الجامعة التقنية الشمالية
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الاجهزة

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Photometer

It is instrument measure the absorbance optical density. This equipment will measure the transmission and optical density depending on color filter which give the complement color and in this case the reading less accurate than spectrophotometer which is use monochrometer.

Visible light spectrum:

When a beam of light passes through a colored solution, it interacts with matters in the solution and be refraction, absorption and transmission among others .

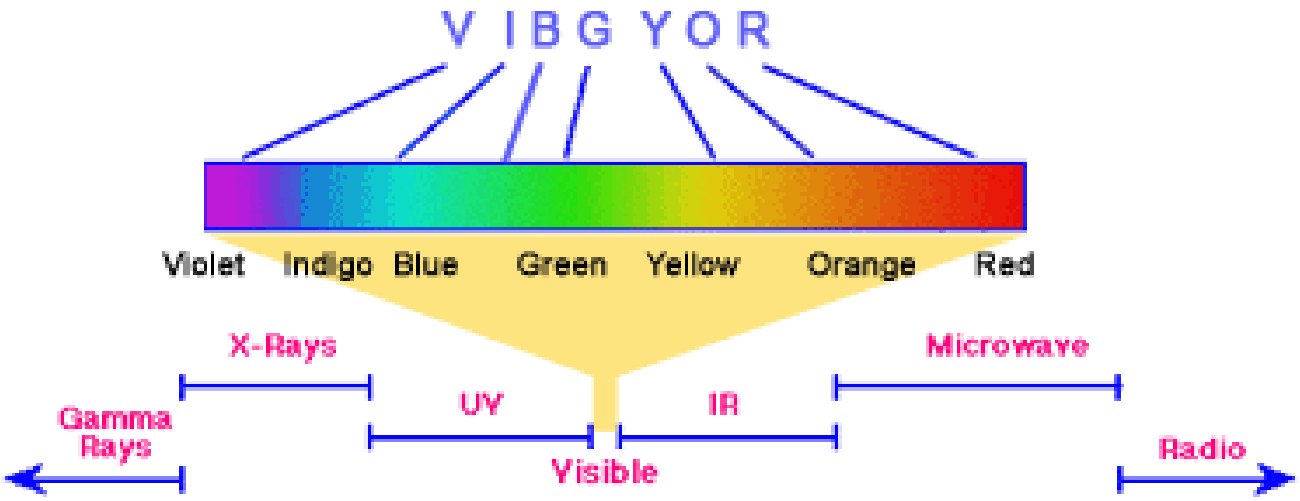
Refraction : Is defined as sudden change in the direction of the beam when the light passes from one medium to other with a different physical density .

Reflection : Is a situation where some components of the light (colors) are retained or absorbed.

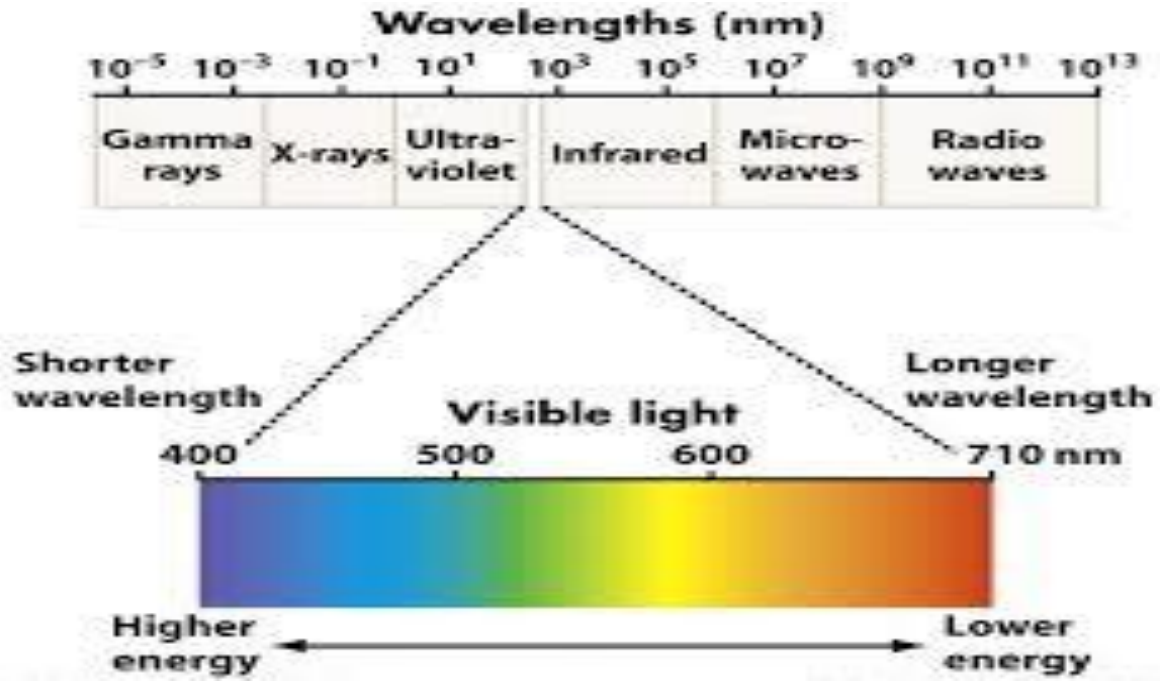
Transmission : Refers to the situation where some portions of the light permitted to pass through a given medium .

