Al-Mustaqbal University College
Department of Medical Instrumentation Techniques Engineering
Class: second
Subject: Clinical Chemistry
Lecturer: Mustafa Fahad
Lecture: Two

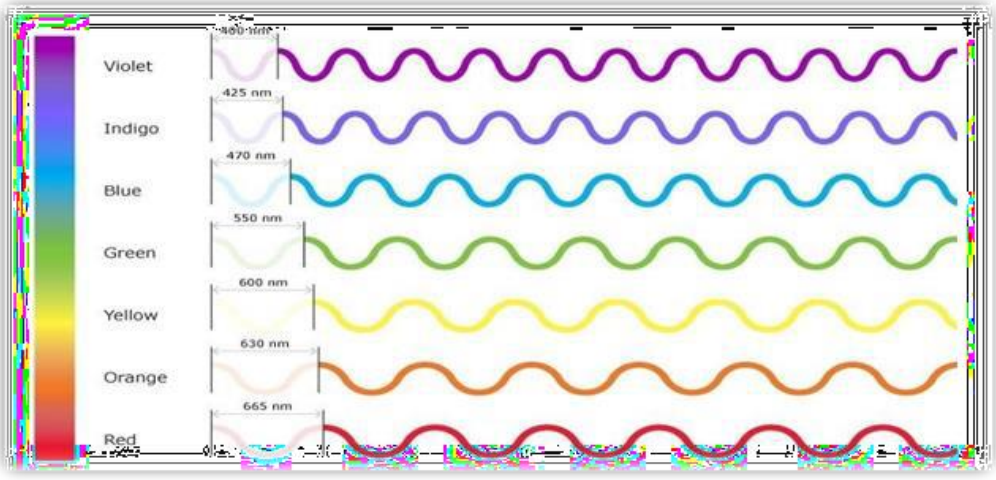
Spectrum Instruments and Uses

In clinical chemistry laboratory there is a continual need for the use of quantitative method one of the methods used most frequently in clinical chemistry is photometry.

A photometer: is a device used to measure the properties of light. Among other characteristics, the luminous intensity, color, luminous flux of a light source can be measured using a photometer.

Photometers measure light wavelengths and atomic emissions by collecting radiation emitted by the light source. Through a series of steps the photometer converts the radiation data input into a measurement called spectral irradiance. Photometers may be used to measure infrared or ultraviolet light.

The human eye respond to radiant energy or light with wave length between 380 - 750 nm which is called the visible light. **The visible light :** is the small part within the electromagnetic spectrum that human eyes are sensitive to and can detect. It is consist of the: Violet, blue, green ,yellow, orange, and red.



Al-Mustaqbal University College
Department of Medical Instrumentation Techniques Engineering
Class: second
Subject: Clinical Chemistry
Lecturer: Mustafa Fahad
Lecture: Two

-The following table shows the relation between the wave length and color absorbed:

Wave length	Name	Color absorbed
180-320	Short uv	Not visible
320-380	Long uv	Not visible
380-440	Visible	Blue
500-580	Visible	Green
580-600	Visible	Yellow
600-620	Visible	Orange
620-750	Visible	Red
750-2000	Short infrared	Not visible

The principle of Photometer:

It used to measure the color solutions, to obtain one type of wave lengths, to absorb unknown solution then measure its concentration. there are basic steps to achieve this purpose:

The solution must be colors ,if it not then should treat him chemically to be colors because the light intensity increase proportionally with solution concentration.

Passing single light through the solution to determine intensity light color, so the darkness color solution is more absorbance and less transmittance.

From this two steps will be achieve the proportional relation for three factors: Solution concentration , light intensity, transmittance light quantity.

