

Spectroscopy

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Introduction:

- In terms of the modern quantum theory, electromagnetic radiation is the **flow of photons** (also called *light quanta*) through space.
- Photons are packets of energy $h\nu$ that always move with the universal speed of light. The symbol h is Planck's constant, while the value of ν is the same as that of the frequency of the electromagnetic wave of classical theory.
- **Photons** having the same energy $h\nu$ are all alike, and their **number density corresponds to the intensity** of the radiation.
- Electromagnetic radiation exhibits a multitude of phenomena as it interacts with charged particles in atoms, molecules, and larger objects of matter.
- These phenomena as well as the ways in which electromagnetic radiation is created and observed, the manner in which such radiation occurs in nature, and its technological uses depend on its frequency ν .
- The spectrum of frequencies of electromagnetic radiation extends from very low values over the range of radio waves, television waves, and microwaves to visible light and beyond to the substantially higher values of ultraviolet light, X-rays, and gamma rays.

Spectroscopy

- Studying materials by measuring their response to different frequencies of radiation.
- Note that while a few forms of spectroscopy use other forms of radiative energy, such as acoustic or matter waves, spectroscopy is virtually always understood to use electromagnetic radiation to probe matter.
- Applications ranging from materials characterization to astronomy and medicine.
- Spectroscopy techniques are commonly categorized according to the wavelength region used, the nature of the interaction involved, or the type of material studied.

What is EMR?

- Energy propagated through free space or through a material medium in the form of electromagnetic waves.
- Examples include radio waves, infrared radiation, visible light, ultraviolet radiation, X-rays, and gamma rays.
- Electromagnetic radiation exhibits wavelike properties such as reflection, refraction, diffraction, and interference, but also exhibits particle-like properties in that its energy occurs in discrete packets or quanta.
- Though all types of electromagnetic radiation travel at the same speed, they vary in frequency and wavelength and interact with matter differently.
- A vacuum is the only perfectly transparent medium; all others absorb some frequencies of electromagnetic radiation.

What is EMR?

EMR Theory:

- ▶ **Electro (Stationary** electric charges produce an electric field)
- ▶ **Magnetism (Moving** electric charges produces both electric and magnetic fields)
- ▶ Radiation is the transportation of energy from one point to another.
- ▶ Particle is bombarded with EMR to generate Electromagnetic spectrum.

EMR □ (Interaction) □ Matter

- ▶ Spectroscopy: Deals with the study of the interaction between EMR with particles (*atoms* in the particle).

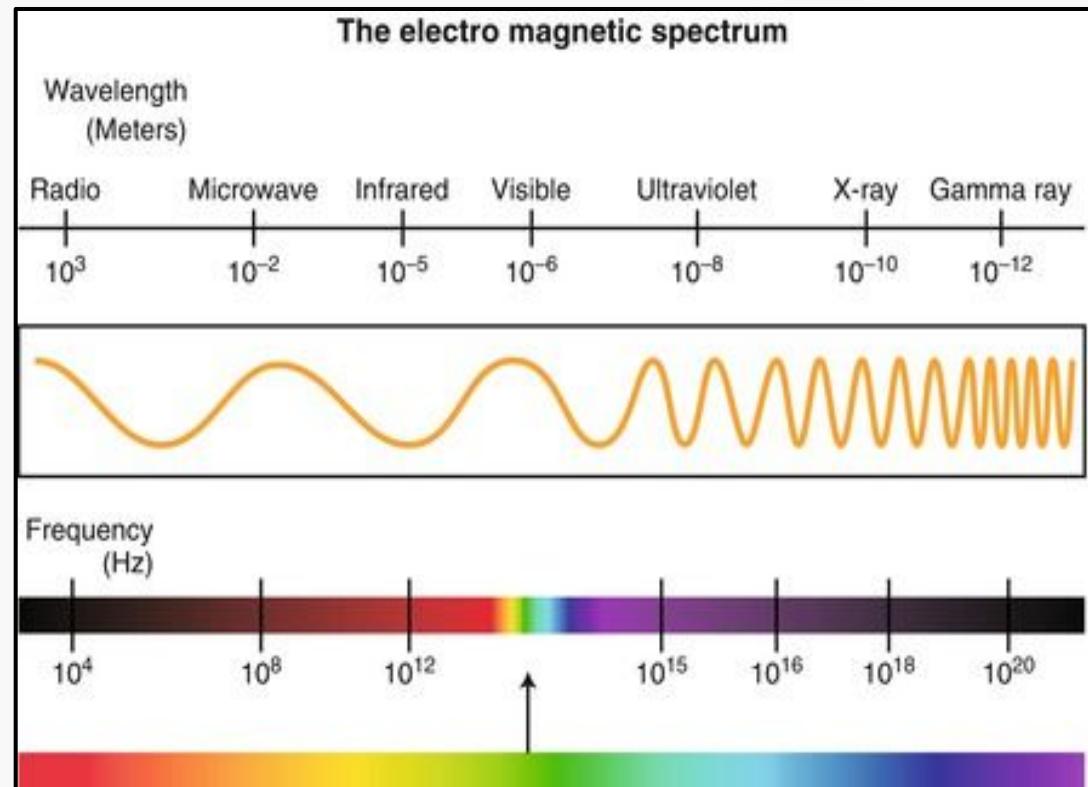


Figure 1: Electromagnetic Spectrum

What is EMR:

EMR Theory:

- EMR is a form of radiation energy that has both particle and wave nature.
- Wave nature – Electric field and magnetic field

