CHAPTER # 13
SPECTROSCOPY

www.pakcity.org
EDUCATION
pakcity.org

“Spectroscopy is the study of interaction of electromagnetic radiation of light with matter".

Spectroscopy has a wide scope of applications in various fields of science and technology and significantly favored over conventional methods because;

- It is easier and takes very little time to analyze a sample.
- It requires a very small amount of substance which is to be analyzed.
- It provides more reliable information about chemical molecule.

Some applications of spectroscopy are given as:

(i) Analysis of chemical comnpounds
(i) Quality control of drugs syntheses
(iii) Analysis of water pollution
(iv) Determination of protein structure
(v) Analysis of forensic materials
(vi) It helps in structure analysis

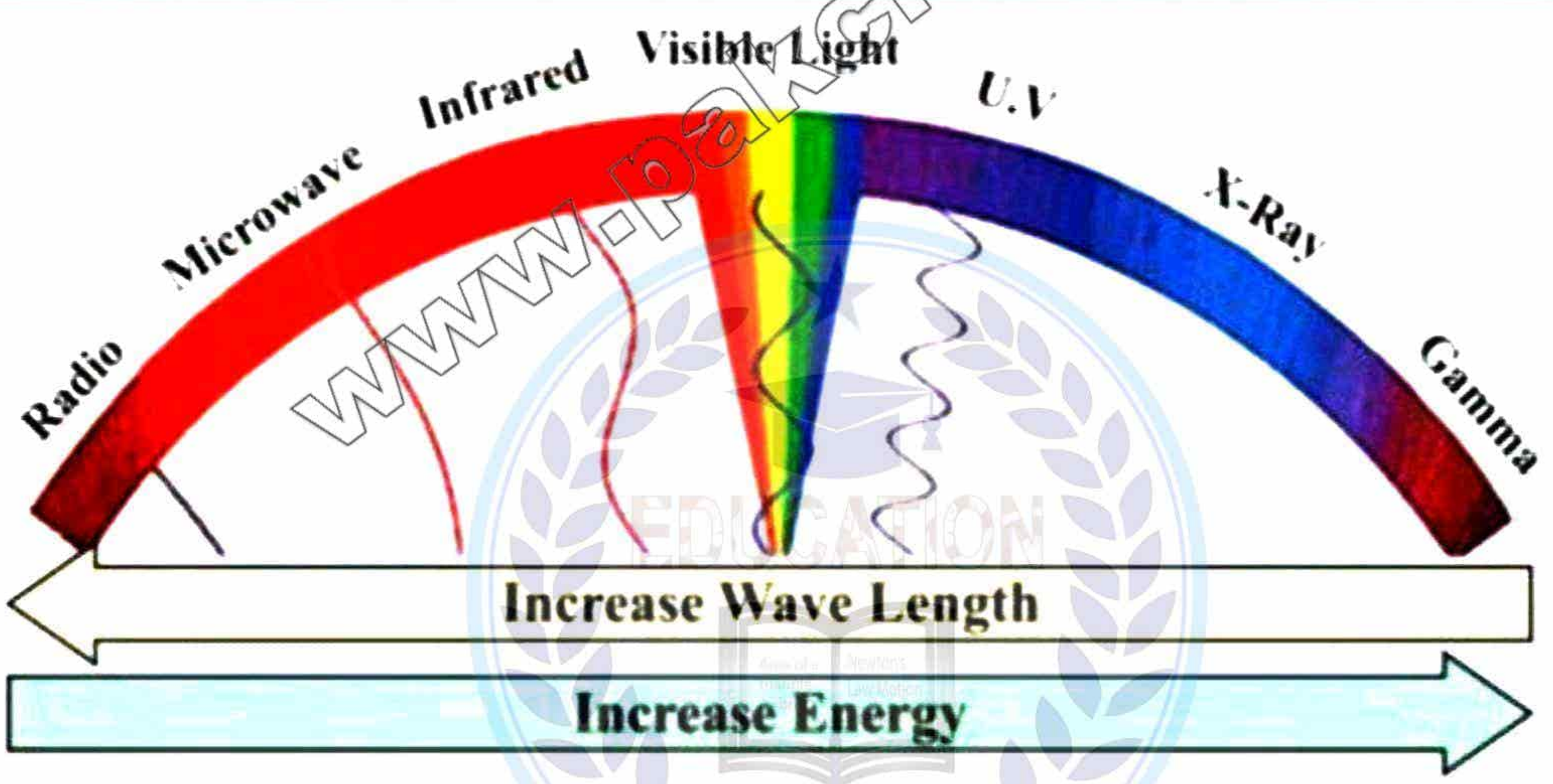


Fig 13.1: Electromagnetic spectrum from low-energy radiowaves to high energy gamma rays

METHODS OF SPECTROSCOPY

Sunlight consists of a wide range of electromagnetic waves including radio waves, microwaves, infrared radiations, visible radiations, ultra-violet radiations, etc. When electromagnetic radiations interact with molecule, some of the rays are absorbed by the molecules while other are transmitted. The wave length and frequency of absorbed light can provide valuable information about the structure of molecule. Various methods of spectroscopy are commonly used to analyze and characterize the compounds.

