

At the end of this Lecture, students should be able to:

1. Know a brief history of the microscope
2. Describe the working principle of the microscope
3. Understand the theoretical Principles of Microscopy
4. Identify the essential parts of a microscope.
5. Know the different types of microscope and their applications.
6. Correctly use a microscope
7. Take care of and maintain a microscope.

INTRODUCTION

Microscopy is the technical field of using microscopes to view samples and areas of objects that cannot be seen with the un-aided eye (i.e., objects that are not within the resolution range of the normal eye).

Microbiology owes its existence to Antonie van Leeuwenhoek. In 1673, with the aid of a crude microscope consisting of a biconcave lens enclosed in two metal plates, Leeuwenhoek introduced the world to the existence of microbial forms of life with his observation of the bee's mouth parts and stings, and later in 1676, the single-celled organisms (Animacules – small animals). Over the years, microscopes have evolved from the simple, single-lens instrument of Leeuwenhoek, with a magnification of about 275X, to the present-day electron microscopes and Scanning probe Microscopes capable of magnifications up to 1,000,000X and 100,000,000X respectively. Microscopes are broadly classified under light microscopes, electron microscopes and others. Light microscope uses visible light or ultraviolet rays to illuminate specimens. They include bright-field, dark-field, phase-contrast, and fluorescent microscopes. Fluorescent microscopes use ultraviolet radiations whose wavelengths are shorter than those of visible light and are not directly perceptible to the human eye. Electron microscopes use beams of electron instead of light rays, and magnets instead of glass lenses to observe sub-microscopic particles. Other microscopes use different other technique. They include: Confocal microscopes – use laser for illumination, Acoustic microscopes – use sound waves, Scanning probe microscopes – use probe, X-Ray microscopes – use X-rays, etc. It is also noteworthy that many optical microscopes produced these days send their images to computer screens rather than an eyepiece. Hence they are often referred to as **digital microscopes**; but the light source and interior parts of the



microscope are still the same. These have become much more popular over the years, because it's way easier to look at a large screen than a tiny eyepiece.

WORKING PRINCIPLE OF THE MICROSCOPE:

Microbiology is a science that studies living organisms that are too small to be seen with the unaided eye. Therefore, such a study must involve the use of a good compound microscope. Although there are many types and variations, they all fundamentally consist of a two-lens system, a variable but controllable light source, and mechanical adjustable parts for determining focal length between the lenses and specimen. They also work on the same principle as the conventional microscope (Fig.1.0).

In the light microscope, light rays which have passed through the specimen are transmitted through two sets of lenses, the objective, which is nearest to the specimen, and the eyepiece, which is further away from the specimen. The magnified image of the specimen is first produced by the objective. This is known as the primary image. The eyepiece then magnifies the primary image into the final one that is seen by the observer. The total magnification obtainable by the microscope is the product of the magnification of the objective and that of the eyepiece.

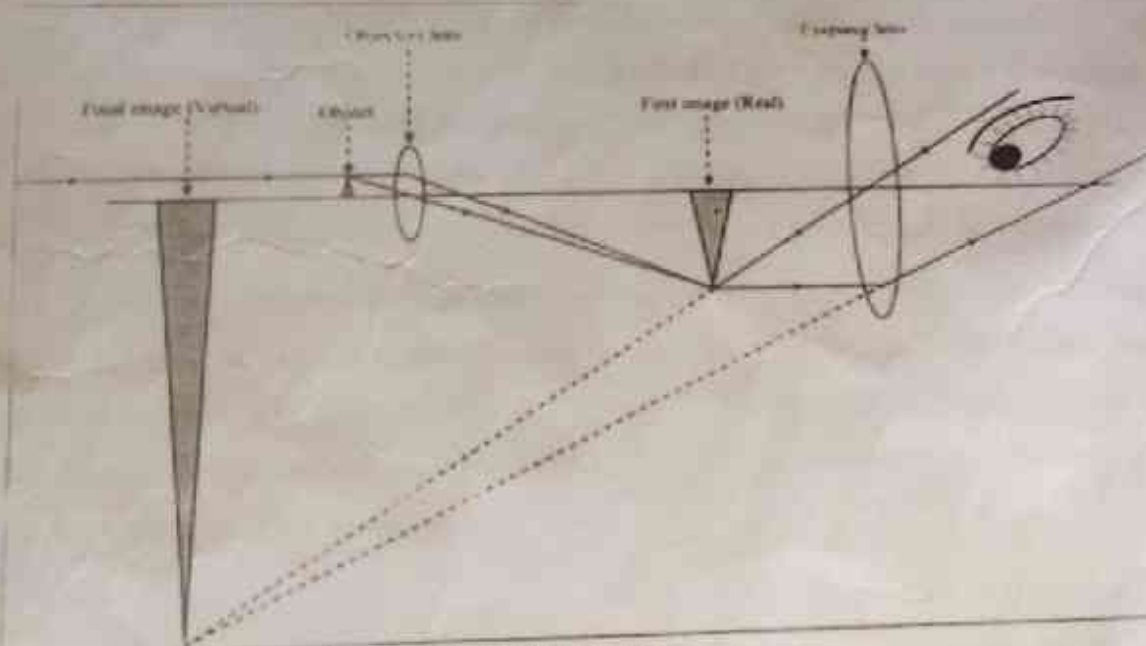


Fig.1.0: Ray diagram of a compound Microscope

MAIN PARTS OF A MICROSCOPE

The conventional microscope consists basically of the optical parts and the mechanical parts (which carries the optical parts/illuminating apparatus) [fig.2.0, fig. 2.1].