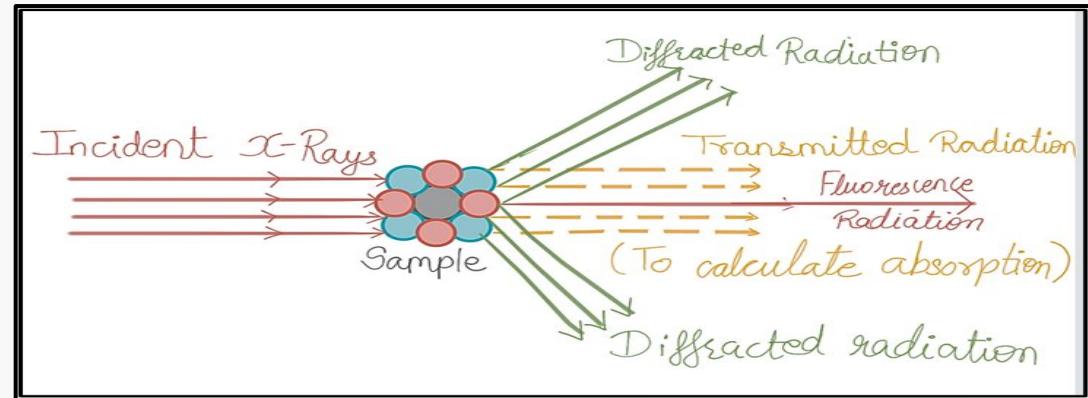


Figure 2: X-Ray striking particle

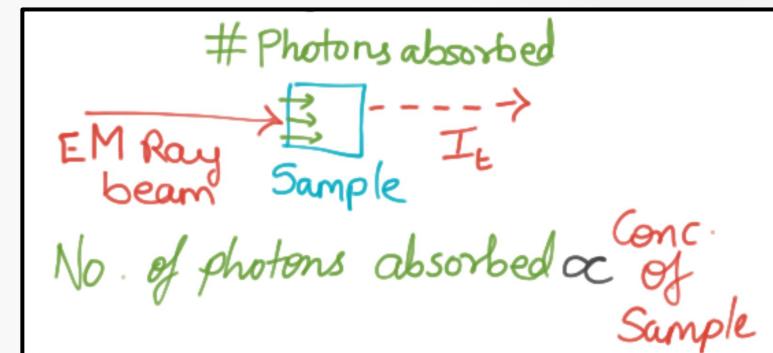


Figure 3: EMR process

Advantages of Spectroscopy:

1. Understanding Atomic and Molecular Structure

- Detailed Insight: Spectroscopy allows scientists to probe the electronic, vibrational, and rotational transitions of molecules. By analyzing the spectra, one can identify functional groups, molecular bonds, and even the overall structure of complex organic compounds.
- Non-destructive Analysis: Spectroscopic techniques are often non-invasive, meaning that the molecular structure can be studied without altering or destroying the sample, preserving its integrity for further analysis.

2. Less Time-Consuming

- Rapid Data Acquisition: Modern spectroscopic techniques, such as UV-Vis, IR, NMR, and mass spectrometry, provide rapid results, often within minutes or even seconds. This makes them highly efficient compared to other methods like crystallography or chemical degradation.
- Automated Systems: With the advent of automated spectrometers and advanced software for data analysis, the time required for both data collection and interpretation is significantly reduced, streamlining the overall process.

Advantages of Spectroscopy:

3. Small Amount of Sample Required

- High Sensitivity: Many spectroscopic techniques are highly sensitive and require only microgram to milligram quantities of the sample. This is particularly advantageous when dealing with expensive, rare, or hazardous materials.
- Preservation of Sample: The ability to use small sample sizes also means that more of the sample can be preserved for other analyses, which is critical when working with limited or valuable materials.

4. Cost-Effective in the Long Run

- Low Operational Costs: Once the initial investment in spectroscopic equipment is made, the ongoing operational costs are relatively low. Reagents, consumables, and maintenance are minimal compared to other analytical techniques.
- Broad Applicability: The versatility of spectroscopic methods means that the same equipment can be used for a wide range of analyses, reducing the need for multiple specialized instruments and further driving down costs over time.
- Reduction in Reagents: Many spectroscopic methods do not require reagents or additional chemicals, which not only saves costs but also reduces the environmental impact.

Rapidity comparison:

Compared to methods like crystallography, chemical degradation, and chromatography, spectroscopy offers significant time savings. The ability to rapidly acquire data, often within minutes, combined with automation in modern spectroscopic instruments, makes spectroscopy a far less time-consuming and more efficient choice for many types of molecular analysis.

1. Crystallography

- **Time-Consuming Process:** X-ray crystallography, while highly detailed, is a time-intensive process. It requires growing high-quality crystals, which can take days to weeks. The data collection itself, though advanced, still takes considerable time, followed by extensive data processing to solve the structure.
- **Rapid Data Acquisition in Spectroscopy:** In contrast, techniques like UV-Vis, IR, and NMR spectroscopy can provide valuable structural information within minutes. For example, an NMR spectrum can be obtained in seconds to minutes, offering insight into molecular structure without the need for crystallization.

2. Chemical Degradation

- **Laborious and Sequential:** Chemical degradation methods, such as Edman degradation for protein sequencing, are stepwise and often require sequential reactions and purifications. This process can take hours to days to complete, depending on the complexity of the molecule.
- **Efficiency of Spectroscopy:** Spectroscopic methods allow for the direct analysis of a compound's structure without the need for sequential degradation. Mass spectrometry, for example, can quickly identify molecular fragments, providing structural information in seconds.

Rapidity comparison:

3. Chromatography

- **Separation and Detection Time:** Techniques like gas chromatography (GC) and high-performance liquid chromatography (HPLC) require the sample to pass through a column for separation, which can take from several minutes to hours, depending on the complexity of the sample and the length of the column. After separation, additional detection methods are required to analyze the components.
- **Direct Analysis in Spectroscopy:** While chromatography is essential for separating complex mixtures, spectroscopy can often be applied directly to the sample without the need for separation, especially for pure compounds. This bypasses the time-consuming separation step, leading to much quicker results.

