

The study of the interaction b/w matter & electromagnetic radiation as a function of wavelength (or) frequency of the radiation is called Spectroscopy.

$$(\text{Absorbance}) A = \frac{I_0}{I}$$

Lambert's Law:-

→ Intensity of beam of parallel monochromatic radiation decreases exponentially as it passes through a medium of homogenous thickness

→ Absorbance is directly proportional to thickness.

(path length)

$$\log \left( \frac{I_0}{I} \right) = \frac{K' L}{2.303}$$

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

→ A measure of the capacity of a substance to absorb light of a specified wavelength

$L$  = path length.

$$\frac{K'}{2.303} = \text{absorptivity.}$$

→ the degree to which something absorbs energy, a liquid  
(or) another substances.

Beer's Law:-

- Intensity of beam of parallel monochromatic radiation decreases exponentially with the no. of absorbing molecules.
- Absorbance is directly proportional to concentration

$$\log \left( \frac{I_0}{I} \right) = \frac{k'c}{2.303} \quad c \rightarrow \text{Concentration}$$

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

- A measure of the capacity of a substance to absorb light of specified wavelength.

$$\frac{k'}{2.303} = \text{absorptivity}$$

- Absorbance A is directly proportional to l & c

$$A = \epsilon l c$$

Where

A = Absorbance (optical density)

l = length of sample cell (cm)

c = Concentration

$\epsilon$  = molar absorptivity (molar extinction

Coefficient)

## \* Instrumentation of UV-Visible Spectrophotometer

Components :- Source,

Filter & monochromator,

Sample compartment,

Detector,

Recorder.

## principle and instrumentation of colorimetry.

Components :- 1. Light source,

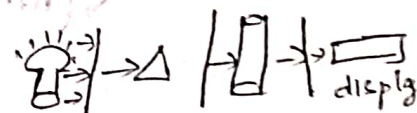
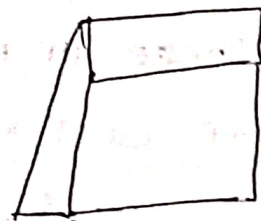
2. Condenser,

3. Monochromator, (prism)

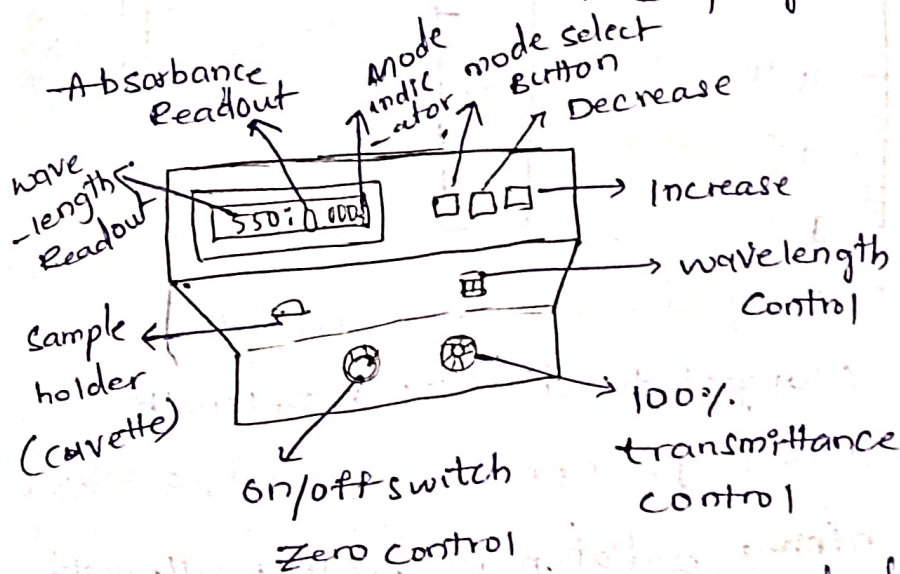
4. slit,

5. Sample holder, (cuvette)

6. Detector, (photocell)



7. Display.



Electromagnetic Spectrum :- The electromagnetic spectrum

is the entire distribution of electromagnetic radiation according to frequency (or) wavelength.

