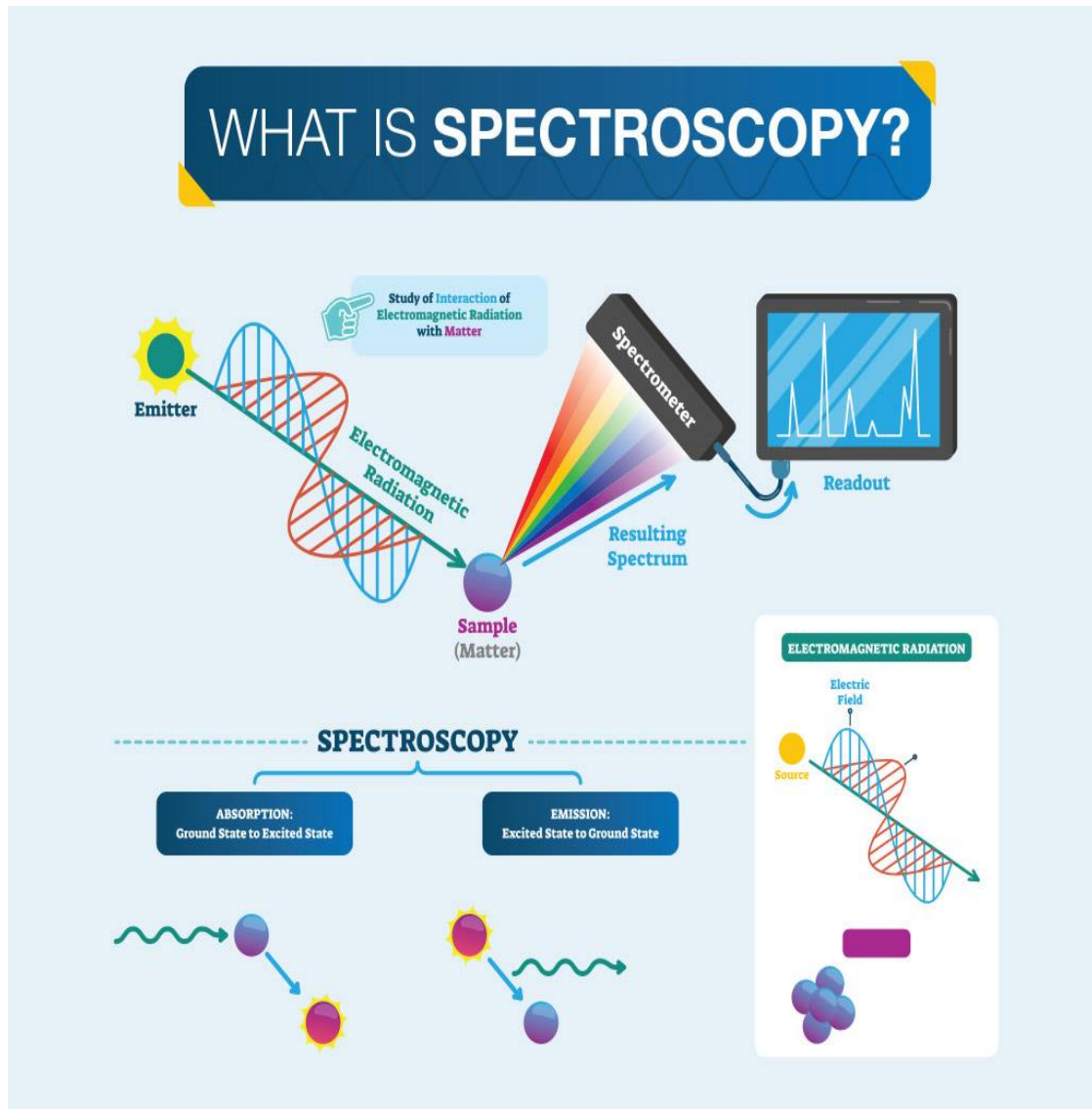


Lab. Instrument

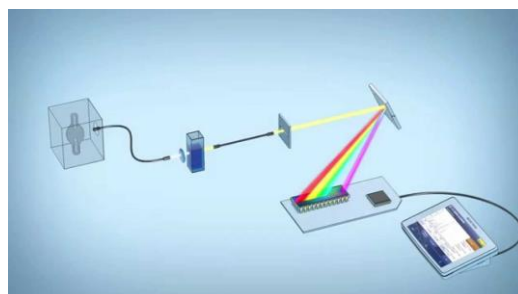
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Lecture \ 4

Spectroscopy ,Beer and Lambert Law



What is Spectroscopy?



spectroscopy was defined as the study of the interaction between radiation and matter as a function of wavelength .

1- During a spectroscopy experiment, electromagnetic radiation of a specified wavelength range passes from a source through a sample containing compounds of interest,

2- resulting in absorption or emission. During absorption, the sample absorbs energy from the light source.

3- During emission, the sample emits light of a different wavelength than the source's wavelength.