4. Automated Systems

- **Manual Interpretation in Other Techniques:** In many traditional analytical techniques, significant manual effort is required to interpret results. For example, crystallography often needs expert interpretation of electron density maps, and chemical degradation methods require careful monitoring of each step.
- **Automation in Spectroscopy:** Modern spectrometers are equipped with advanced automation capabilities, allowing for rapid, high-throughput analysis with minimal human intervention. Automated systems can handle multiple samples in sequence, perform real-time data processing, and even suggest interpretations based on databases and machine learning algorithms. This dramatically reduces the time from data acquisition to result interpretation.

Properties of EMR:

- **Photons:** Electromagnetic radiation is emitted as discrete packets of energy called photons. These photons carry light energy and travel at the constant speed of light.
- **Quantized Harmonic Waves:** Electromagnetic radiation can be described as quantized harmonic waves. These waves exhibit specific frequencies and energy levels.
- Categorization by **Wavelength:** Electromagnetic radiation is grouped into different categories based on its wavelength within the electromagnetic spectrum. Each category has distinct properties and applications.
- **Perpendicular Electric and Magnetic Fields:** Electromagnetic waves consist of perpendicular electric and magnetic fields. These fields oscillate in directions that are perpendicular to each other and to the direction of wave propagation.
- **Wavelength, Amplitude, and Frequency:** Electromagnetic radiation exhibits properties such as wavelength, amplitude, and frequency. Wavelength (λ) represents the distance between two adjacent points in phase, while frequency determines the number of wave cycles per second. Amplitude refers to the maximum displacement of the wave from its equilibrium position.
- **Travel through Empty Space:** Unlike other types of waves, electromagnetic radiation can travel through empty space. It does not require a medium such as a solid, liquid, or gas for propagation.
- **Constant Speed of Light:** The speed of light in a vacuum, denoted as “c,” is a fundamental constant of approximately 2.99792458×10^8 meters per second. This speed remains constant for all forms of electromagnetic radiation.

Understanding the properties of electromagnetic radiation allows us to comprehend its behavior, interactions with matter, and various applications across scientific disciplines and everyday life.

Interaction of Light with Matter

- Interaction of Electromagnetic waves and matter is of 2 types:

1. Absorption (Beer Lambert's Law)
2. Emission
3. Scattering

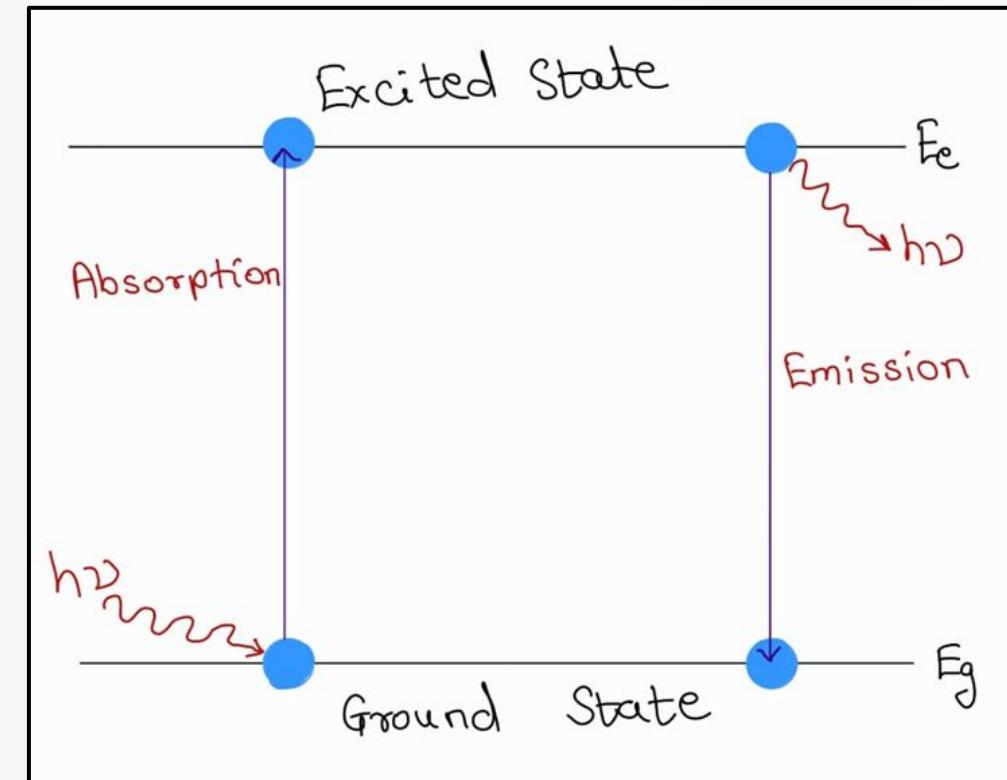


Figure 13: Absorption & Emission

Spectra and Spectrophotometer

- ▶ Spectra: Energy change or frequency if EMR emitted or absorbed can be recorded (spectra) using a spectrophotometer.

- ▶ 4 Types of Energy change:

$$E_{\text{total}} = E_{\text{translational}} + E_{\text{vibrational}} + E_{\text{rotational}} + E_{\text{electronic}}$$