13.1.1 Infrared (IR) Spectroscopy

"Infrared spectroscopy is used to detect the type of bonds and the functional groups present in molecule".

I.R frequency is expressed in the unit of wave number (cm^{-1}). The most useful IR range lies between 4000-670 cm^{-1}.

Applications of IR Spectroscopy

(i) I.R spectroscopy provides information for the presence of different functional groups in the organic molecules.

(ii) I.R spectroscopy is also useful for identifying the impurities present in the sample to be analyzed.

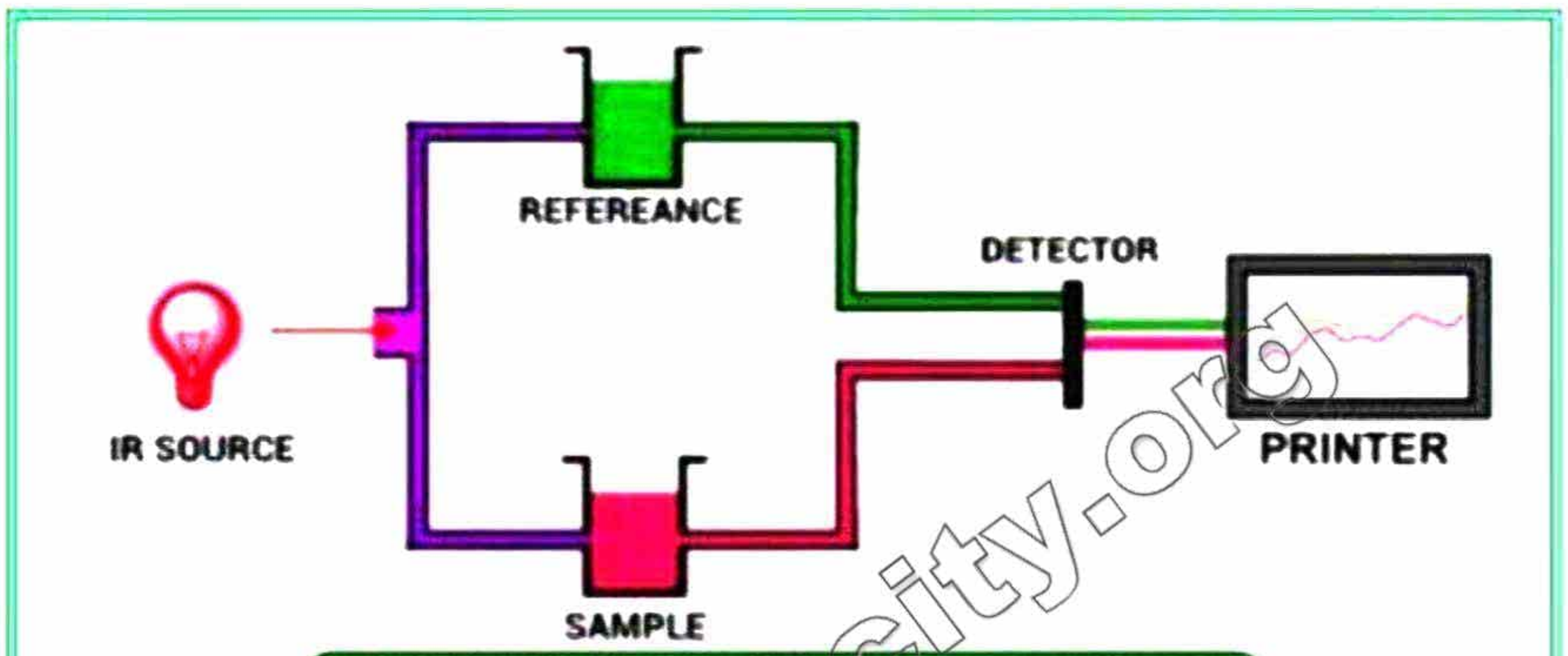


Fig 13.4:Block diagram of Infrared spectrophotometer

www.pakcity.org

Ultra-violet/Visible (UV-Vis) Spectroscopy

"Ultra-violet-visible spectroscopy is used to determine the presence of double and triple bonds as well as conjugated system in the molecule".

The U.V region of electromagnetic spectrum extends from 200nm to 400nm and the visible region extends from 400nm to 800nm.