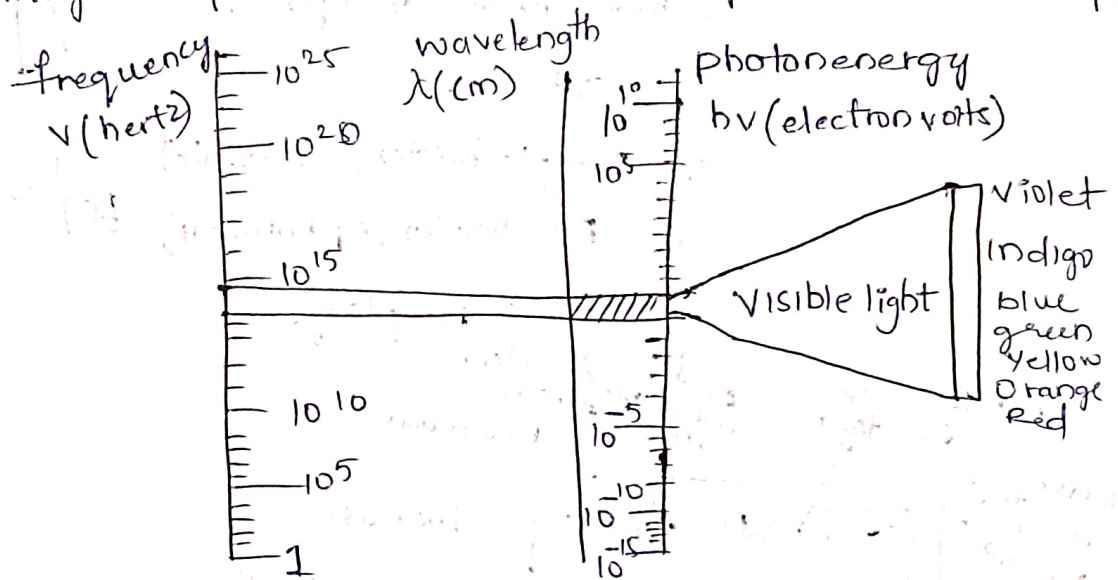
→ Although all electromagnetic waves travel at the speed of light in a vacuum, they do so at a wide range of frequencies, wavelengths, and photon energies.



→ The electromagnetic spectrum comprises the span of all electromagnetic radiation and consists of many subranges, commonly referred to as portions, such as visible light (or) ultraviolet radiation.

→ The various portions bear different names based on differences in behavior in the emission, transmission & absorption of the corresponding waves and also based on their different practical applications.

→ There are no precise accepted boundaries b/w any of these contiguous portions, so that ranges tend to overlap.



→ The entire electromagnetic spectrum, from the lowest to highest frequency (longest to shortest wavelength) includes all radio waves (e.g.:— Commercial radio & television microwaves, radar (infrared radiation, visible light, ultraviolet radiation, x-rays, and gamma rays).

→ Nearly all frequencies and wavelengths of electromagnetic radiation can be used for Spectroscopy.

**Types of Electromagnetic Radiation:**—

wavelength :→ used to broadcast radio and television

Radio microwaves :→ used in cooking, radar, telephone and other signals.

infrared :- transmits heat from sun, fires, radiators.

visible light :- makes things able to be seen.

ultraviolet :- absorbed by the skin, used in fluorescent tubes.

x-rays :- used to view inside of bodies and objects.

gamma rays :- used in medicine for killing cancer cells.

\* Absorption of Radiation : Beer-Lambert's Law :-

→ when a monochromatic light of initial intensity ( $I_0$ ) passes through a solution in a transparent container, some of the light is absorbed so that the intensity of the transmitted light ( $I$ ) is less than  $I_0$ .

→ There is some loss of light intensity from scattering by particles in the sol<sup>n</sup> & reflection at the interfaces, but mainly from absorption by the solution.

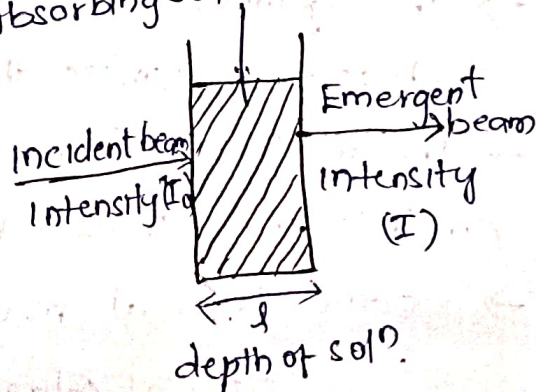
→ The relation b/w  $I$  &  $I_0$  depends on path length of the absorbing medium,  $I$ , and concentration of absorbing solution. 'c'

These factors are related to Lambert & Beer.