- ▶ **Translational energy** is negligible in molecular spectroscopy

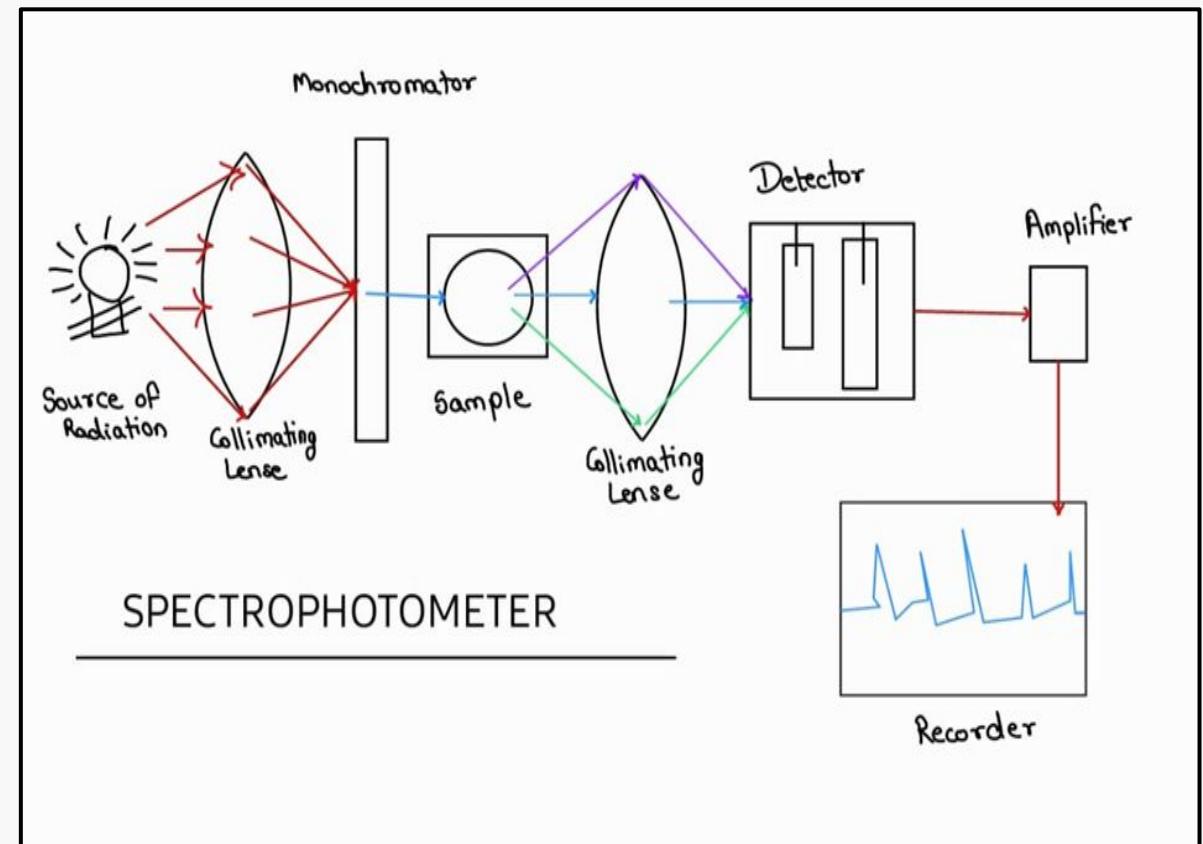


Figure 5: Spectrophotometer

Types of Spectra:

- ▶ **Rotational** (Microwave spectra): *Far IR and Microwave region.* Low energy to change the rotational level on the same vibrational and transitional level, but not enough to cause translation spectra.
- ▶ **Vibrational & Rotational** (Infrared spectra): *Near IR region.* Same electronic level, and the same transitional level but a higher vibrational level
- ▶ **Transitional** (Band spectra): *Visible and UV region.* Exciting energy is large enough to cause a transition from one electronic level to another electronic level (E_0 to E_1).

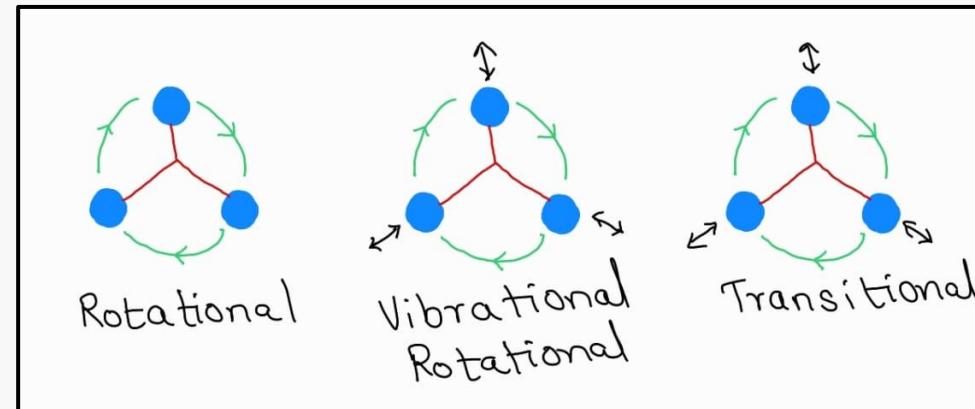


Figure 6: Molecular representation

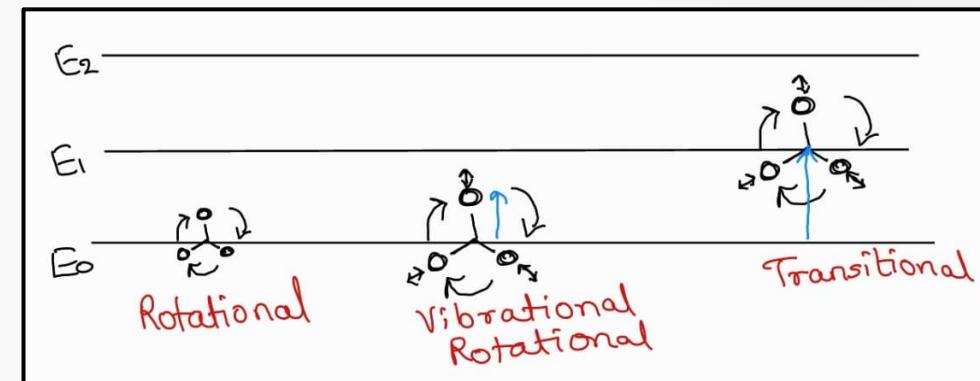


Figure 7: Energy level representation

Types of Spectroscopy:

1. Infrared (IR) Spectroscopy

- Photons in the infrared region of the electromagnetic spectrum have characteristic energies corresponding to those of molecular vibrations; this means IR spectroscopy currently remains the primary tool to study the vibrational and rotational modes of molecules.
- IR spectrometers typically measure the relative absorption of different frequencies in the IR region by a sample. This absorption spectrum can then be used to identify the types of molecular bonds present in the sample, indicating the type of molecular structures present in a sample.