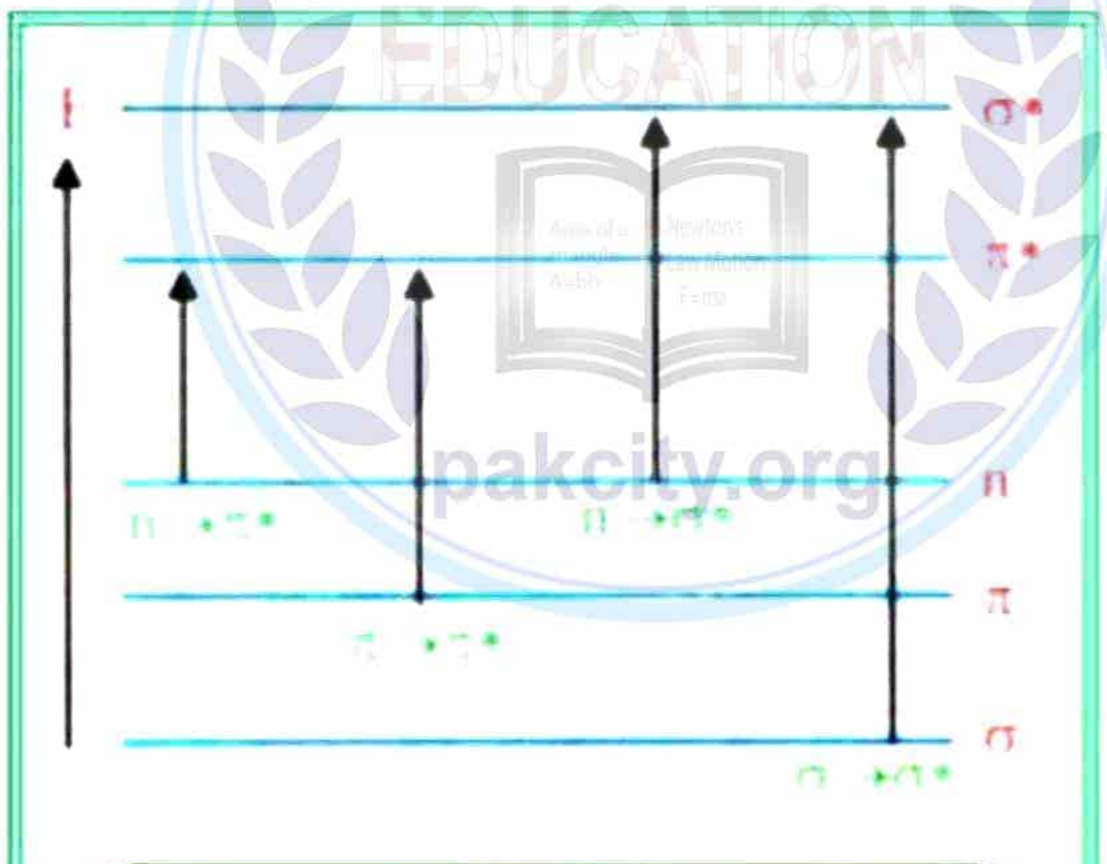


Fig 13.5:
Electron transition in organic compound due to interaction with U.V-visible radiations

When a molecule absorbs electromagnetic radiations of U.V- visible range (200nm-800nm) electronic transitions Occur. Its electrons are promoted from lower energy level to higher energy level. The transition of electrons may be of the following types.

(i) σ – σ* transition:

It requires very high energy which is beyond the range of U.V-visible radiations therefore carbon-carbon single bond do not absorb U.V- visible radiations and cannot be detected.

(ii) π – π* transition:

This transition occurs in the molecule that contain double or triple bonds or aromatic rings. The range of wavelength for the absorption of T-* transition is 180-320 nm.

(ii) n – π* transition:

This transition is associated with the molecules in which double or triple bond is connected with a hetero atom (N, 0, S) for example aldehyde and ketone, etc. The range of wavelength of n-* transition can vary depending upon the specific molecular system, however it is approximately 200- 500nm.

(iv) n – σ* transition:

This transition is concerned with the saturated molecule with hetero atoms like alkyl halide, alcohol etc. The range of n-g* transition roughly falls around 150-300nm.

Application of U.V-visible spectroscopy

(i) U.V spectroscopy can provide valuable information regarding the structure of a compound particularly in relation to the presence of double bond, triple bond, aromatic system and hetero atoms.

(ii) U.V spectroscopy is extensively used for determining the concentration of unknown compounds in a solution by using Beer-Lambert's law.

Nuclear Magnetic Resonance (NMR) Spectroscopy

The nucleus of certain elements like 'H (proton) exhibits random spin and behave like a tiny magnet due to their charged nature. When an external magnetic field is applied, the spin of nucleus aligned in two ways.

(ii) It can align in the same direction of the applied magnetic field and it is said to be low energy spin state.

(i) It can be in the opposite direction of the applied magnetic field and said to be high energy spin state.