→ Beer-Lambert's law is obeyed when a single type species is available at relatively low concentration.

Monochromatic light

Absorbing sol<sup>n</sup>



→ Beer-Lambert's law is not obeyed when the sample used for analysis is at higher concentrations, availability of fluorescent compounds, solute & solvent form complexes.



## \* Five Basic optical instrument components.

- source :- A stable source of radiant energy at the desired wavelength (range)
- Wavelength selector :- A device that isolates a restricted region of the EM spectrum used for measurement. (monochromators, prisms & filters).
- Sample container :- A transparent container used to hold the sample (cells, cuvettes, etc)
- Detector/ photoelectric transducer :- converts the radiant energy into a useable signal (usually electrical)
- Signal processor & Readout :- Amplifies (or) attenuates the transduced signal & sends it to a readout device as a meter, digital readout, chart, recorder, computer and etc...

## \* Sample Compartment :-

- Spectroscopy requires all materials in the beam path other than the analyte should be as transparent to the radiation as possible.
- The geometries of all components in the system should be such as to maximize the signal & minimize the scattered light
- The material from which a sample cuvette is fabricated controls the optical window that can be used.
- some typical materials are :-

Optical Glass :- 335 - 2500 nm [nanometers]

Special optical glass :- 320 - 2500 nm

Quartz (infrared) - 220 - 3800 nm

Quartz (far-UV) - 170 - 2700 nm

## \* principle and Instrumentation of colorimetry

**Light source:**— The source of light should produce energy with enough intensity to cover the entire visible spectrum (380-780nm). Commonly, tungsten lamps are used as a light source for measurement in the visible spectrum and near infrared ranges. Halogen deuterium is suitable for measurement in the UV range (200-400nm).

**Slit:**— It reduces unwanted (or) stray light by allowing a light beam to pass through.

**Condensing lens:** → parallel beam of light emerges from Condensing lens after the light passes through slit incidents on it.

**Monochromator:** → It filters the monochromatic light from polychromatic light which absorbs unwanted light wavelength and permits only monochromatic light. These are of three types prism, grating and glass.

**Prism:**— It facilitates the refraction of light when it passes from one medium to another.

**Glass:**— It selectively transmits light in certain ranges of wavelength.

**Gratings:**— These are made of graphite which separates light in different wavelength.

**Cuvette (sample cell):**— The monochromatic light from the filter passes through the colored sample solution placed in the Cuvette. Their sizes range from square & rectangle round and have a fixed diameter of 1cm.



# Instrumentation of flame photometry.

→ various process involved in the flame photometry

Desolvation

Vaporization

Atomization

Excitation of elements

Emission.

Principle of flame photometer:-

→ Is based on the measurement of the emitted light intensity when a metal is introduced into the flame.

→ The wavelength of the colour gives information about the element and the colour of the flame gives information about the amount of the element present in the sample.

→ Flame photometry is one of the branches of atomic absorption spectroscopy.

→ It is also known as flame emission spectroscopy.