2. Ultraviolet-Visible (UV/Vis) Spectroscopy

- The ultraviolet (UV) and visible regions of the electromagnetic spectrum correspond to electron energy level transitions in atoms and molecules. UV/Vis spectroscopy can therefore be used to probe the electronic structure of molecules in a sample, subsequently enabling the identification of the compounds present. UV/Vis spectroscopy is particularly useful for identifying peptide bonds, certain amino acid sidechains, and certain prosthetic groups and coenzymes. This type of spectroscopy is useful for many wearable sensor technologies.

Types of Spectroscopy:

3. Nuclear Magnetic Resonance (NMR) Spectroscopy

- Nuclear magnetic resonance spectroscopy is a technique used to measure the magnetic fields that exist around atomic nuclei. NMR spectroscopy uses radio waves to excite atomic nuclei in a sample. When nuclei start to resonate, this is detected by sensitive radio receivers.
- As the resonant frequency of an atomic nucleus depends on the electronic structure of the molecule of which it is a part, NMR spectroscopy provides detailed information about the structure and reaction state of molecules. It is hence a powerful tool for deducing the exact nature of monomolecular organic compounds.

4. Raman Spectroscopy

- Raman spectroscopy is solely concerned with the inelastic scattering of photons, known as Raman scattering, where the apparent wavelength of a photon is changed when it interacts with the sample.
- Raman scattering uses a source of monochromatic light that is used to illuminate the sample. When the laser light interacts with molecular vibrations or other excitations in the molecular system, the energy of the photons is shifted either up or down. Accurate measurement of these energy shifts enables a detailed analysis of the types of chemical bonds present in the sample. Raman scattering provides similar yet complementary data to IR spectroscopy.

Types of Spectroscopy:

5. X-Ray Spectroscopy

- The use of X-ray spectroscopy began with the development of X-ray crystallography in 1912. Father-and-son team William Henry Bragg and William Lawrence Bragg showed that the diffraction patterns created by X-rays passing through crystalline materials could be used to deduce the nature of the crystal structure.

Fun fact: X-ray astronomy has revealed very strong sources of X-rays in deep space. In the Milky Way Galaxy, of which the solar system is a part, the most-intense sources are certain double-star systems in which one of the two stars is thought to be either a compact neutron star or a black hole. The ionized gas of the circling companion star falls by gravitation into the compact star, generating X-rays that may be more than 1,000 times as intense as the total amount of light emitted by the Sun. At the moment of their explosion, supernovae emit a good fraction of their energy in a burst of X-rays.

Wavelength & Energy:

Spectral Region	Wavelength Involved	Spectroscopy
Microwave	1 to 100 cm ⁻¹	Microwave spectroscopy (Rotational spectroscopy)
Infrared	500 to 400 cm ⁻¹	Infrared spectroscopy (Vibrational-Rotational spectroscopy)
Visible & UV	12500 to 25000 cm ⁻¹ (Visible) 25000 to 70000 cm ⁻¹ (UV)	UV – visible spectroscopy (Electronic spectroscopy)

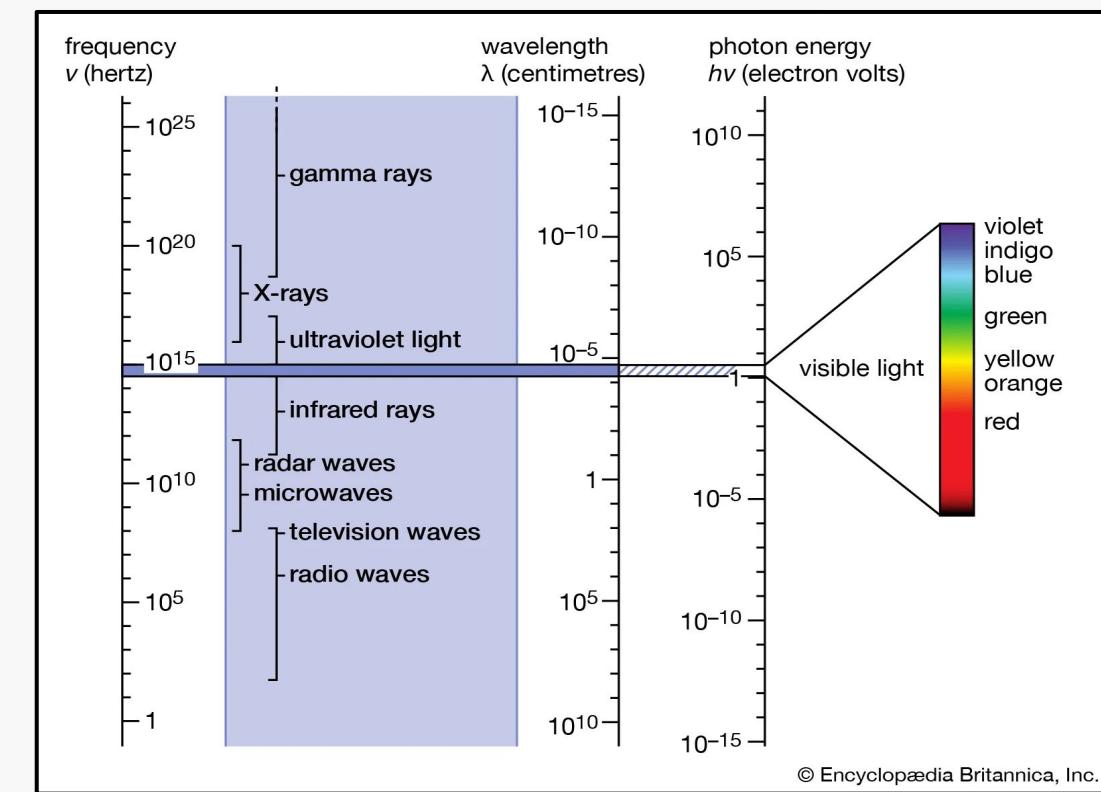


Figure 8A: Electromagnetic Spectrum

Wavelength & Energy:

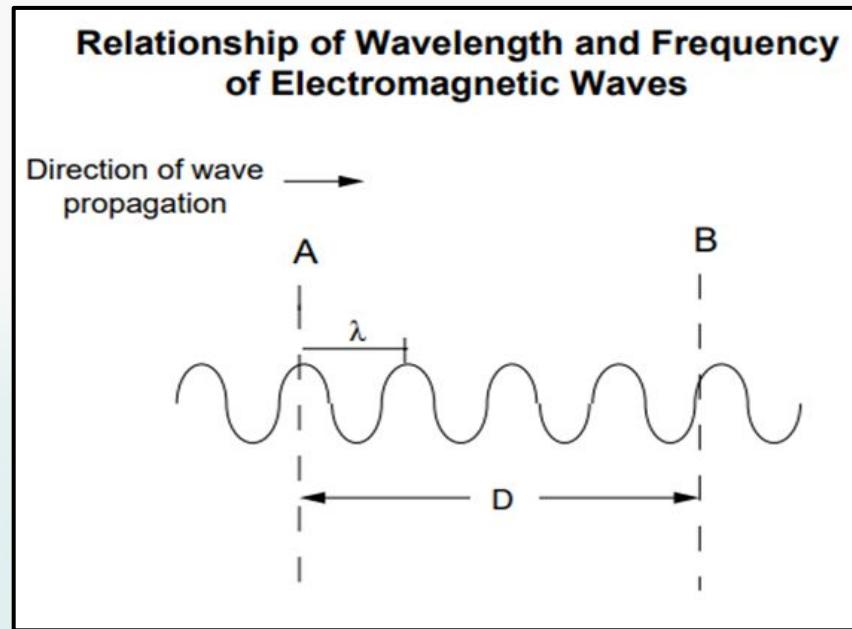


Figure 8B: Relation between wavelength, frequency and EMR

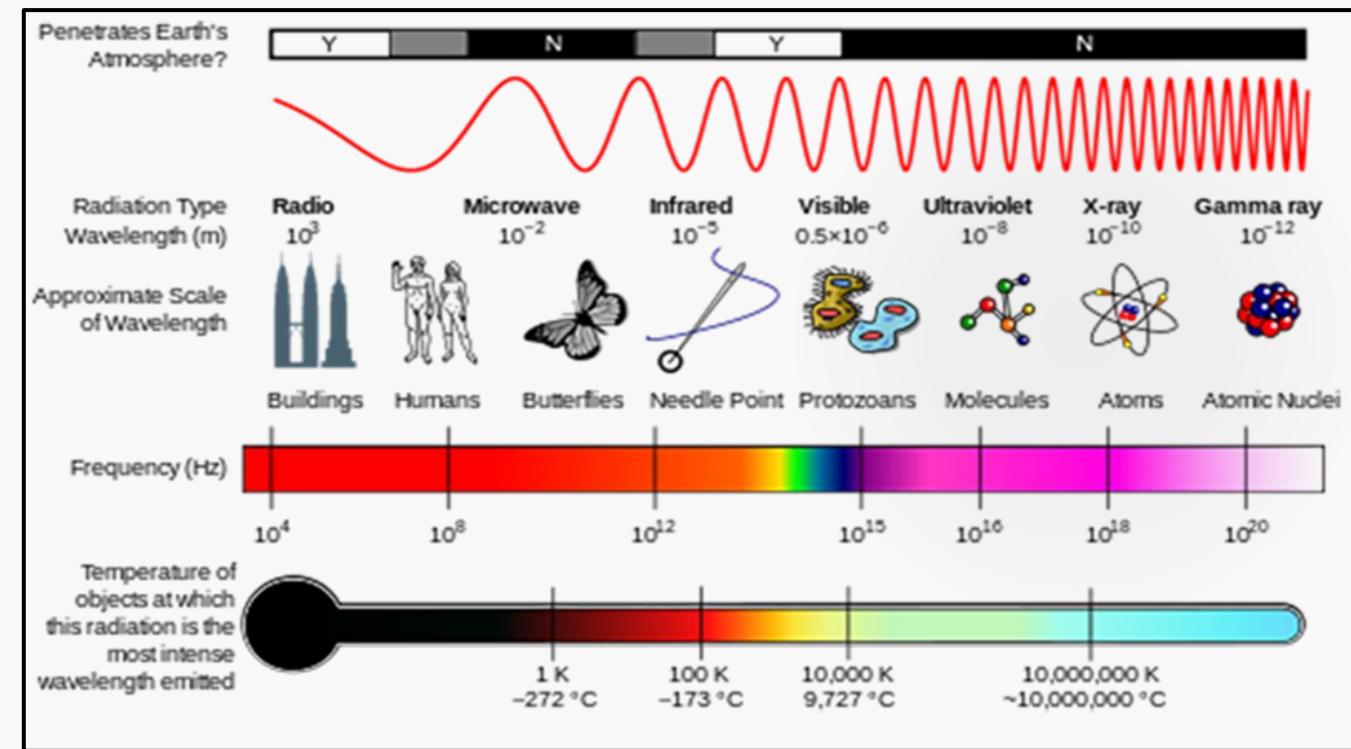


Figure 8C: Electromagnetic Spectrum Comparisons

Example working of Spectroscopy

High voltage (up to 60kV)

Tungsten filament is heated

Emits an electron (Striking electron)

