



Spectrophotometer

Medical Laboratory Instruments

1st Stage, Laboratory 4

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What is a Spectrophotometer

Measures the intensity of light absorbed by a sample.

Determines chemical concentrations by measuring light intensity.

Types of Spectrophotometers

1. UV-Visible Spectrophotometer

Wavelength range: 180–750 nm.

2. Infrared (IR) Spectrophotometer

Wavelength range: 750–15,000 nm.

Single vs. Double Beam Spectrophotometers

Single Beam: Operates between 325–1000 nm.

One light beam used for sample and blank measurements.

Double Beam: Operates between 185–1000 nm
Splits light into two beams: one for reference, one for the sample..

Components of a Spectrophotometer

1. Light Source

Tungsten lamp (visible spectrum). Deuterium lamp (UV). Nernst filament/globar (IR).

2. Collimator Lens

Produces parallel rays from focused light.

3. Monochromator

Uses a prism or diffraction grating to select specific wavelengths.

4. Sample Holder (Cuvettes): Made of plastic, glass, or quartz for visible light.

5. Detector: Generates current proportional to light intensity.

6. Recorder: Measures and records light intensity.

Uses of a Spectrophotometer

Quantification of DNA, RNA, and proteins.

Measuring concentration of color/colorless compounds.

Monitoring reaction phases.

Identifying compounds using absorption spectrum.

Calibration Curve

Plots signal vs. concentration.

Follows linear equation: $y = mx + b$.

Non-linear data is excluded.

Preventive Measures

1. Turn on device 10–15 minutes before use.
2. Calibrate before each use.
3. Choose the optimal wavelength.
4. Avoid reactive substances in samples.
5. Ensure proper sample concentration.

Conclusion

A spectrophotometer is an essential tool in medical and chemical analysis.

Accuracy depends on calibration, preparation, and preventive measures.



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