Al-Mustaqbal University College
Department of Medical Instrumentation Techniques Engineering
Class: second
Subject: Clinical Chemistry
Lecturer: Mustafa Fahad
Lecture: Two

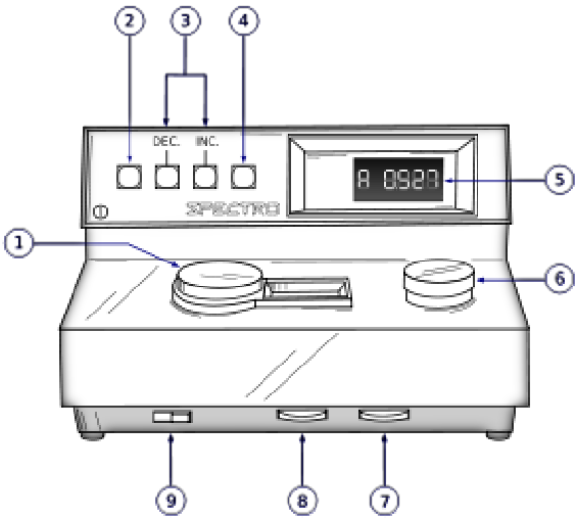
The main types of spectrum instrument:

- Colorimeter.
- Spectrophotometer.
- Flame photometer.

A colorimeter: is a device used for measuring colors, It measures the absorbance of different wavelengths of light in a solution.

The most important parts of a colorimeter are:

- a light source, which is usually an ordinary filament lamp
- an aperture (exit slit),used for colorimeter more accurate when the slit opening is small.
- a set of filters (optical filter) in different colors, used to select the wavelength of light which the solution absorbs the most
- a detector (photocell) that convert light intensity to electrical energy
- Cuvette : In a manual colorimeter the cuvettes are inserted and removed by hand, is made of quartz or plastic or glass.
- Output: The output of the colorimeter may be shown in graphs or tables, by an analogue or digital meter. The data may be printed on paper, or stored in a computer.



(1)Wavelength selection, (2) printer button , (3) concentration factor adjustment , (4) UV mode selector ,(Deuterium lamp) , (5) Readout, (6) Sample compartment, (7) Zero control (100% T), (8) Sensitivity switch, (9)ON/OFF switch.

Al-Mustaqbal University College
Department of Medical Instrumentation Techniques Engineering
Class: second
Subject: Clinical Chemistry
Lecturer: Mustafa Fahad
Lecture: Two

Spectrophotometer: is a device used to measure how much a chemical substance absorbs light as it passes through a sample. Not much different from the photometer, only **optical filters** are replaced by **prisms, diffraction grating** to generate monochromatic light beams

The basic principle of this method is that a different compound absorbs or transmits light over a certain range of wavelength. Depending on the range of wavelength of light source, spectrophotometers can be classified into ultraviolet- visible and infrared spectrophotometer.