Hits the metal target (Anode)

X-Rays produced in random directions

Directed by and pass through Beryllium Window

Collimator – Narrows the beam

Monochromator – Monochromatic radiation

Hit the sample and diffract

Detector detects the signal

Amplifier amplifies the signal

Recorder reads the signal

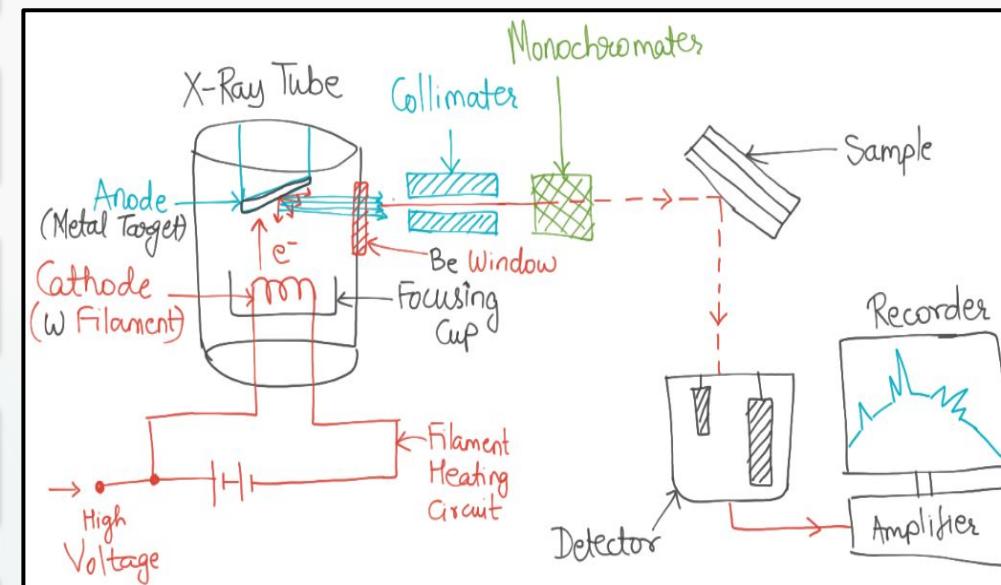


Figure 9: Instrumentation of XRD for Working.

Example: Instrumentation of X-Ray Spectroscopy:

- **Source (X-Ray Tube to produce X-Rays):**
 - Large vacuum tube with a heated **cathode** of tungsten filament and a target metal (Cu or Mo) **anode**.
- **Collimator** (narrows X-Ray beam):
 - 2 closely packed metal plates, separated by a small **gap**.
- **Monochromator:** (Filter & Crystal-(Plate & Curved))
 - Absorbs undesirable radiations, and filters the required radiations.
- **Detector:** (Photographic Method & Counter Methods)
 - A film is developed when X-rays pass through the sample and hit the film.
 - Counter methods – GM counter, Proportional Counter, Scintillation Detector, SS-SC detector, SC detector.

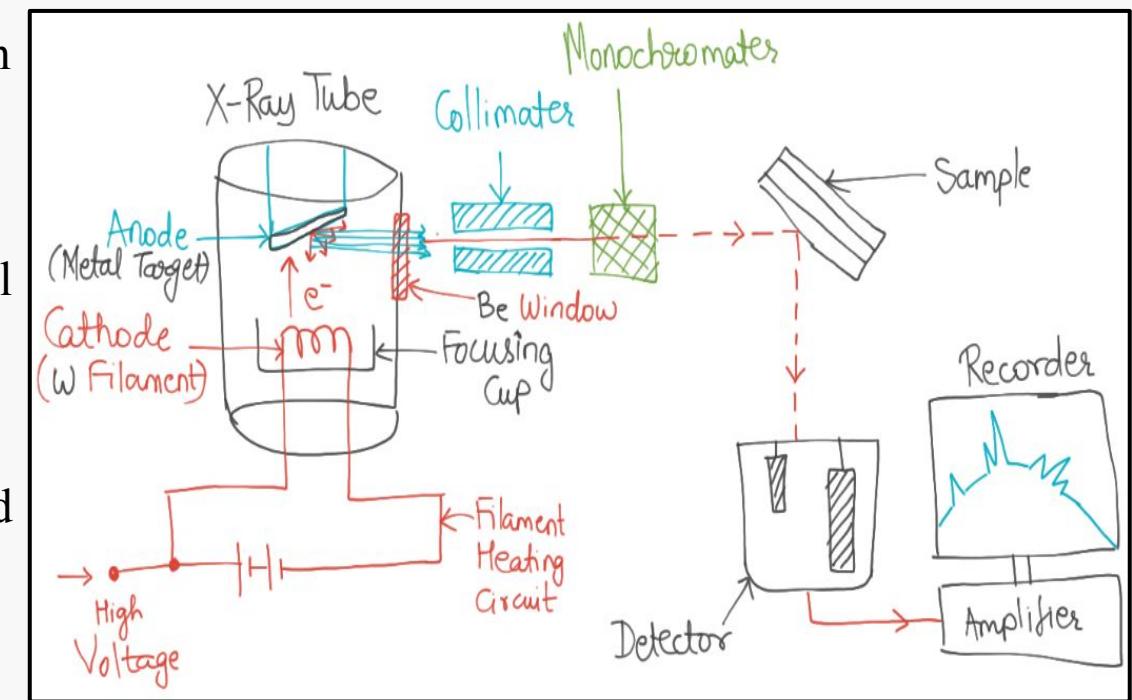


Figure 10: Instrumentation of X-Ray Diffraction

Example: X-Ray Spectroscopy:

- 2 types of **Monochromator**:

1. Filter

- Partly monochromatized
- Absorbs undesirable radiations and passes the required radiations
- If during analysis we require only K-alpha and not K-beta, we must use suitable filter (Zr filter for Mo radiation)
- However, results in slight decrease in the intensity of K-alpha