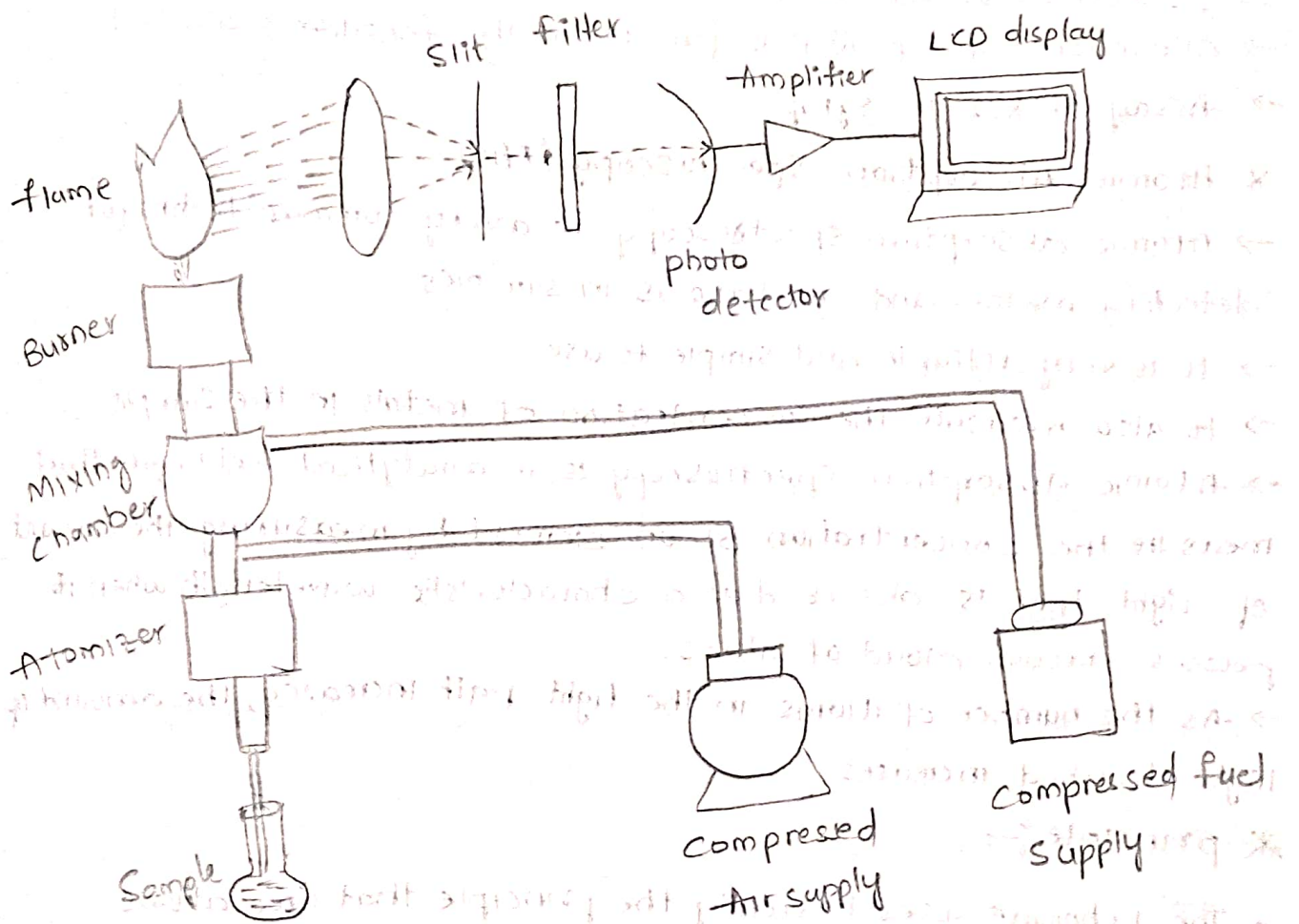
→ The compounds of the alkali and alkaline earth metals (Group II) dissociate into atoms when introduced into the flame.

→ Some of these atoms further get excited to even higher levels. But these atoms are not stable at higher levels.

Hence, these atoms emit radiations when returning back to the ground state.