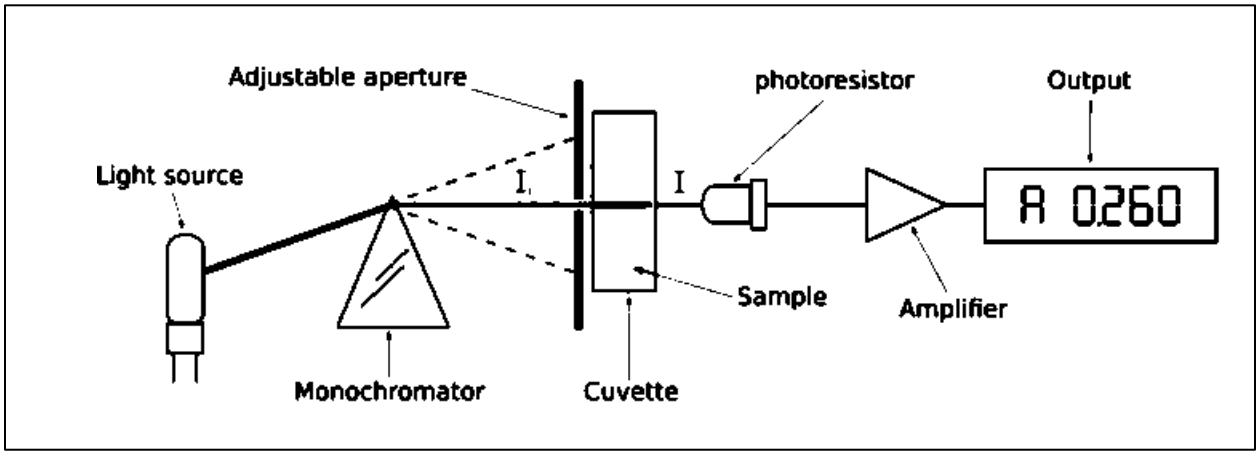
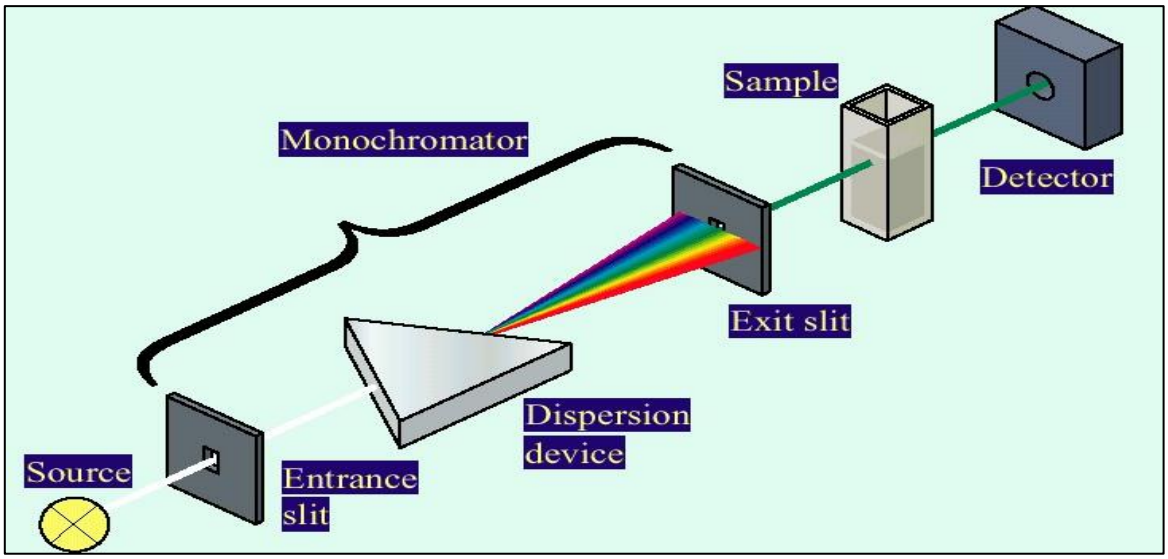


Fig (1) The components of spectrophotometer

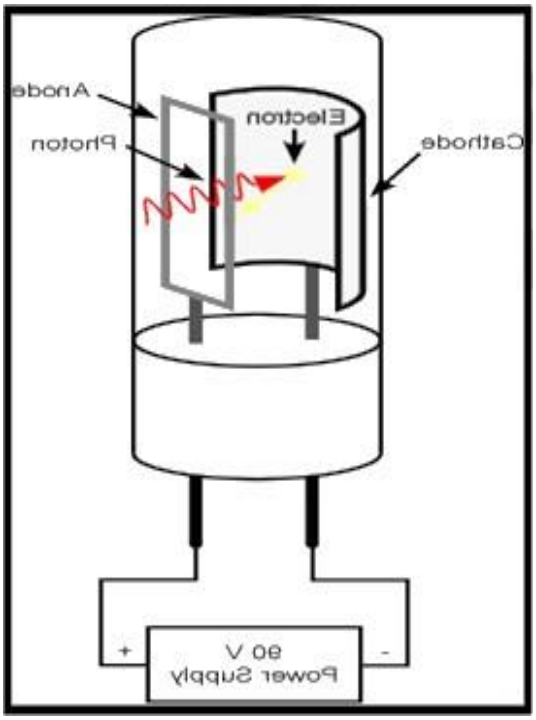


Al-Mustaqbal University College
Department of Medical Instrumentation Techniques Engineering
Class: second
Subject: Clinical Chemistry
Lecturer: Mustafa Fahad
Lecture: Two

- **Light source (lamp):** tungsten filament lamp emits light in the visible region (400-800 nm) , hydrogen lamp is used to get ultraviolet (340nm).
- **Monochromator:** a system for isolating the desired wave length of light band excluding that of other wave length this is may be filter (colored) thin glass which is transferred or prism or diffraction grating.
- **Slit:** this is to adjust the intensity of light emitted through the monochromator i.e. isolate a narrow beam of light and improve its chromatic purity.
- **Photocell:** it is to convert light energy into electrical energy ,the type of photocell is photomultiplier tube as shown below :

Photomultiplier tube

The diagram shows a photocell circuit. The principle depend on:



The cathode is made of photosensitive material (e.g. semiconductor (selenium)). When a beam of light falls on the cathode, photoelectrons are ejected out and are attracted by the anode generating electric field. Thereby sending a current through the circuit that is measure by the galvanometer connected in the circuit.

