Spectrophotometry

Spectrophotometer

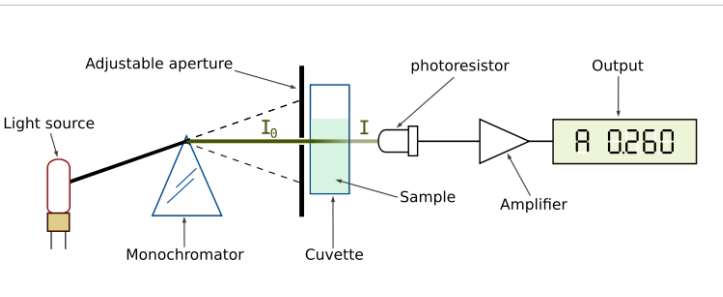
In chemistry, **spectrophotometry** is the quantifiable study of electromagnetic spectra. It is more specific than the general term electromagnetic spectroscopy in that spectrophotometry deals with visible light, near-ultraviolet, and near-infrared. Also, the term does not cover time-resolved spectroscopic techniques.

Spectrophotometry involves the use of a spectrophotometer. A spectrophotometer is a photometer (a device for measuring light intensity) that can measure intensity as a function of the color (or more specifically the wavelength) of light. Important features of spectrophotometers are spectral bandwidth and linear range of absorption measurement.

Perhaps the most common application of spectrophotometers is the measurement of light absorption, but they can be designed to measure diffuse or specular reflectance. Strictly, even the emission half of a luminescence instrument is a type of spectrophotometer.

The use of spectrophotometers is not limited to studies in physics. They are also commonly used in other scientific fields such as chemistry, biochemistry, and molecular biology.[1] They are widely used in many industries including printing and forensic examination.

Design



Single beam spectrophotometer

There are two major classes of devices: single beam and double beam. A double beam spectrophotometer compares the light intensity between two light paths, one path containing a reference sample and the other the test sample. A single beam spectrophotometer measures the relative light intensity of the beam before and after a test sample is inserted. Although comparison measurements from double beam instruments are easier and more stable, single beam instruments can have a larger dynamic range and are optically simpler and more compact.

Historically, spectrophotometers use a monochromator containing a diffraction grating to produce the analytical spectrum. There are also spectrophotometers that use arrays of photosensors. Especially for infrared spectrophotometers, there are spectrophotometers that use a Fourier transform technique to acquire the spectral information quicker in a technique called Fourier Transform InfraRed.

The spectrophotometer quantitatively compares the fraction of light that passes through a reference solution and a test solution. Light from the source lamp is passed through a monochromator, which diffracts the light into a

"rainbow" of wavelengths and outputs narrow bandwidths of this diffracted spectrum. Discrete frequencies are transmitted through the test sample. Then the intensity of the transmitted light is measured with a photodiode or other light sensor, and the transmittance value for this wavelength is then compared with the transmission through a reference sample.

In short, the sequence of events in a spectrophotometer is as follows:

1. The light source shines into a monochromator.
2. A particular output wavelength is selected and beamed at the sample.
3. The sample absorbs light.

Many spectrophotometers must be calibrated by a procedure known as "zeroing." The absorbency of a reference substance is set as a baseline value, so the absorbencies of all other substances are recorded relative to the initial "zeroed" substance. The spectrophotometer then displays % absorbency (the amount of light absorbed relative to the initial substance).[1]

UV and IR spectrophotometers

The most common spectrophotometers are used in the UV and visible regions of the spectrum, and some of these instruments also operate into the near-infrared region as well.

Visible region 400–700 nm spectrophotometry is used extensively in colorimetry science. Ink manufacturers, printing companies, textiles vendors, and many more, need the data provided through colorimetry. They take readings in the region of every 10–20 nanometers along the visible region, and produce a spectral reflectance curve or a data stream for alternative presentations. These curves can be used to test a new batch of colorant to check if it makes a match to specifications e.g., iso printing standards.

Traditional visual region spectrophotometers cannot detect if a colorant or the base material has fluorescence. This can make it difficult to manage color issues if for example one or more of the printing inks is fluorescent. Where a colorant contains fluorescence, a bi-spectral fluorescent spectrophotometer is used. There are two major setups for visual spectrum spectrophotometers, d/8 (spherical) and 0/45. The names are due to the geometry of the light source, observer and interior of the measurement chamber. Scientists use this instrument to measure the amount of compounds in a sample. If the compound is more concentrated more light will be absorbed by the sample; within small ranges, the Beer-Lambert law holds and the absorbance between samples vary with concentration linearly. In the case of printing measurements two alternative settings are commonly used- without/with uv filter to control better the effect of uv brighteners within the paper stock.

Samples are usually prepared in cuvettes; depending on the region of interest, they may be constructed of glass, plastic, or quartz.

IR spectrophotometry

Spectrophotometers designed for the main infrared region are quite different because of the technical requirements of measurement in that region. One major factor is the type of photosensors that are available for different spectral regions, but infrared measurement is also challenging because virtually everything emits IR light as thermal radiation, especially at wavelengths beyond about 5 μm.

Another complication is that quite a few materials such as glass and plastic absorb infrared light, making it incompatible as an optical medium. Ideal optical materials are salts, which do not absorb strongly. Samples for IR spectrophotometry may be smeared between two discs of potassium bromide or ground with potassium bromide and pressed into a pellet. Where aqueous solutions are to be measured, insoluble silver chloride is used to construct the cell.

Spectroradiometers

Spectroradiometers, which operate almost like the visible region spectrophotometers, are designed to measure the spectral density of illuminants in order to evaluate and categorize lighting for sales by the manufacturer, or for the customers to confirm the lamp they decided to purchase is within their specifications. Components:

1. The light source shines onto or through the sample.
2. The sample transmits or reflects light.
3. The detector detects how much light was reflected from or transmitted through the sample.
4. The detector then converts how much light the sample transmitted or reflected into a number.

See also

- Atomic Absorption Spectrophotometry
- Atomic emission spectroscopy
- Inductively coupled plasma atomic emission spectroscopy
- Inductively coupled plasma mass spectrometry
- Spectroradiometry

External links

- Optical systems using concave gratings [2]

References

[1] Rendina, George. *Experimental Methods in Modern Biochemistry* W. B. Saunders Company: Philadelphia, PA. 1976. pp. 46-55

[2] http://gratings.newport.com/information/handbook/chapter7.asp

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