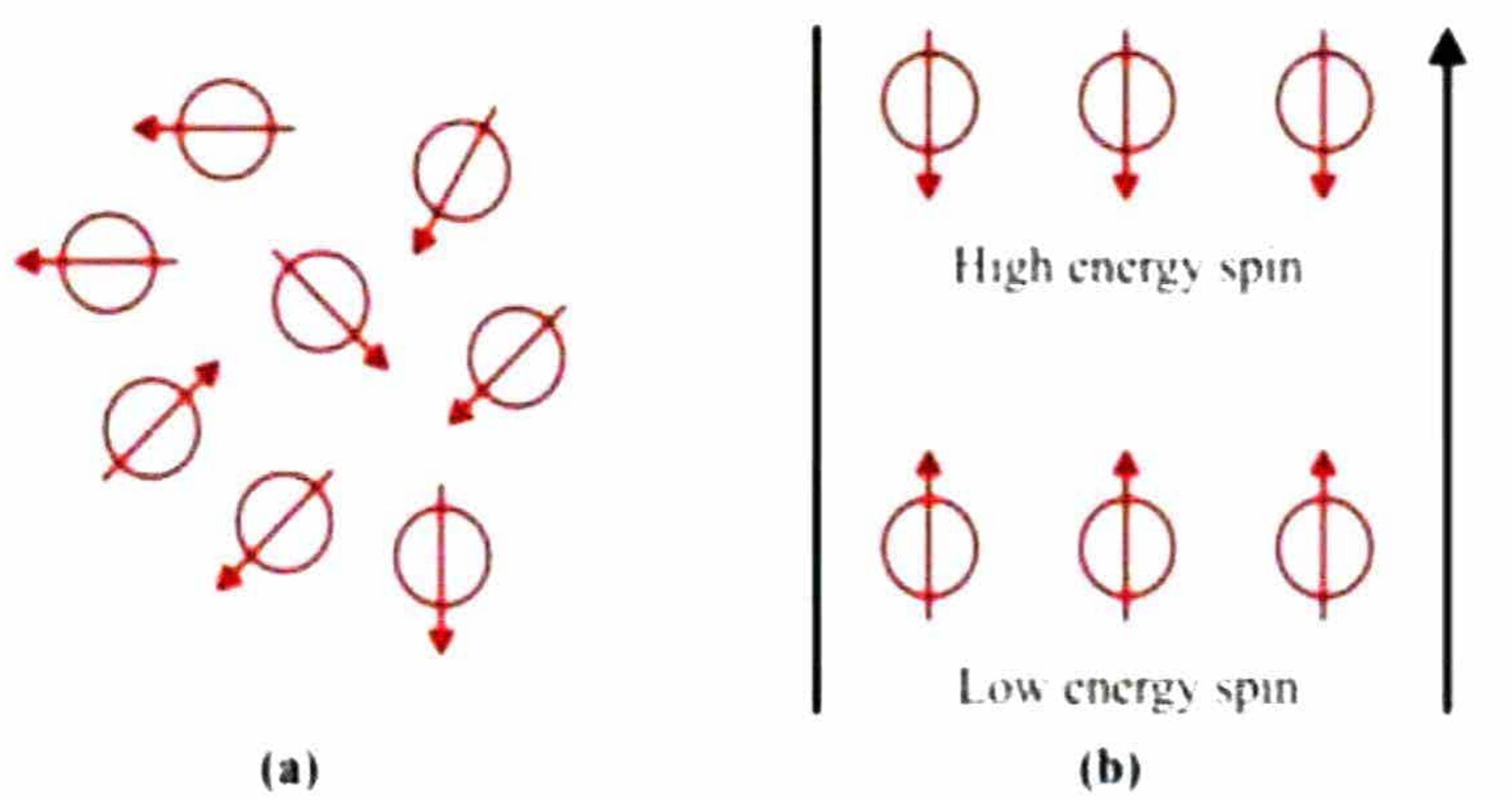


Fig 13.7: Spinning of proton (^1H) (a) in the absence of magnetic field (b) in the presence of magnetic field

When a sample of a compound is placed in a strong magnetic field and subjected to radio frequency radiation, the nuclei with specific spin state absorbs energy and flip into a high energy state, this absorption of energy is detected as signals in the NMR spectrum. The solvent used in NMR spectroscopy is usually D_20 or DMSO, since it does not interfere with NMR of the sample but with the conditions that compound is soluble in both solvents.

The graph of ^1H NMR consists of following parameters.

(i) X-axis represents chemical shift which shows position of proton signals relative to TMS (tetramethylsilane).

(i) y-axis represents absorption which shows shift. the intensity of NMR-signals.

(iii) Peaks represent splitting pattern (singlet, doublet, triplet, quartet) due to neighboring protons.

To understand ^1H NMR spectrum let us consider the example of ethanol. H NMR spectrum of ethanol has three types of protons.

- Methyl (CH_3) protons appear as triplet at around 1.1 to 1.3 ppm.
- Methylene protons (CH_2) appear as a quartet around 3.5 to 4 ppm.
- The hydroxyl proton appears as a broad singlet around 4 to 5 ppm.