Al-Mustaqbal University College
Department of Medical Instrumentation Techniques Engineering
Class: second
Subject: Clinical Chemistry
Lecturer: Mustafa Fahad
Lecture: Two

WAVELENGTH SELECTION

There are several options open to the manufacturer of a colorimeter when deciding how to select the wavelength i.e. produce monochromatic radiation (one wavelength band) from polychromatic radiation (white light). These basic options are:

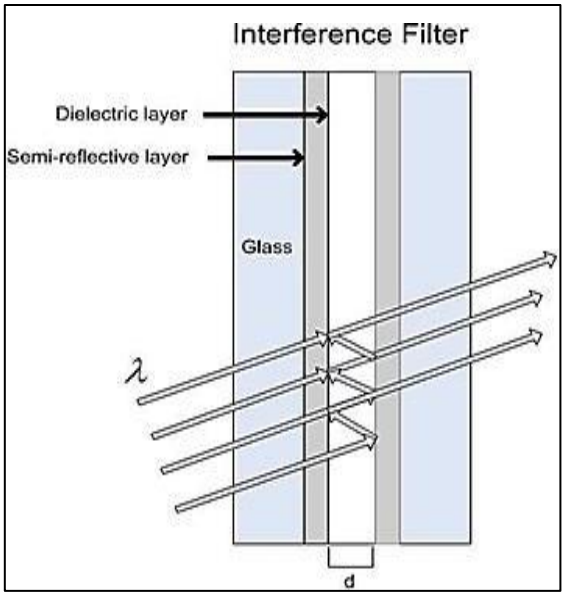
- Gelatin filter
- Glass filter
- Interference filter
- Diffraction grating
- Prism.

1. Gelatin Filters : These are low cost selection devices which produce or transmit a wide band of radiation usually ą 20 nm. The most common type of gelatin filter is constructed by sandwiching a thin layer of dyed gelatin of the desired color between two thin glass plates. They absorb approximately 30-40% of all incident radiation there by reducing energy throughput to the detector.

2. Glass Filters : Colored glass filters are now more or less historical selection devices in colorimeters and have very wide band posses often up to 150nm. Specific wavelengths can however be achieved by using a combination of glass filters.)

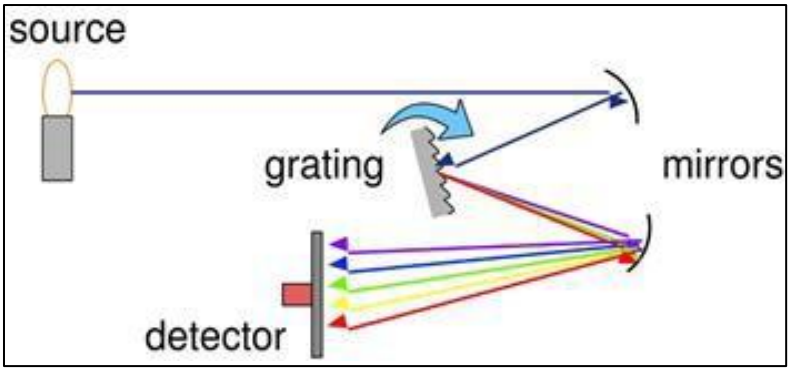
Al-Mustaqbal University College
Department of Medical Instrumentation Techniques Engineering
Class: second
Subject: Clinical Chemistry
Lecturer: Mustafa Fahad
Lecture: Two

3. Interference Filters



These are used to select wavelengths more accurately by providing a narrow band pass typically of around 10nm. The interference filter also only absorbs approximately 10% of the incident radiation over the whole spectrum thereby allowing light of higher intensity to reach the detector.

4. **Diffraction grating:** In optics, a diffraction grating is an optical component with a periodic structure that diffracts light into several beams travelling in different directions (i.e., different diffraction angles). The emerging coloration is a form of structural coloration.



5. **Prism:** An optical prism is a transparent optical element with flat, polished surfaces that refract light. Typical prism materials include glass, acrylic and fluorite.