*Radiation is characterized by waves on which basis the electromagnetic radiation spectrum could be divided in many regions including gamma rays , x-rays, ultraviolet rays, visible light , infrared , microwaves and radio waves .

*Visible region is the radiant energy to which the human eye responds and their wavelength varies between (400-700) nm, wavelength of about 700nm are seen by the eyes as red colors while those of progressively shorter wavelengths give in descending order to orange ,yellow, green , blue, indigo and finally violet colors which is produced in the short wavelength of 400nm.



Visible light spectrum



Electromagnetic spectrum

Beer's and Lambert's Law:

Most colorimetric analytical tests are based on the Beer's - Lambert's law which states that under the correct conditions the absorbance of a solution when measured at the appropriate wavelength is directly proportional to its concentration and the length of the light path through the solution.

Using a standard, this law can be applied to measuring the concentration of a substance in unknown (test) solution by using the formula.

$$\textbf{Concentration of test } (C_t) = \frac{\text{Absorbance of test (At)}}{\text{Absorbance of standard (As)}} \text{X Concentration of standard (Cs)}$$

Types of filter:

1-Blue filter : it will pass the wavelength between (400 – 495) nm.

2-Green filter : it will pass the wavelength between(500 – 580) nm.

3-Red filter : it will pass the wavelength between (600 – 800) nm.

Photometer device

Spectrophotometer

It is an instrument that measures the amount of photons (intensity of light) absorbed after it passes through sample solution.

In the spectrophotometer, the concentrations of a known chemical substance can be determined by measuring the intensity of light detected depending on the range of wavelength of light source.

Types of Spectrophotometer:

1-UV-visible spectrophotometer: uses light of the ultraviolet range

(185- 400 nm) and visible range (400-700nm) of electromagnetic radiation spectrum.

2-IR spectrophotometer: uses light of the infrared range

(700 - 15000 nm) of electromagnetic radiation spectrum.

Types of photocell:

1-Red photocell (600 –800) nm.

2-Blue photocells (400 – 495)nm.

Source of light:

1-Tungsten filament: The most commonly used

source for visible light ranging from (400 – 700) nm.

2-Halogen lamp: It will give more bright light with

minimum of the red and has a long life in use.

3-Deuterium lamp: It will use in ultraviolet spectra

measurement ranging(190 – 400) nm .

It is use only in spectrophotometer

Diffraction Grating:

It is an optical device consisting of many closely spaced parallel slits or grooves. In a transmission type of grating, light passes through the narrow transparent slits that lie between the dark lines on a glass or plastic plate. In a reflecting grating, light is reflected by the many parallel, narrow, smooth surfaces and absorbed or scattered by the lines cut in the reflecting surface of the grating .A diffraction grating does not bend anything. It shifts the position of wave crests so that they add together at different angles.

Prism:

It is An object made up of a transparent material like glass or plastic that has at least two flat surfaces that form an acute angle (less than 90 degrees).White light is comprised of all the colors of the rainbow. When white light is passed through a prism, the colors of the rainbow emerge from the prism (it changes the speed of light differently for different colors -it bends the light differently for the different colors).

Photometer	Spectrophotometer
1-It use filter which give approximate a wavelength according to the color	It use prism or 1-grating(monochrometer) which give exact wavelength
2- Lower cost	2-More expensive, but may be necessary
3-Smaller number of calibrations	3- Unlimited number of calibrations
4- Fixed applications	4-Complete spectral coverage accommodates new processes
5- Simpler chemistries	5-More complex chemistries
6-Photometers are “tuned” for a specific application	6-Enhanced spectral processing
7-Do not require in-house chemometrics (PLS) expertise	7-Generally requires chemometrics (PLS) expertise within the company
8-They will not work for other applications that require different wavelengths unless modified	

Balance:

Is essential laboratory instruments that are widely used for determining weight of various substances (powders ,crystals and chemical materials) in the laboratory for used to prepare reagents, stains and culture media , balances are required to weight accurately within the needed range.

The balance should be kept clean and located in an area away from:

- large pieces of electrical equipment.

-open windows to minimize any vibration as interference that may happen.

Types of balance:

1-Beam Balance: This type of balance uses a comparison technique in the form of a beam from which a weighing pan and scale pan are suspended. The object to be weighed is placed on the measuring pan, and standard weights are added to the scale pan until the beam is in equilibrium.