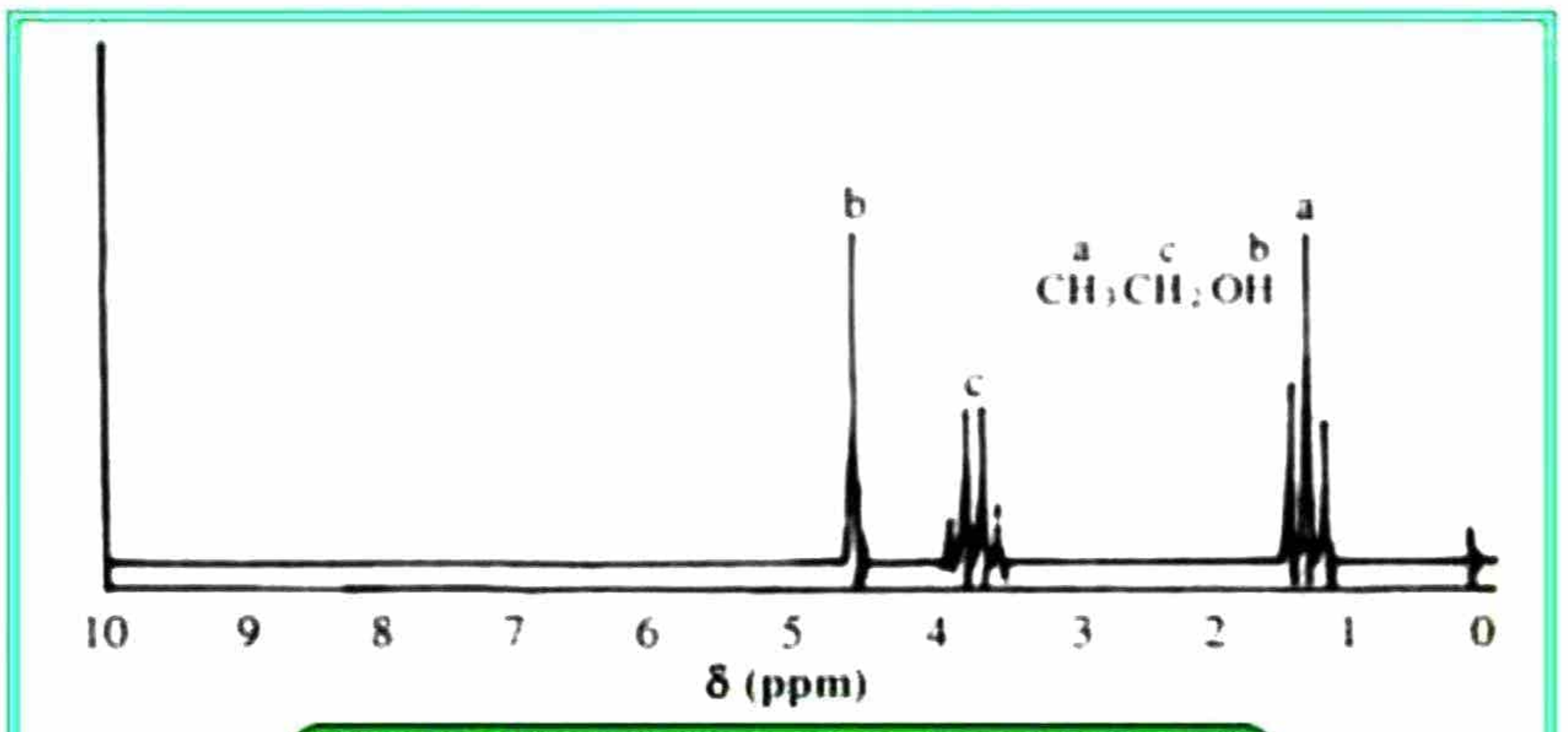


Fig 13.8: ^{1}H NMR spectrum of ethanol (C_2H_5OH)

Applications of NMR Spectroscopy

NMR spectroscopy is a powerful analytical technique, it provides valuable information about the chemical structure of organic compounds.
Each organic compound exhibits a unique NMR spectrum acting as a "fingerprint" that allows chemists to identify them. This technique is successfully applied in drug analysis, material science, forensic analysis and many other fields.

Atomic Absorption and Emission Spectroscopy

Atomic absorption and emission spectroscopy are established techniques used to identify elements in various samples including metal compounds. Within an atom, electrons are distributed in different energy levels, when atom receive energy from an external source like heat or an electric discharge, electrons can be promoted to higher energy level. These excited electrons than undergo transition involving the absorption or emission of electromagnetic radiations.

Atomic absorption spectroscopy

In atomic absorption spectroscopy, the sample is exposed to a wide range of light, the atom selectively absorbs specific wavelength of light that align with the energy needed to elevate electrons to higher energy level. The absorbed wavelengths of light appear as dark lines in a unique pattern specific for that element. By examining the absorbed wavelength, a chemist can identify the presence of specific element in the given sample.