2. Crystal

- Made of suitable crystalline materials like NaCl, LiF, Quartz
- 2 types: (Plate crystal and curved crystal)

Example: X-Ray Spectroscopy:

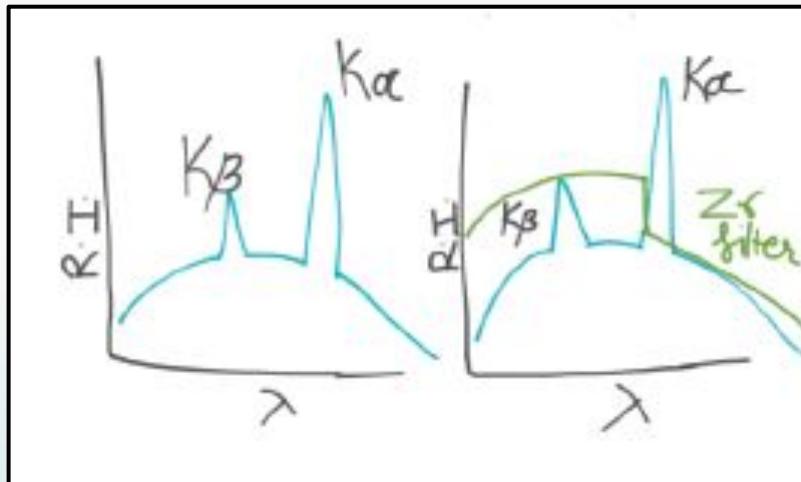


Figure 11: Filter monochromator for Mo radiation

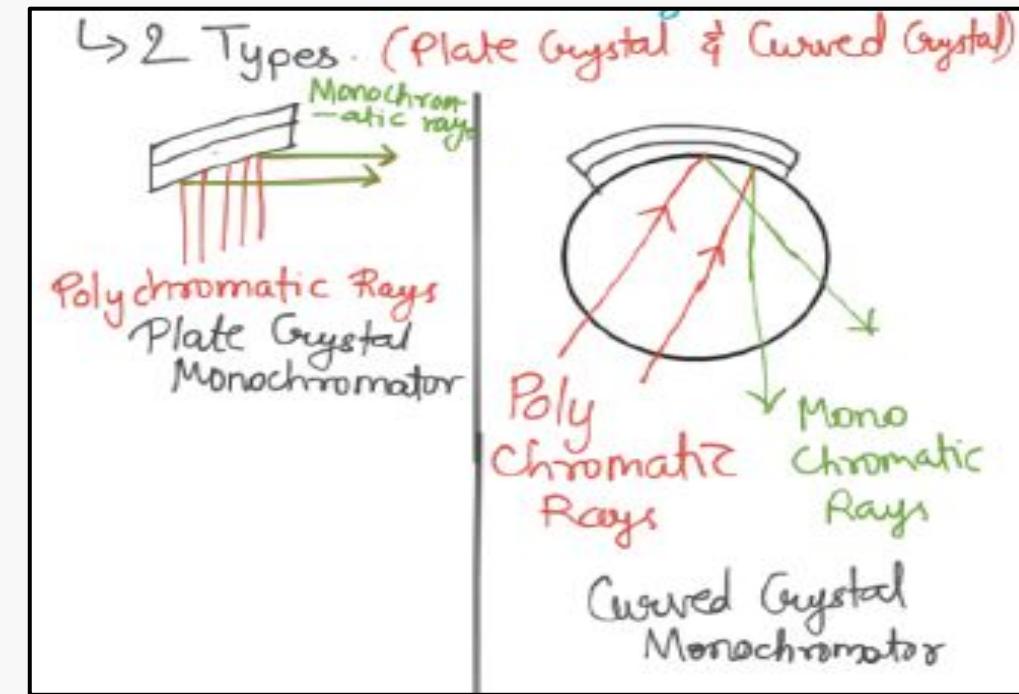


Figure 12: Two types of Crystal monochromators- Plate & Curved

Absorption spectroscopy:

- Principle: When a beam of monochromatic light passes through a solution or matter some of its radiation may be absorbed.
- Beer Lambert's Law is based on 2 laws.
- Lambert's Law: When a beam of monochromatic light passes through a transparent medium the intensity of transmitted light decrease as the thickness of absorbing material increases.
- Beer's Law: The intensity of transmitted monochromatic light decreases as the concentration of absorbing substance increases.



$$A = \varepsilon bc$$

$A \rightarrow$ Absorbance

$\varepsilon \rightarrow$ Molar Absorptivity

$b \rightarrow$ Path length

$c \rightarrow$ Concentration of the sample

Beer Lambert's Law: (Law of Absorption)

- Beer-Lambert's law for absorption spectroscopy is a linear relationship between the absorbance and the concentration of an absorbing species. The states imply that type, as well as the concentration of the molecules, are necessary.
- I_0 (incident radiation) and I_t (transmitted radiation).
- When a monochromatic beam passes through a homogeneous absorbing sample, the rate of decrease of I (intensity) of radiation with a thickness of the absorbing medium is proportional to the incident radiation.
- $T = I_t/I_0$

$$A = \log^{I_0}/I = \varepsilon cl$$

Numerical problems

Absorption spectroscopy numericals:

1. Frequency and Wavelength Calculation

Problem: Calculate the frequency (ν) of light with a wavelength (λ) of 500 nm. The speed of light (c) is 3×10^8 m/s.

2. Energy of a Photon

Problem: Calculate the energy of a photon with a frequency of 4×10^{14} Hz. Planck's constant (h) is 6.626×10^{-34} J·s.

3. Absorbance Calculation

Problem: A solution has a molar absorptivity of $2000 \text{ L/mol}\cdot\text{cm}$ and a path length (l) of 1 cm. If the concentration (c) is 0.01 M, calculate the absorbance (A).

Thank-You!