2-Analytical Balance: It is used to measure mass to a very high degree of precision. The weighing pans are inside see-through enclosure with doors so that dust does not collect and so any air currents in the room do not affect the delicate balance.

These balances are used :

1-To weigh small quantities usually in milligram (mg) range.

2-When great accuracy is required.

Types of Analytical Balance:

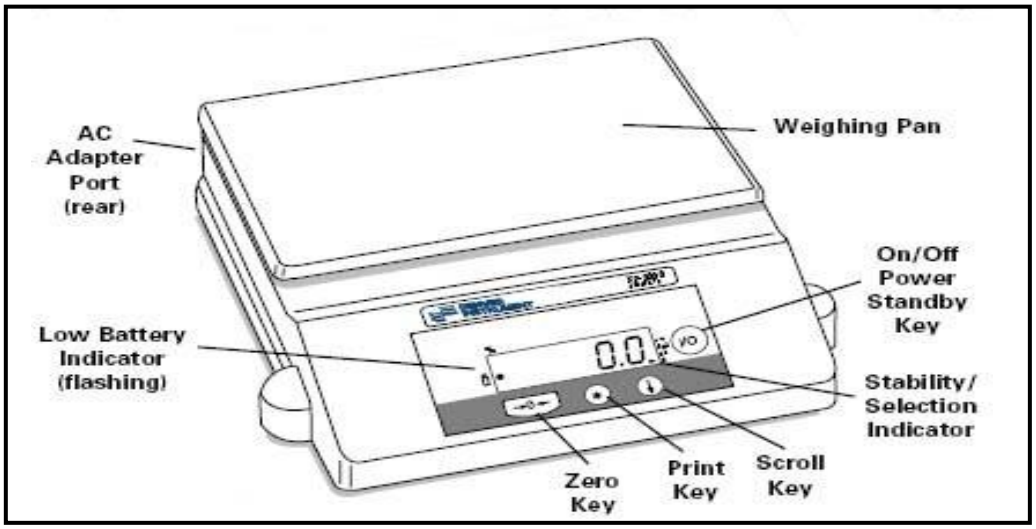
A- Single-Pan Mechanical Balance: These consist of abeam with two

knife-edges, one to support the weighing pan and the other acting as a pivot fixed counterweight balances the load on the pan.

B-Two-Pan Analytical Balance: These balances consist of a symmetrical beam and three knife-edges. The two terminal knives support the pans and a central knife-edge acts as a pivot about which the beam swings.

C-Electronic Single-Pan Balance: These are top loading balances with the applied load being measured by an electromagnetic force unit or a strain gauged load cell. The mass of the load is proportional to the current needed to balance it. Single-pan electronic balances give a direct reading of the mass applied.

D-Microbalance: This type of analytical balance is capable of measuring samples to at least (1) million parts of a gram. The more sensitive quartz crystal microbalance (QCM) measures mass by measuring the change in frequency of a piezoelectric quartz crystal.



Microbalance

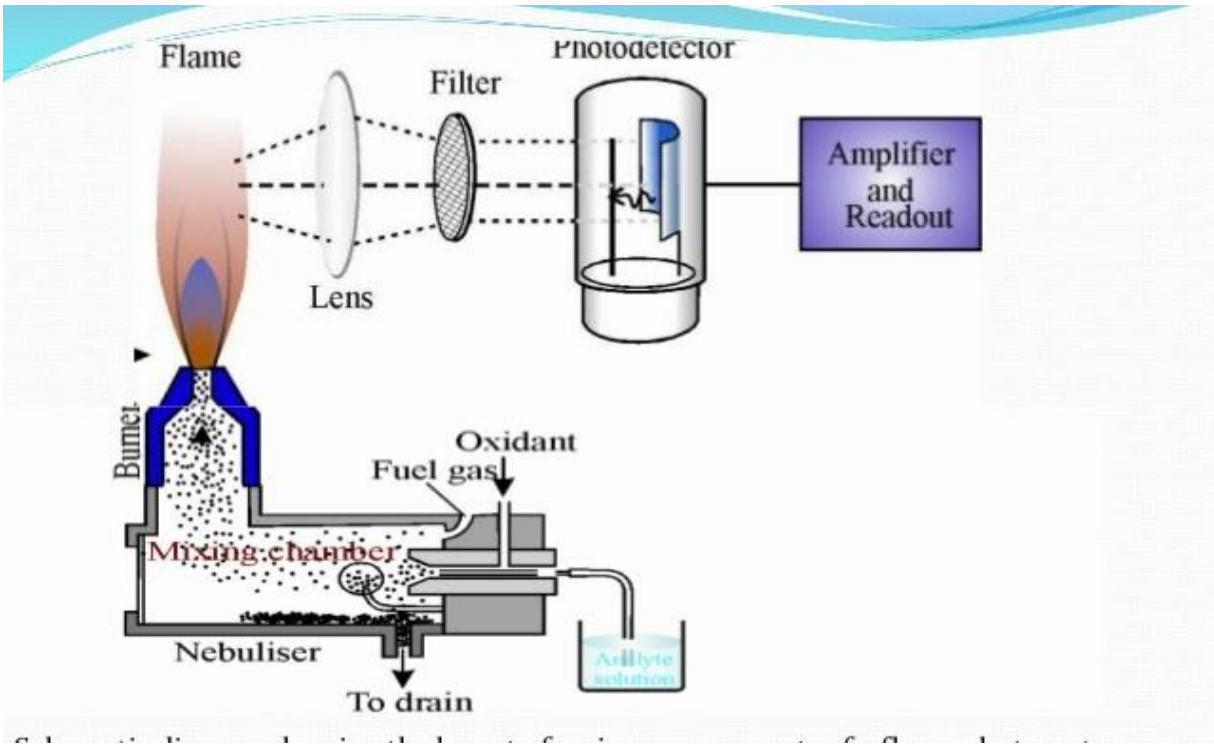
Flame photometer:

It a device used for the determination of electrolytes in a given solution. It is most commonly used for the quantitative analysis of sodium (Na) and potassium (K) ions in body fluids. It used for measuring the wavelength specific for (K, Na) by special filter depending on intensity and emission light.

There are two types of atomizers burner:

1-Total consumption burner: It is draws the solution directly into the base of small flame where it is vaporized burned and locally droplets of fluid to the flame .

2-A premix burner: The large droplets full the floor of the chamber and only the rare fine droplets are carried up word into the flame by the flow of gas.



Component of flame photometer

Centrifuges

It is a device for separating two or more substances from each other by using centrifugal force. Centrifugal force is the tendency of an object traveling around a central point to continue in a linear motion and fly away from that central point.

Centrifugation can be used to separate substances from each other because materials with different masses experience different centrifugal forces when traveling at the same velocity and at the same distance from the common center.

Principles work of centrifuges:

The basic physics work of centrifuges is gravity and generation of the centrifugal force to sediment different fraction.

centrifugal field (G) depend on: 1-Angular velocity (w) in radians / sec.

2- Radial distance (r in cm) of particle from axis of rotation.

$$G = w^2 r$$

Rate of sedimentation depends on:

-Mass of particle (density and volume).

-Density of medium.

-Shape of particle.

-Friction.

Types of centrifuges:

There are many types of centrifuges, but the basic principle is the same

all use centrifugal force.

1-Low speed centrifuges:

-It is very simple and small.

-maximum speed of 3000 rpm.

-Don't has any temperature regulation system.

-Used normally to collect rapidly sedimenting substance such as blood cell.

Pic. 2

Head of centrifuge ***Centrifuge drive***

2-High speed centrifuges:

-Maximum speed of 25000 rpm.

-Temperature maintained at 0 – 4 ^{0}C by thermocouple.

-Used to collect microorganism, cell, large cellular organelles.

-Useful in isolating sub cellular organelles (nuclei , mitochondria and lysosomes).

3-Ultracentrifuges:

-Operate at speed of 75000 rpm.

-Rotor chamber is sealed and evacuated by pump to attain vacuum.

-Refrigeration system temperature 0 – 4 ^{0}C.

There are two types of ultracentrifuge:

A- Analytical centrifuge.

B- Preparative centrifuge.

Applications of centrifuges:

1-Remove cellular elements from blood to provide cell free plasma or serum for analysis.

2 -Isolate chemical precipitated protein from an analytical specimen.

3 -Separate protein bound from free ligand in immunochemical.

4-Separate lipid components .

pH Meter:

It is a scientific instrument that measures the hydrogen-ion concentration in a solution indicating its acidity or alkalinity expressed as pH . pH is the unit that describes the degree of acidity or alkalinity. It is measured on a scale of 0 to 14.

*The pH value of a substance is directly related to the ratio of the hydrogen ion [H^+] and the hydroxyl ion [OH^-] concentrations.

If the H^+ concentration is greater than OH^-, the material is acidic

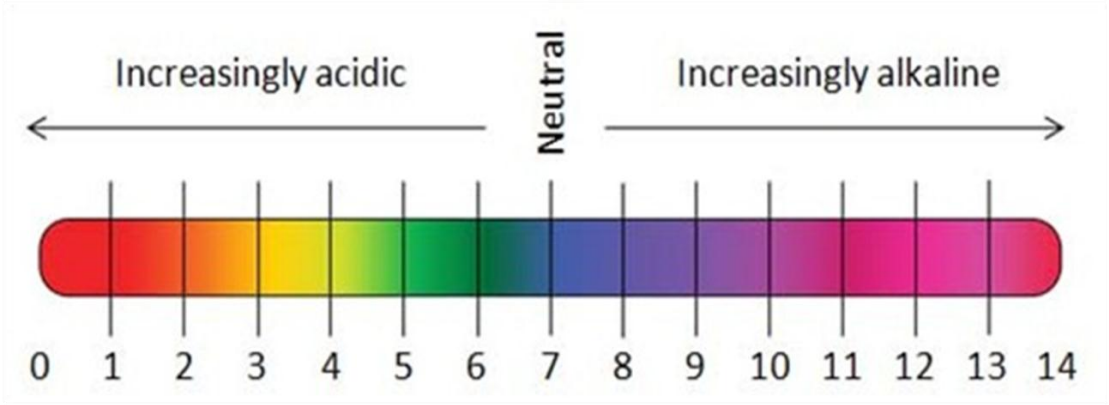
(pH value is less than 7).

