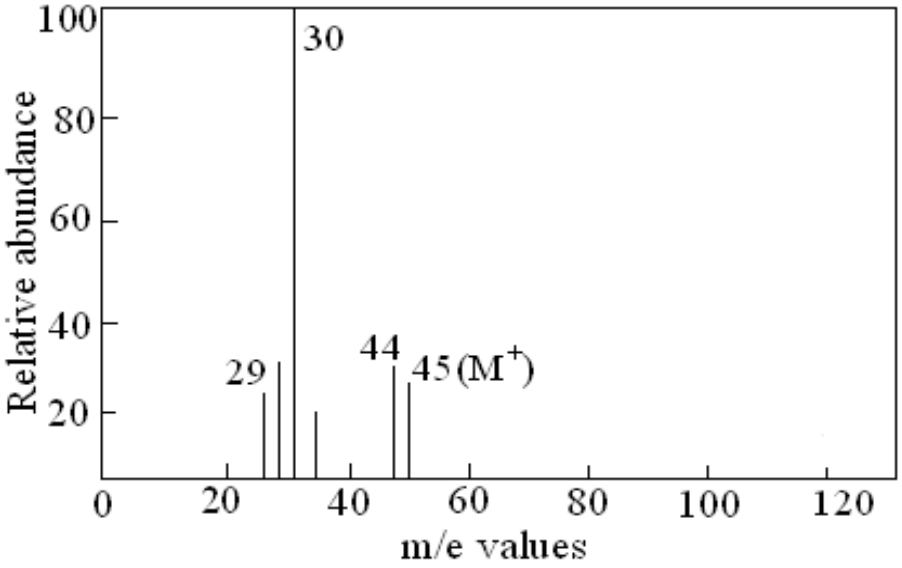


(iv) Ethyl bromide

The $C_2H_5^+$ ion (m/e 29) is the base peak (100 % abundance) and formed due to the removal of bromine. The peak at m/e 108 is the molecular ion peak and the peak at m/e 110 is the $(M+2)^+$ ion peak. These two peaks are of equal intensity. The peaks at m/e 79 and at m/e 81 are the isotope peaks.