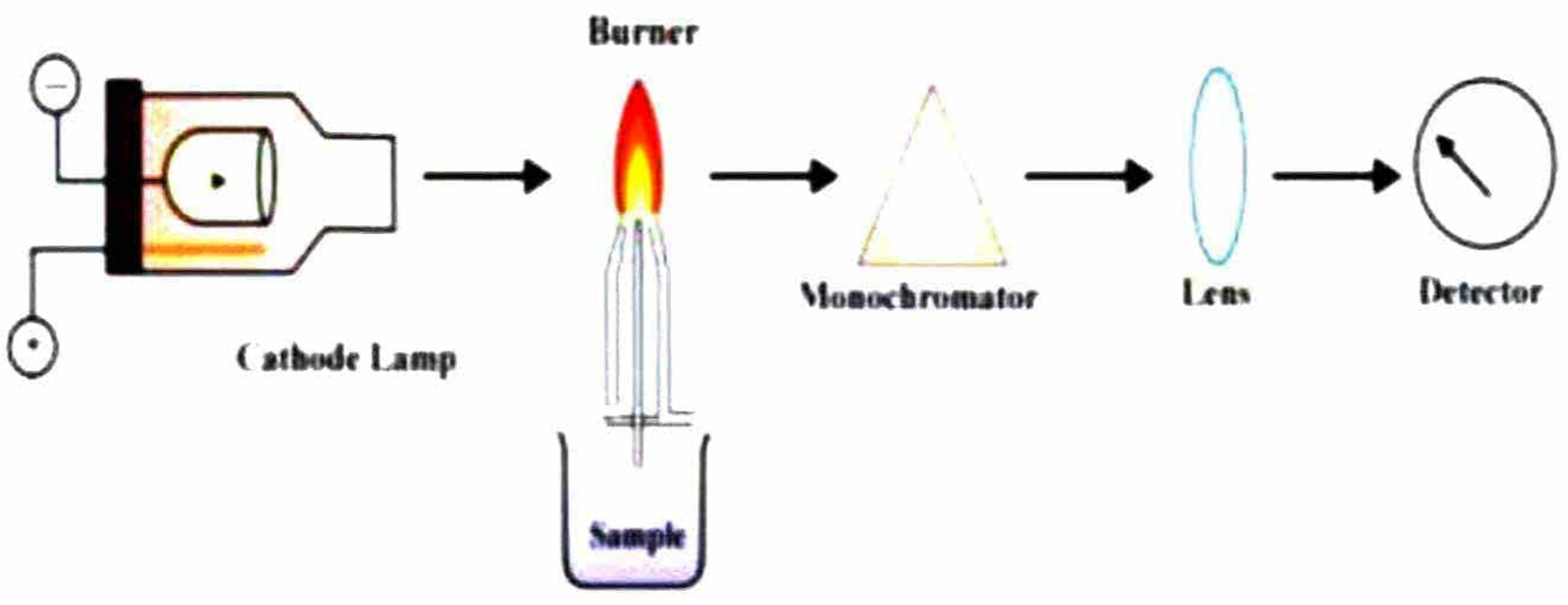


Fig 13.9: Atomic absorption spectrophotometer

In atomic emission
spectroscopy the electron in an atom is first excited by providing energy from external source such as heat or electrical energy. When the excited electrons return to their ground state, they emit excess energy in the form of light of specific wavelengths.
This emitted light commonly found U.V visible or rarely in IR range. This emitted light appears as a series of bright lines against a dark background. Since each element has its distinct set of bright lines, chemist can identify the element based on this information.

Mass Spectrometry
"Mass spectrometry is technique used to determine the mass to charge ratio (m/z) of ions in a sample".

It provides information about the mass of different fragments of the molecule and play an important role in the structure elucidation of molecule.