*if the OH^- concentration is greater than H^+, the material is basic

(pH value greater than 7).

*if equal amounts of H^+ and OH^- ions are present, the material is neutral (pH of 7).

Acids and bases have free hydrogen and hydroxyl ions, respectively. The term pH is derived from (p) the mathematical symbol for negative logarithm and (H) the chemical symbol for Hydrogen.



The pH Scale

Types of pH meters:

1-Manual pH meter.

2-Digital pH meter.

Applications of pH meter:

1-know the acidity of the water.

2-Chemical laboratory analyses.

3-Soil measurements in agriculture.

4-Water quality for municipal water supplies.

5-Environmental remediation.

6-Brewing of wine or beer.

7-Healthcare and clinical applications such as blood chemistry.

Microtome:

is It an instrument which is used for cutting of materials in different thickness that are used as microscope slides, allowing samples to be observed under transmitted light or electron radiation.

Microtome is a method for the preparation of thin sections of materials such as bones, minerals and teeth for thorough examination. Microtome sections can be made thin enough with section thickness between 50 nm and 100 µm.

Types of Microtome :

There are several types of microtome each designed for a specific purpose although many have a functional role , there are basic types of microtome are named according to the machine as following:

1-Rotary microtome : It is an excellent machine for research and is valuable from the preparation of serial section.

2-Freezing microtome : It is using for cutting section when:

1-Speed of the utmost in portances .

2-When it is required to demonstrate fat histological.

3-When neurological structure are to be studies.

3-Ultra microtome.

4-Laser microtome.

5-Saw microtome : It is used for hard material such as teeth or bones.

6-Vibrating microtome : It used for difficult biological samples.

7-Sledge microtome.

8-cryo microtome.

Types of knives according to material that made of :

1-Steel blades : There are used to prepare sections of animal or plant tissues for light microscopy histology.

4-Gem quality diamond knives : are used for slicing thin sections for electron microscopy.

3-Diamond knives : are used to slice hard materials such as bone, teeth and plant matter for both light microscopy and for electron microscopy.

2-Glass knives : are used to slice sections for light microscopy and to slice very thin sections for electron microscopy.

Uses of microtomes:

1-Fix the paraffin block on block holder.

2-Rotate the operating handle and close the knife.

3-The section cutting effected by the vertical and fall

of the object and fix the knife edge.

Microtome drive

Microscopes:

Is an important device that produces a magnified image of objects too small to be seen with the naked eye .The microscope is widely used in medicine and bology.

Types of microscope:

1-Light microscope : The types of light microscope including:

A- Bright - field microscope: It used to view stained or naturally pigmented specimens .The name "bright field" is derived from the fact that the specimen is dark and contrasted by the surrounding bright viewing field. Simple light microscopes are sometimes referred to as bright field microscopes.

B- Dark - field microscopy: is used to illuminate unstained samples causing them to appear brightly lit against a dark background. This type of microscope contains a special condenser that scatters light and causes it to reflect off the specimen at an angle.

C -Ultraviolet microscope: It has quartz lenses and slides and uses ultraviolet radiation as the illumination. The use of shorter wavelengths than the visible range enables the instrument to resolve smaller objects and to provide greater magnification than the normal optical microscope.

D -Fluorescent microscope : It used to examine material that fluoresces under ultraviolet light. Fluorescence microscopy is based on the principle that fluorescent materials emit visible light when they are irradiated with ultraviolet rays or with violet-blue visible rays.

E - Phase contrast microscope:

Transparent microorganisms suspended in a fluid may be difficult and sometimes impossible to see. One method of making them more visible is to use phase contrast.

Phase contrast is useful for examining:

•Unstained bacteria : cholera vibrios in specimens and cultures.

•Amoebae in faecal preparations.

Cerebrospinal fluid, lymph gland fluid.

• Urine sediments.

2-Electronic microscope:These types are:

A- Scanning electron microscope (SEM): This microscope helps in viewing three-dimensional images of microorganisms and other specimens. Gold and palladium is used to stain the specimens mounted on a scanning electron microscope.

B-Transmission electron microscope (TEM) : is used to study cells.