(v) Ethyl amine



$$CH_3 - CH_2 - \dot{N}H_2 \longrightarrow \dot{C}H_3 + CH_2 = NH_2$$

(m^+) m/e 45 (Base peak) m/e 30

$$\xrightarrow{-H} CH_3 - CH_2 = N^+H$$

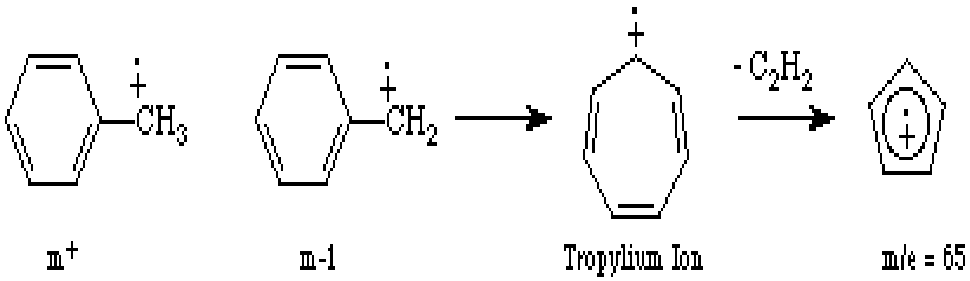
(m+ -1) m/e 44

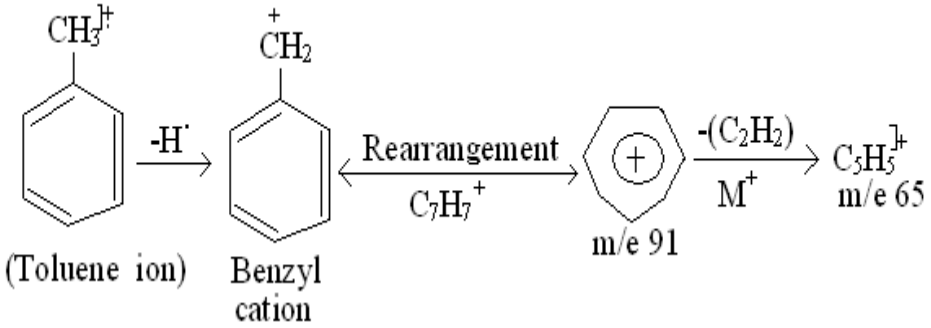
(vi) Toluene

Two strong peaks at m/e 91 and at m/e 65 are formed. The peak at m/e 91 is due to the formation of tropylium cation (stable) which loses a molecule of acetylene (26 mass units) to give $C_5H_5^+$ (m/e 65).

The mass spectrum of toluene (methyl benzene) is shown below. The spectrum displays a strong molecular ion at m/e = 92, small m+1 and m+2 peaks, a base peak at m/e = 91 and an assortment of minor peaks m/e = 65 and below.

The molecular ion, again, represents loss of an electron and the peaks above the molecular ion are due to isotopic abundance. The base peak in toluene is due to loss of a hydrogen atom to form the relatively stable benzyl cation. This is thought to undergo rearrangement to form the very stable tropylium cation, and this strong peak at m/e = 91 is a hallmark of compounds containing a benzyl unit. The minor peak at m/e = 65 represents loss of neutral acetylene from the tropylium ion and the minor peaks below this arise from more complex fragmentation.

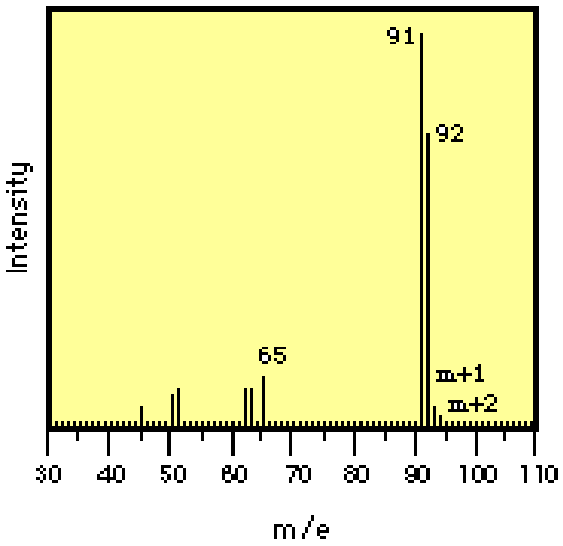


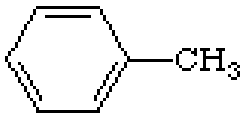


Suppose the transition $C_7H_7^+$ (91) to $C_5H_5^+$ (65) occurs in the second field free region, then a metastable peak is formed. The position of the broad metastable peak is determined as:

$$m^* = \frac{m^2_1}{M_1} = \frac{65 \text{ X } 65}{91} = 46.4$$

A metastable peak in case of toluene appears at 46.4 in the mass spectrum.





Toluene

Important questions

1. What are the principles of nuclear magnetic resonance?
2. Explain chemical shift
3. How will you determine the number of NMR signals for a compound?
4. What are the factors affecting chemical shift?
5. Why TMS is taken as standard in NMR spectroscopy?
6. Write notes on shielding and deshielding in NMR spectroscopy
7. Write notes on spin-spin coupling
8. What are the factors affecting splitting constant?
9. What are the basic principles of Mass spectroscopy?
10. Discuss the instrumentation of mass spectroscopy
11. How will you interpret a mass spectrum?
12. What are molecular ion peak and metastable peak?
13. Explain the NMR spectra of toluene, anisole and benzaldehyde
14. Explain the mass spectrum of toluene, ethylamine and acetone
15. Write notes on base peak and isotopic peak.
16. Explain nitrogen rule and ring rule

NOTES

NOTES