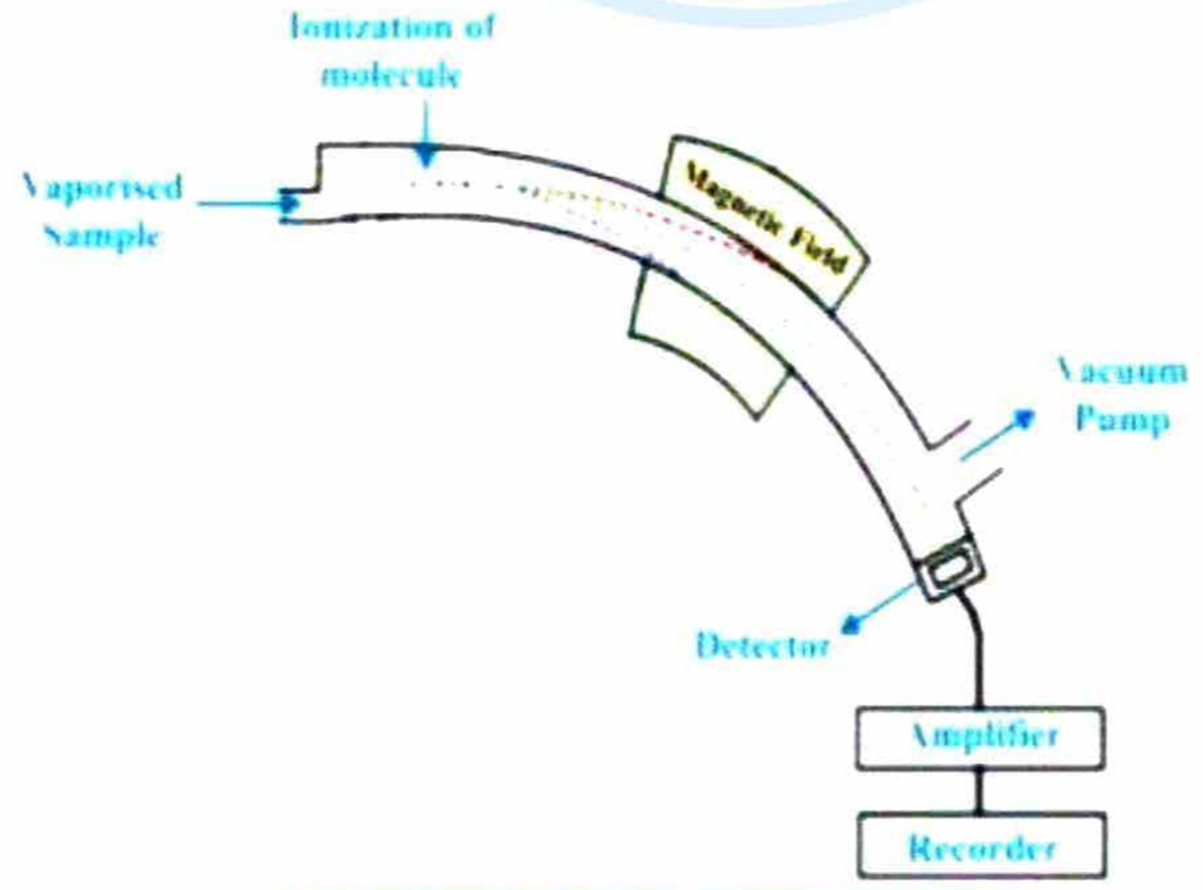


Fig 13.13: Mass spectrometer

Applications of mass spectroscopy

(i) It is used to determine the molecular mass of unknown compounds on the basis of mass to charge ratio.

(i) It is used for the identification and purification of drugs and other pharmaceutical products.

Short Questions

1. What types of nuclei are detected in proton NMR spectroscopy?

Proton NMR (^1H NMR) spectroscopy specifically detects the nuclei of hydrogen atoms (protons). This is because the technique relies on the magnetic properties of the proton and its interaction with an applied magnetic field. Other nuclei with magnetic moments, like ^{13}C (carbon-13) or ^{19}F (fluorine-19), can be detected using different NMR spectroscopy techniques with appropriate frequencies.

2. Name the components which represents x-axis and y-axis of a proton NMR spectrum.

In a proton NMR spectrum:

- **X-axis (chemical shift, δ):** This axis represents the resonance frequency of the protons relative to a reference standard (usually tetramethylsilane, TMS). The chemical shift is measured in ppm (parts per million) and reflects the electronic environment surrounding each proton. Protons in different chemical environments experience slightly different magnetic fields due to shielding effects from neighboring electrons, leading to variations in their resonance frequencies.
- **Y-axis (intensity):** This axis represents the integrated signal intensity, which is proportional to the number of protons giving rise to that particular resonance peak. The integration allows you to determine the relative abundance of protons in different environments within the molecule.

3. Differentiate between atomic absorption and emission spectroscopy.

- **Atomic Absorption Spectroscopy (AAS):**
    - In AAS, the sample is vaporized using high temperatures.
    - Ground state atoms in the vapor absorb specific wavelengths of light from a source element (same element being analyzed).
    - The amount of light absorbed is measured and is related to the concentration of the element in the sample.
    - AAS is a quantitative technique used to determine the concentration of specific elements in a sample.
- **Emission Spectroscopy:**

- In emission spectroscopy, the sample is excited using an energy source (heat, electricity, etc.).
- Excited atoms in the sample emit light at specific wavelengths as they return to their ground state.
- The emitted light is analyzed by a spectrometer to identify the elements present based on their characteristic emission wavelengths.
- Emission spectroscopy is a qualitative technique used to identify the elements present in a sample and can sometimes offer semi-quantitative information.

4. What the purpose of U.V-visible spectroscopy? What is its applications in chemistry and biology?

- **Purpose:** U.V.-visible spectroscopy measures the absorbance or reflectance of ultraviolet (UV) and visible light by a sample. It allows us to study how molecules interact with light at these specific wavelengths.
- **Applications in Chemistry and Biology:**
  - **Chemical analysis:** Identify unknown compounds by comparing their absorption spectra with reference data.
  - **Concentration determination:** Quantify the concentration of colored solutions using Beer-Lambert Law.
  - **Monitoring reactions:** Track the progress of reactions that involve changes in colored components.
  - **Biomolecular studies:** Analyze the structure and interactions of biomolecules like proteins and nucleic acids, which absorb UV light due to aromatic amino acids and conjugated double bonds.

Descriptive Questions

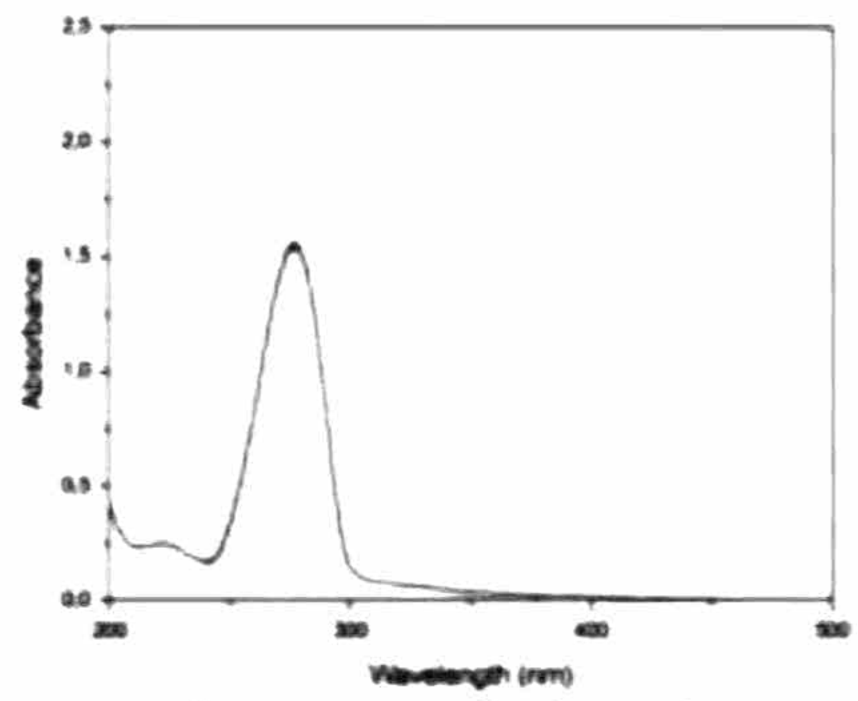
1. What information about the structure of a molecule we can get from mass spectroscopy? Give the applications of mass spectroscopy.