Ultrathin slices of microorganisms like viruses are placed on a wire grid, then these cells are stained with gold or palladium and then used to observe under a transmission electron microscope. The electron beam is deflected on the densely coated parts of the cells and the image is observed on dark and light background.

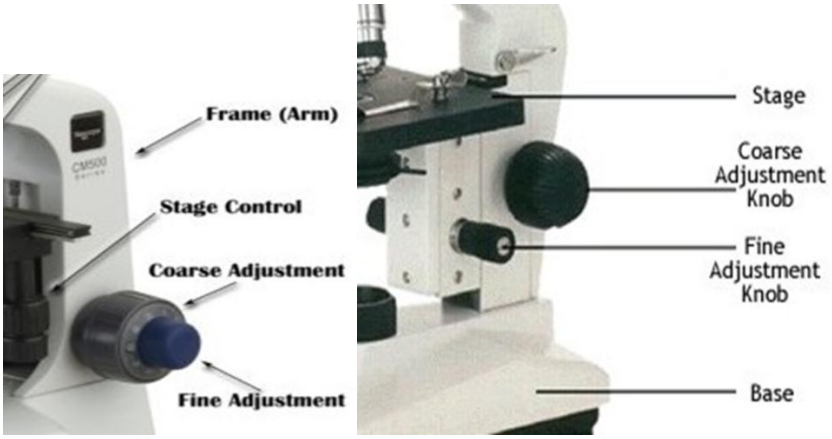
Parts of Microscope:

A- Frame work.

B- Mechanical adjustments (Focusing system):

2-Fine adjustment : This knob is a sub part of the Coarse adjustment knob. It is used to bring the specimen into sharp focus.

1-Coarse adjustment : is a knob present on the arm of a microscope. The main function of this knob is to move the specimen back or forth to adjust the slide containing specimen in order to bring it to focus and show the best image possible. The coarse adjustment should be carefully moved and adjusted to attain desired results.



3-Slide adjustment:

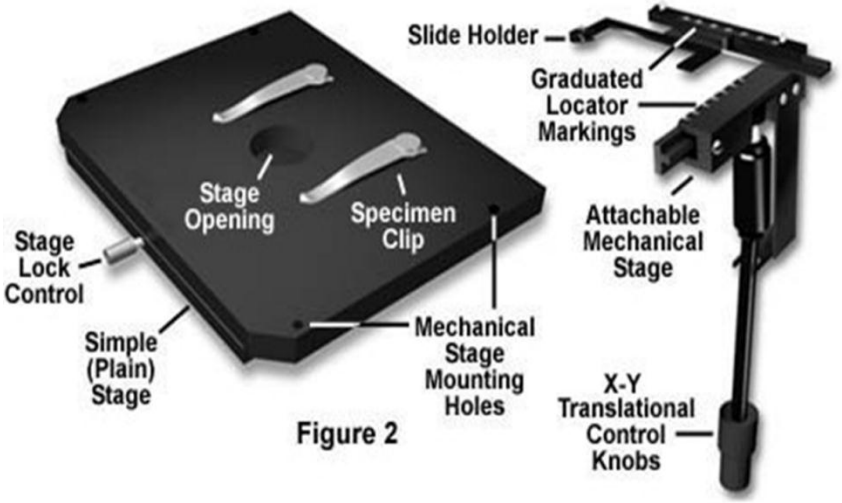
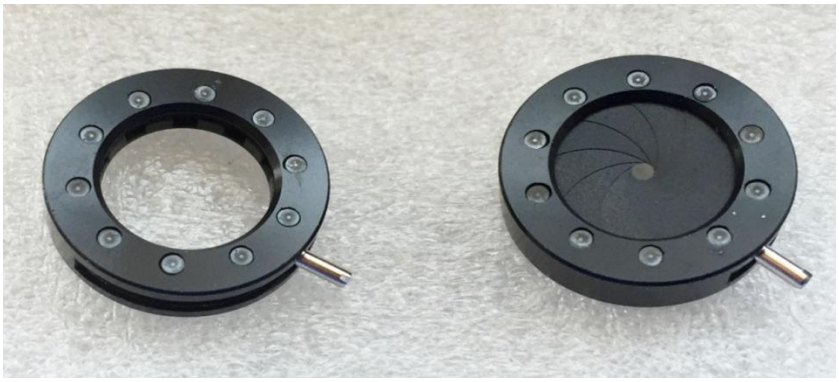


Figure 2

*4-Condenser adjustment :*The condenser is used to condense the light required for visualization. The condenser aperture is adjusted by the iris diaphragm ,which is found just below the condenser.



C- Magnification system (Lenses):

1-Eyepiece (Ocular lens): is a magnifying lens attached to the microscope which helps in magnifying the sample object. It is called an eyepiece as we need to place our eye near it in order to see the magnifying image of the sample Eyepiece may be monocular or binocuar. Eye lenses magnification 10X , 12X.

