A colorimeter is a light-sensitive device used for measuring the transmittance and absorbance of light passing through a liquid sample. The device measures the intensity or concentration of the colour that develops upon introducing a specific reagent into a solution.

There are two types of colorimeters — colour densitometers, which measure the density of primary colours, and colour photometers, which measure the colour reflection and transmission.

Design of Colorimeter:

The three main components of a colorimeter are a light source, a cuvette containing the sample solution and a photocell for detecting the light passed through the solution.

The instrument is also equipped with either coloured filters or specific LEDs to generate colour. The output from a colorimeter may be displayed by an analog or digital meter in terms of transmittance or absorbance.

In addition, a colorimeter may contain a voltage regulator for protecting the instrument from fluctuations in mains voltage. Some colorimeters are portable and useful for on-site tests, while others are larger, bench-top instruments are useful for laboratory testing.

Working Principle:

Colorimetry is a widely used technique applied in biological system. It involves the measurement of a compound or a group of compounds present in a complex mixture. The property of colorimetric analyses is to determine the intensity or concentration of compounds in coloured solution.

This is done by passing light of specific wavelength of visible spectrum through the solution in a photoelectric colorimeter instrument and observes the galvanometric reading of reflection sensitizing the quantity of light absorbed.

Based on the nature of colour compounds, specific light filters are used. Three types of filters are available — blue, green and red — with corresponding light wavelength transmission rays from 470-490 nm, 500-530 nm and 620-680 nm, respectively.

There are two fundamental laws of absorption which are highly important in colorimetric estimation. These are Lambert's law and Beer's law. Lambert's law states that when monochromatic light passes through a solution of constant concentration, the absorption by the solution is directly proportional to the length of the solution.

In contrary, Beer's law states that when monochromatic light passes through a solution of constant length, the absorption by the solution is directly proportional to the concentration of the solution.

Thus both the laws can be expressed as:

Lambert's law: $\log_{10} I_0/I = K_1 I$

Beer's law: $\log_{10} I_0/I = K_2 I$

[Where I_0 = Intensity of incident light (light entering a solution);

I = Intensity of transmitted light (light leaving a solution);

l = Length of absorbing solution;

c = Concentration of coloured substance in solution;

K_1 and K_2 = Constants.]

Both Beer-Lambert laws are combined together for getting the expression transmittance (T).

$$\mathbf{T = I/I_0}$$

(Where I_0 is the intensity of incident radiation and I is the intensity of transmitted radiation).

A 100% value of 'T' represents a totally transparent substance, with no radiation being aborted, whereas a zero value of 'T' represents a totally opaque substance that, in effect, represents complete absorption. For intermediate value we can define the absorbance (A) or extinction (E) that is given by the logarithm (to base 10) of the reciprocal of the transmittance:

$$\mathbf{A = E = \log_{10} (I/T) = \log_{10} (I_0/I)}$$

Absorbance used to be called optical density (OD) but continued use of this term should be discouraged. Also, as absorbance is a logarithm it is, by definition, unit-less and has a range of values from 0 (= 100% T) to cc (= 0% T).

Thus the variation of colour of the reaction mixture (or system) with change of substrate concentration forms the basis of colorimetric analysis.

The formation of colour is due to the reaction between substances and reagents in appropriate proportion. The intensity of colour observed is then compared with that of reaction mixture which contains a known amount of substrate. The optical spectrophotometry is based on identical principles of colorimetry.

Applications:

Colorimeters are widely used to monitor the growth of a bacterial or yeast culture. They provide reliable and highly accurate results when used for the assessment of colour in bird plumage. They are used to measure and monitor the colour in various foods and beverages, including vegetable products and sugar. Certain colorimeters can measure the colours that are used in copy machines, fax machines and printers.

Besides being used for basic research in chemistry laboratories, colorimeters have many practical applications such as testing water quality by screening chemicals such as chlorine,

fluoride, cyanide, dissolved oxygen, iron, molybdenum, zinc and hydrazine. They are also used to determine the concentrations of plant nutrients such as ammonia, nitrate and phosphorus in soil or haemoglobin in blood. Colorimetry is also used in colour printing, textile manufacturing and paint manufacturing for precise quality inspection.

Colorimeter vs. Spectrophotometer:

Like colorimeters, spectrophotometers are used to measure the colour absorbing properties of a substance. The key difference between the two is that the spectrophotometer measures the transmittance and reflectance as a function of wavelength, whereas the colorimeter measures the absorbance of specific colours.

Spectrophotometers measure the transmittance and reflectance for all colours of light, and shows how they vary as the colour is changed. Colorimeters operate only in the visible portion of the electromagnetic spectrum whereas spectrophotometers work with infrared as well as visible light. Spectrophotometers will produce valid results for Beer's law and can effectively be used as colorimeters, but are much higher in cost and complexity.