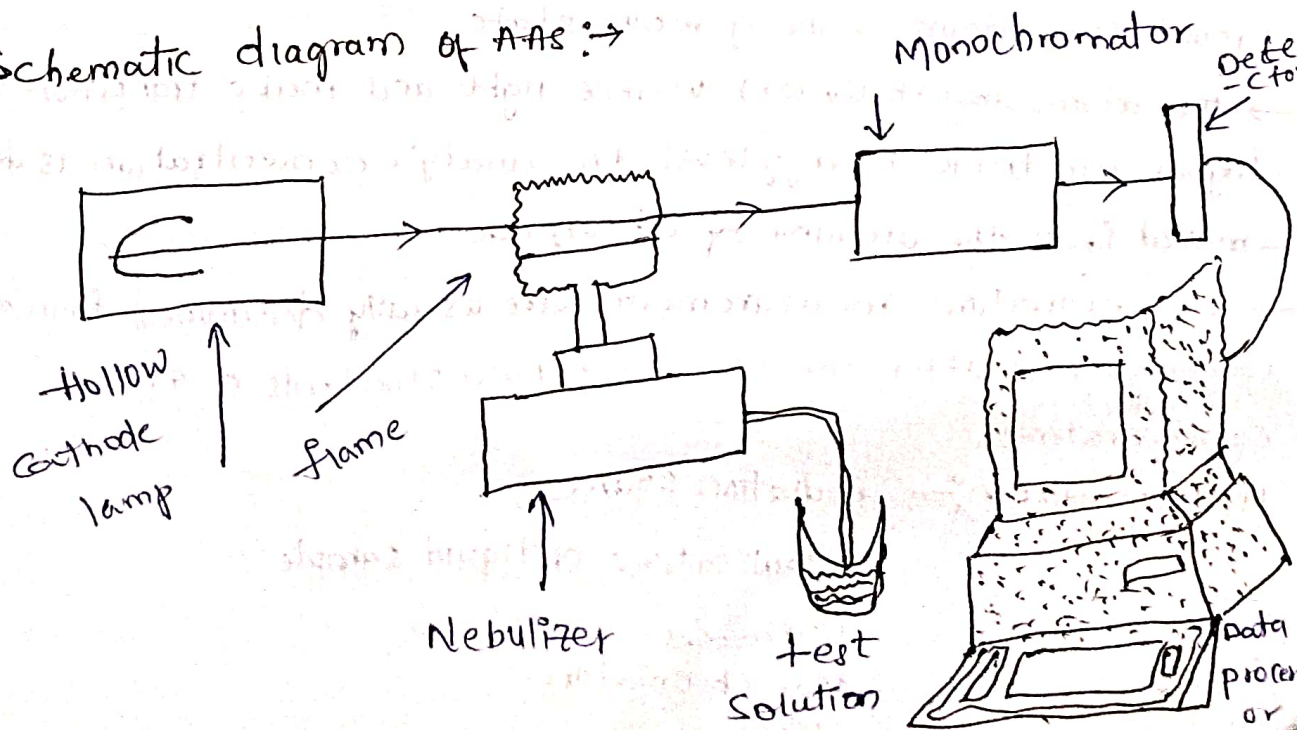


- These radiations generally lie in the visible region of the spectrum.
- Each of the alkali and alkaline earth metals has a specific wavelength.



## \* Atomic Absorption Spectroscopy (AAS)

Schematic diagram of AAS:→



## Applications.

**Qualitative analysis:**— Flame photometry is used to identify the elements present in the sample.

**Quantitative Analysis:**— Concentration of the sample can be determined by this method.

- concentration of Calcium in Serum.
- concentration of N, P, K present in the fertilizers and soil
- Assay of KCl in Syrup.

### \* Atomic Absorption Spectroscopy (AAS)

- Atomic absorption spectroscopy is a very common technique detecting metals and metalloids in samples.
- It is very reliable and simple to use.
- It also measure the concentration of metals in the sample.
- Atomic absorption spectroscopy is an analytical technique that measure the concentration of an element by measuring the amount of light that is absorbed at a characteristic wavelength when it passes through cloud of atoms.
- As the number of atoms in the light path increases, the amount of light absorbed increases.

### \* principle:

- The technique uses basically the principle that free atoms generated in an atomizer can absorb radiation at specific frequency.
- Atomic absorption spectroscopy quantifies the absorption of ground state atoms in the gaseous state.
- The atoms absorb UV (or) visible light and make transition to higher electronic energy level. the analyte concentration is determined from the amount of absorption.
- concentration measurements are usually determined from a working curve after the instrument with standards of known concentration.

**instrumentation:**— Radiation Source

• Nebulisations of liquid sample

• Atomizers

Monochromators

Detector



Radiation Source:- Hollow cathode lamp are the most common radiation source in AAS.

- It contains a tungsten anode and a hollow cylindrical cathode
- These are sealed in a glass tube filled with an inert gas (mainly neon and argon)
- Each element has its own unique lamp which must be used for that analysis.

Nebulizer:- Nebulizer suck up liquid sample at controlled rate

- Create a fine aerosol spray for introduction into the flame.
- Mix the aerosol and fuel and oxidant thoroughly for introduction into flame.

Atomizers:- elements to be analyzed needs to be in atomic state.

- Atomization is separation particles into individual molecules and breaking molecules into atoms.

- This is done by exposing the analyte to high temperatures in a flame and graphite furnace.

- The atomizers most commonly used nowadays are:-

- \* (Spectroscopic) flames and

- \* Electro thermal (graphite tube) atomizers.

Flame atomizers:- Nebulizer suck up liquid sample at controlled rate and creates a fine aerosol spray for introduction into flame.

- To create flame, we need to mix an oxidant gas and a fuel gas
- In most of the cases air-acetylene flame (or) nitrous acetylene flame is used.

- Liquids (or) dissolved samples are typically used with flame atomizer.