2- Objective lens: Is the part of microscope responsible for magnifying the image of spicemen . Thar are four Objective lens in a standard microscope as following:

Scanning Objective (4X): This shortest objective is useful for getting an overview of the slide (especially handy with some of the slides that contain whole organs like a section of the spinal cord, lung, digestive tract, ovary.....).

2-Low Power Objective (10X): This next shortest objective is probably the most useful lens for viewing slide.

3-High Power Objective (40X): This objective (sometimes called the "high-dry" objective) is useful for observing fine detail such as the striations in skeletal muscle, the arrangement of Haversian systems in compact bone, types of nerve cells in the retina, etc.

4-Oil Immersion Objective (100X): This longest objective is used for observing the detail of individual cells such as white blood cells, the cells involved in spermatogenesis, etc….. . The lens must be used wit a specially formulated oil that creates a bridge between the tip of the objective and the cover slip. Since the refractive indices of air and this lens are different the oil immersion using to increase the refractive index to make the field clear.

The product magnification depend on:

1-Mechanical tube length.

2-Focal length of objective lenses.

3-Magnifying power of eye lenses.

Magnification of Microscope = Magnification of objective lens x Magnification of eye lens

$$\text{=Magnification of objective lens } \frac{Mechanical\ tube\ length}{Focal\ length\ of\ objective}$$

Focal length : is the distance between it's center and point where parallel rays of light is brought to focus.

D- illumination system:

1-Condenser and iris: Condenser is a large lens with an iris diaphragm , the condenser lens receives a beam from the light source and passes it into the objective. The iris is a mechanical device mounted underneath the condenser and controls the amount of light entering the condenser.

2-Mirror :Is situated below the condenser and iris ,it reflects the beam of light from the light source up wards through the iris into the condenser. The mirror is used to reflect ray or electrical light.

3-Sources of illumination:

Day light : is enough for oil immersion work.

Electric light.

Condenser of microscope:

It is a lens that used to concentrate light from the illumination source that is in turn focused through the object and magnified by the objective lens.

There are three types of condebser:

1-Abb condenser: It has two controls:

*Moves the Abb condenser closer to or further from the stage.

*The iris diaphragm ,which controls the diameter of the beam of light.

A diaphragm: is a thin opaque structure with an opening (aperture) at its center Abb condensers are mainly vital for magnifications of above 400X. It is considered as the simplest type.

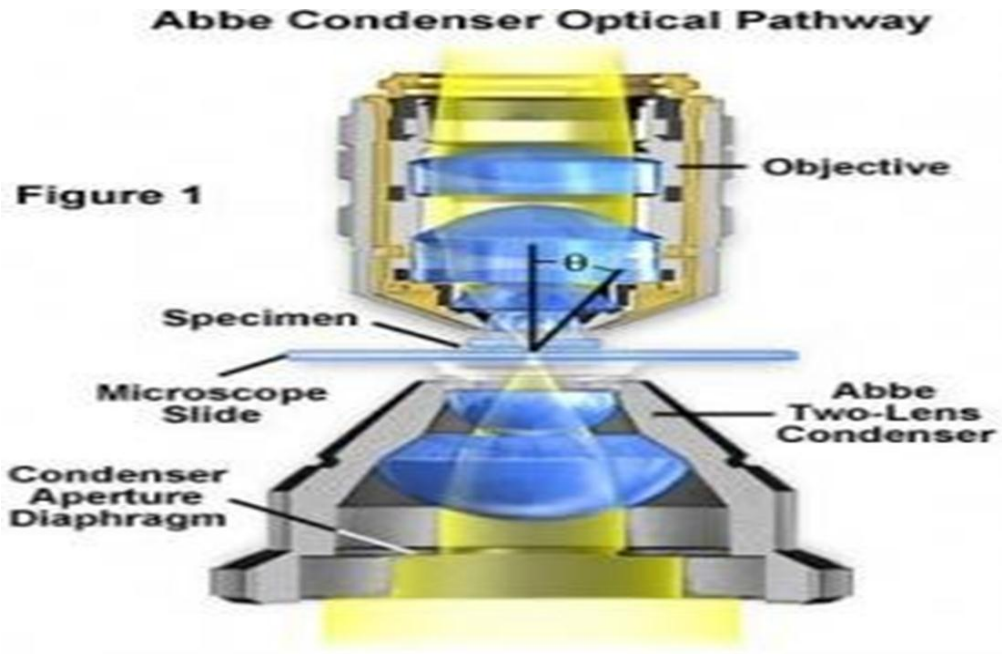


Figure 1

2-Aplanatic condenser: It corrects for spherical aberration in the concentrated light path , spherical aberration is an optical effect observed in an optical device (lens , mirror) that occurs due to the increased when refraction of light rays when they strike a lens or a reflection of light rays they strike a mirror near its edge in comparison with those that strike nearer the center. Plan lenses can reduce or eliminate spherical aberration.