Notes

2. What is proton NMR spectroscopy? How does it work? Give its applications.

Notes

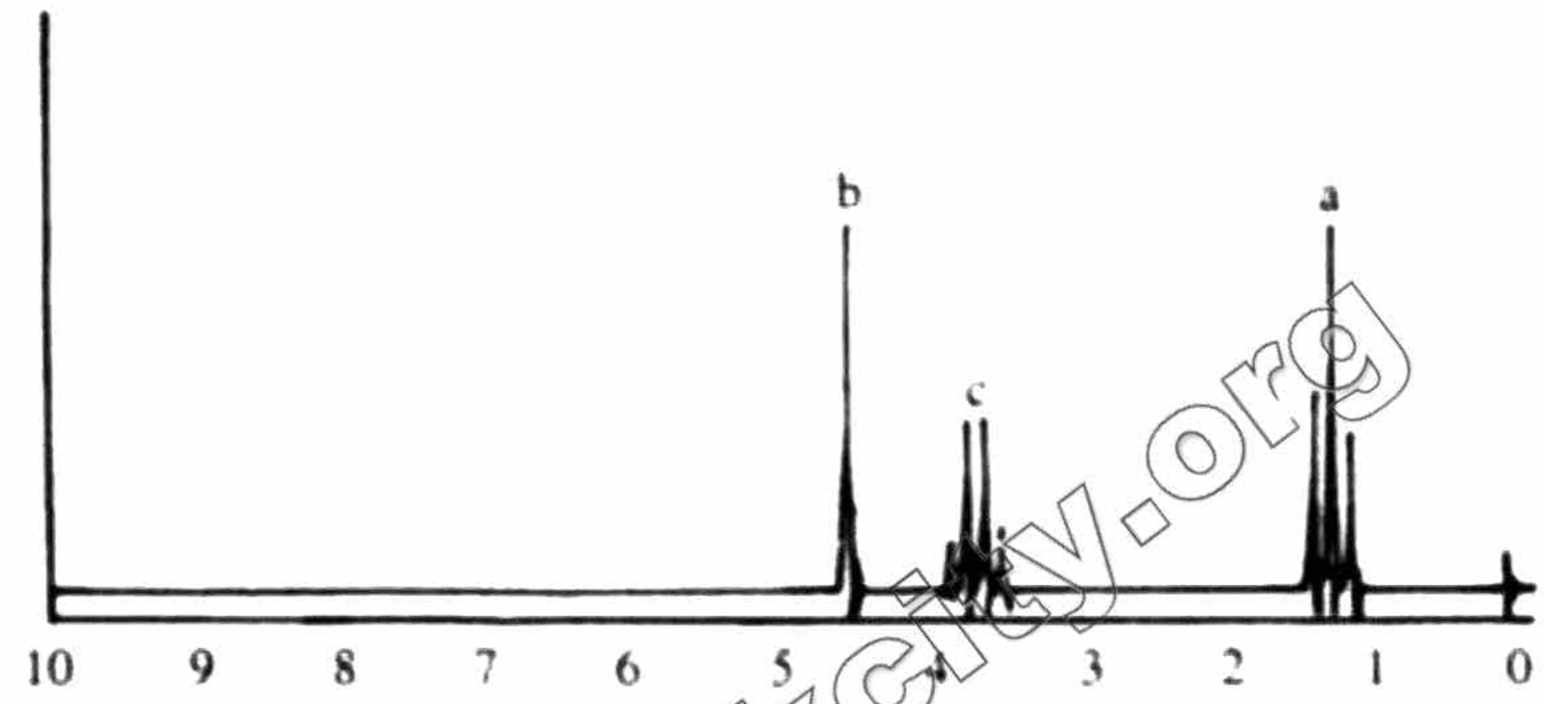
3. Explain max With the help of U.V-visible spectrum of ethanol (C_2H_5OH).



Spectrum of ethanol

Notes

4. Explain the graph of proton NMR of ethanol (C_2H_5OH) proton peaks of OH, CH_2 and CH_3



Notes

www.pakcity.org
EDUCATION
pakcity.org
pakcity.org