4-In absorption spectroscopy, the sample's compounds are excited by the electromagnetic radiation provided by a light source.

5-Their molecules absorb energy from the electromagnetic radiation, become excited, and jump from a low energy ground state to a higher energy state of excitation.

6-A detector, usually a photodiode, on the opposite side of the sample records the sample's absorption of wavelengths, and determines the extent of their absorption.

7-The spectrum of a sample's absorbed wavelengths is known as its absorption spectrum, and the quantity of light absorbed by a sample is its absorbance

Each molecule within a sample will only absorb wavelengths with energies corresponding to the energy difference of the present transition.

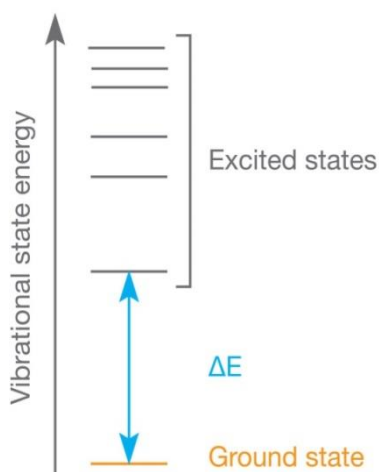
- In simpler terms, this means that a molecule that jumps from ground state 1 to excited state 2, with an energy difference of ΔE , will allow other wavelengths to pass through until it can absorb radiation from a wavelength that corresponds to ΔE .

Absorption that occurs due to an energy difference between the two states is called an absorption line, and a collection of absorption lines creates an absorption spectra

Absorption :Is a situation where some components of - .the light (colors) are retained or absorbed

Transmission: Refers to the situations where some - portions of the light permitted to pass through a given medium.

Refraction: It is defined as a sudden change in the direction of the beam when the light passes from one medium to another with a different physical density.



What is Spectroscopy Used For?

Spectroscopy is used

- 1- in physical and analytical chemistry to detect, determine, or quantify the molecular and/or structural composition of a sample.
- 2- Spectroscopy uses these characteristics to deduce and analyze the composition of a sample.

Examples of Spectroscopy Applications

- Determining the atomic structure of a sample
- Determining the metabolic structure of a muscle
- Monitoring dissolved oxygen content in freshwater and marine ecosystems
- Studying spectral emission lines of distant galaxies
- Altering the structure of drugs to improve effectiveness
- Characterization of proteins
- Space exploration
- Respiratory gas analysis in hospitals.

Spectrometer Components

1-Light Sources

2-Non-dispersive Elements

3-Dispersive elements - Prisms.

4-Dispersive elements - Diffraction Gratings

Beer's and Lambert's Law

Beer's Law: The relative amount of a certain wavelength of light absorbed (A) that passes through a sample is dependent on:

1. **Distance of the light must pass through the sample** (cell path length (b)).
2. **Amount of absorbing chemicals** in the sample (analyte concentration (c)).
3. **Ability of the sample to absorb light** (molar absorptivity (E))

$$A = \epsilon bc$$

Where: A = absorbance (no units)

b = cell path length (cm)

c = concentration of analyte (mol/L)

ϵ = molar absorptivity (L/mole-cm)

Beer's Law, the relative amount of light making it through the sample (I/I₀) is known as the transmit.

$$T = I / I_0$$

Where:

T: Transmittance

I: Intensity of light

I₀: Initial intensity of light

Beer's Law is followed only if the following conditions are met:

- **Incident radiation** on the substance of interest is monochromatic.
- **Solvent absorption** is insignificant compared to solute absorbance.
- **Solute concentration** is within given limits.
- **An optical interferant is not present.**
- **The chemical reaction does not occur** between the molecule of interest and another solute or solvent molecule.

Beer's and Lambert's Law:

Most colorimetric analytical tests are based on Beer's-Lambert's law which states that under the correct conditions the absorbance of a solution, **when measured at the appropriate wavelength, is directly proportional to its concentration and the length of the light path through the solution using a standard.** This law can be applied to measuring the concentration of a substance in an unknown (test) solution by using the formula:

$$\text{Concentration of test (} C_t \text{)} = \frac{\text{absorbance of test (AT)}}{\text{absorbance of standard} * \text{Concentration of standard (CS)}}$$

Beer's and Lambert's Law

$$\text{Light} \bullet \text{ Absorbance (A)} = \log \left(\frac{I_0}{I} \right) = \epsilon LC$$

$$\text{Light Transmission (T)} = \frac{I}{I_0} = 10^{-\epsilon LC}$$

$$\text{Light Absorbance (A)} = \log \left(\frac{I_0}{I} \right) = \epsilon LC$$

$$\text{Light Transmission (T)} = \frac{I}{I_0} = 10^{-\epsilon LC}$$

- I_0 : Light Intensity entering a sample.
- I : Light Intensity exiting a sample.
- C : The concentration of analyst in sample.
- L : The length of the light path in glass sample cuvette.
- ϵ : a constant for a particular solution and wave length.