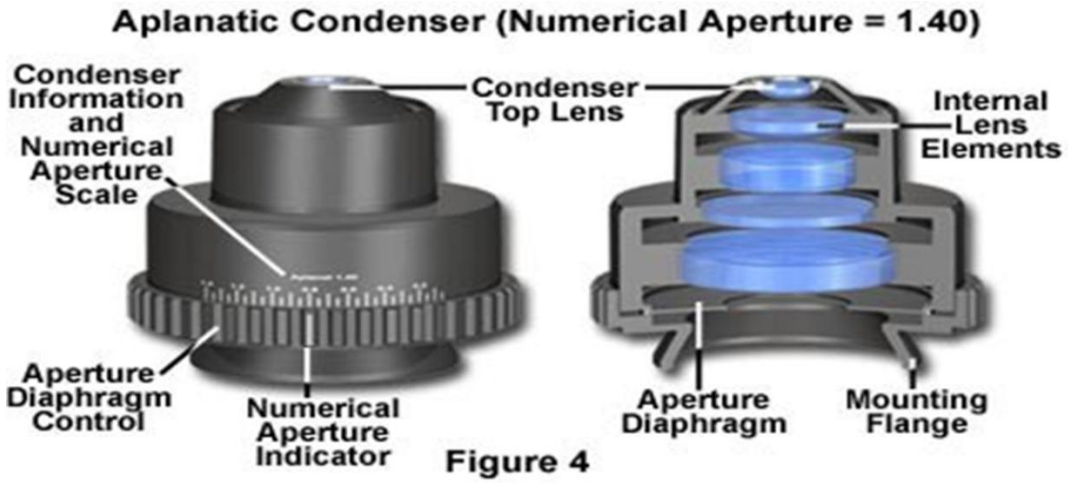
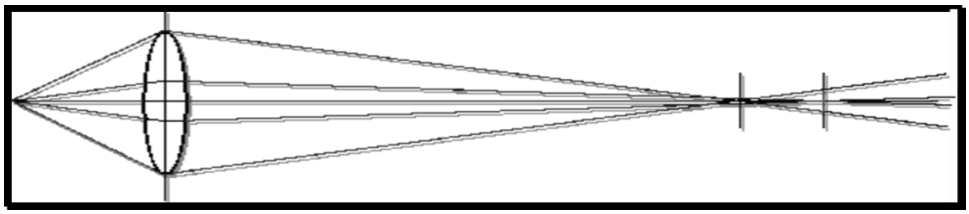


Figure 4

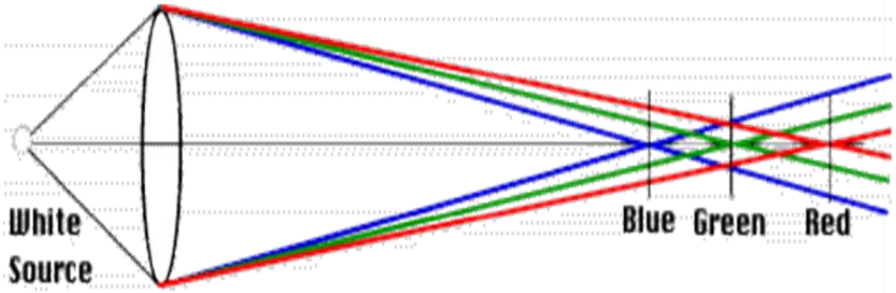


Spherical aberration

3-Achromatic condenser: There are corrected for chromatic aberrations. Achromatic lens is a lens that is designed to limit the effects of chromatic and spherical aberration. Chromatic aberration is a type of distortion in which there is a failure of a lens to focus all colors to the same convergence point.

**Chromatic aberration occurs due to the variation of refractive index with wavelength for a lens material.

**This wavelength dependence results in slightly different focal lengths for different wavelengths of light. Compound lenses, called achromats, can reduce or eliminate chromatic aberration because the components are chosen such that the variation in refractive index as a function of wavelength cancels out.



Chromatic aberration

Numerical Aperture (N.A.):

(n sine θ) and is related to the angular aperture of the lens and the index of refraction of the medium found between the lens and the specimen. The physical size of the lens is important in determining the N.A. of the lens.

in an object being observed. It is derived by a mathematical formul

This is a number that expresses the ability of a lens to resolve fine detail

$NA = N \sin \theta$

Where:

n : is the index of refraction of the medium in which the lens is working.

(1.00 for air, 1.33 for pure water and 1.52 for immersion oil).

θ :is the maximal half-angle of the cone of light that can enter or exit the lens.

Resolution power (Resolving) : It is a capacity to distinguish two adjacent point depend on:

1-numerical aperture of lens.

2-wave length of used light.

Note:

*The human eye can separate dots (0.25 mm apart).

*The light microscope can separate dots (0.25 micron apart).

*The electron microscope can separate dotes less than angstrom.