

## LEARNING PRINCIPLE OF WORKING OF PHOTO COLORIMETER

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**Abstract:** In this article, we talk about principle of colorimeter, working of colorimeter, parts of colorimeter.

**Key words:** photo colorimeter, Beer's law, Lambert's law, Absorption of light, electromagnetic wave.

### COLORIMETER – COMPONENTS, WORKING

A Colorimeter involves the measurement of Color and is the widely used method for finding the concentration of biochemical compounds. It Measures absorbance and wavelength between 400 to 700 nm (nanometer) i.e. from the visible spectrum of light of the electromagnetic spectrum.

**Absorption of light** – Light falling on a colored solution is either absorbed or transmitted. A colored solution absorbs all the colors of white light and selectively transmits only one color. This is its own color.

#### **Beer's Law**

This law states that the amount of light absorbed is directly proportional to the concentration of the solute in the solution.

$$\text{Log}_{10} I_0/I_t = a_s c$$

where,

$a_s$  = Absorbency index

$c$  = Concentration of Solution

#### **Lambert's Law**

The Lambert's law states that the amount of light absorbed is directly proportional to the length and thickness of the solution under analysis.

$$A = \log_{10} I_0/I_t = a_s b$$

Where,

$A$  = Absorbance of test

$a_s$  = Absorbance of standard

$b$  = length / thickness of the solution

*The mathematical representation of the combined form of Beer-Lambert's law is as follows:*

$$\text{Log}_{10} I_0 / I_t = a_s bc$$

If  $b$  is kept constant by applying Cuvette or standard cell then,

$$\text{Log}_{10} I_0/I_t = a_s c$$

The absorbency index  $a_s$  is defined as

$$a_s = A/cl$$

Where,

$c$  = concentration of the absorbing material (in gm/liter).

$l$  = distance traveled by the light in solution (in cm).

**In simplified form,**

The working principle of the colorimeter is based on Beer-Lambert's law which states that the amount of light absorbed by a color solution is directly proportional to the concentration of the solution and the length of a light path through the solution.

$$A \propto cl$$

Where,

$A$  = Absorbance / Optical density of solution

$c$  = Concentration of solution

$l$  = Path length

$$A = \epsilon cl$$

$\epsilon$  = Absorption coefficient

### WORKING OF THE COLORIMETER

When using a colorimeter, it requires being calibrated first which is done by using the standard solutions of the known concentration of the solute that has to be determined in the test solution. For this, the standard solutions are filled in the cuvettes and placed in the cuvette holder in the colorimeter.

There is a ray of light with a certain wavelength that is specific for the assay is directed towards the solution. Before reaching the solution the ray of light passes through a series of different filters and lenses. These lenses are used for navigation of the colored light in the colorimeter and the filter splits the beam of light into different wavelengths and allows the required wavelength to pass through it and reaches the cuvette containing the standard or test solutions. It analyzes the reflected light and compares it with a predetermined standard solution.

When the monochromatic light (light of one wavelength) reaches the cuvette some of the light is reflected, some part of the light is absorbed by the solution and the remaining part is transmitted through the solution which falls on the photodetector system. The photodetector system measures the intensity of transmitted light and converts it into the electrical signals that are sent to the galvanometer.

The galvanometer measures the electrical signals and displays them in the digital form. That digital representation of the electrical signals is the absorbance or optical density of the solution analyzed.

If the absorption of the solution is higher than there will be more light absorbed by the solution and if the absorption of the solution is low then more lights will be transmitted through the solution which affects the galvanometer reading and corresponds to the concentration of the solute in the solution. By putting all the values in the formula given in the below section one can easily determine the concentration of the solution.

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