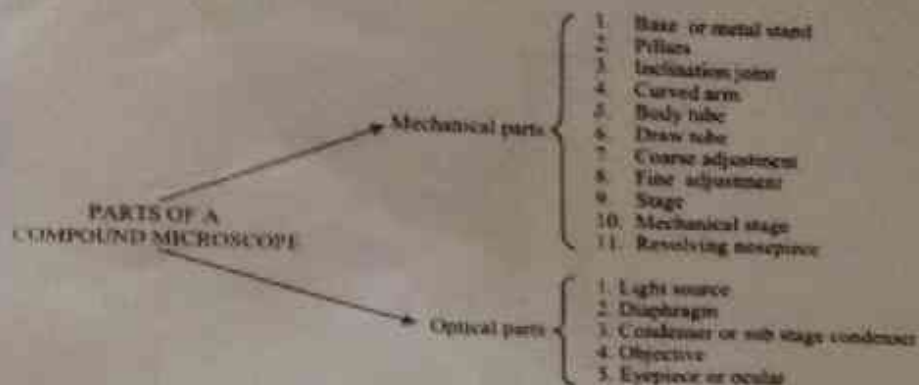


Fig. 2.0: Parts of a compound microscope

The illuminating apparatus

The illuminating apparatus comprises a light source or mirror having plane and concave sides, a condenser below the stage (sub-stage condenser), and an iris diaphragm by which to regulate the amount of light passing up from the light source/mirror. The condenser lens and iris diaphragm are held in place by the sub-stage. The sub-stage can also serve to fit the light filter attachments. The condenser lens can be moved up and down by rotating a knob usually located on the side of the body of the microscope. If the condenser is a lens of the moveable type, it is equipped with two screws that serve to centre the condenser lens. However, the condensers of most modern microscopes are pre-centred and do not require centering. The purpose of the condenser is to increase the illumination and to bring the rays of light from a wide angle to bear on the object.

Always use the condenser focused, and never use the condenser with the concave mirror. The lenses of the condenser are adjusted to give optimum illumination only when focused with light or the flat mirror. The concave mirror is only used for illumination when the condenser is not in use. However, this method of illumination without using the condenser is, for most purposes, inferior to that using the condenser.

Illuminating apparatus

Stage

The tube

The objective

Eye piece

Low power objective x10
High dry objective x40
Oil immersion objective x100
Scanning objective x4

The stage

The stage is a broad flat surface fixed to the body, and on it are placed the objects, usually mounted on slides, which are to be examined. The stage, which is usually rectangular in shape, has a central aperture. The aperture serves as a passage for light from the condenser lens to the objectives. The stage is also used to support the glass slide with the specimen to be examined. The stage can be surmounted by an apparatus, called the mechanical stage. The mechanical stage has a clamp or clamps and spring lever attached to it. The clamps, on both sides of the stage, are used to hold the slide in place. The mechanical stage is equipped with two additional knobs to move the slide either to the left and right or backwards and forwards to line the specimen with the objective lens. The mechanical stage also has vernier scales to help locate a specific field.

The tube

The tube, which may be monocular or binocular, is attached to the body above the stage, and it supports the eyepiece and objective lenses at a known distance and angle. The eyepieces, at the upper end of the tube, can easily be pulled out and replaced with another set of eyepieces having the required magnification. At the lower end, the tube is fitted with a nosepiece into which the objectives are screwed. In some microscopes the tube carrying the two sets of lenses can be lowered and raised by the coarse and fine adjustments, in order to bring the specimen on the slide into focus. The focusing knobs may be located near the base or the upper arm depending on the type of microscope. In some modern microscopes, the body and the tube with its objectives and eyepieces are fixed and the coarse and fine adjustments are incorporated in the stage which is therefore moveable vertically. In some microscopes, focusing knobs are built together as coarse and fine adjustments. Inside the tube there is sometimes an inner tube called the draw tube which can be extended; when an inner tube is present, the eyepiece is fitted into the upper end of this. On the inner tube a scale is engraved for the purpose of adjusting the tube-length; the length required for the lenses varies in different microscopes.

The objective

It is the most important lens in a microscope. Usually three objectives (and one optional) with different magnifying powers are screwed to the revolving nosepiece. They are:

(a) *Low power objective (X 10):*

It produces ten times magnification of the object.



(b) **High dry objective (X 40):**

It gives a magnification of forty times.

(c) **Oil-immersion objective (X100):**

It gives a magnification of hundred times, when immersion oil fills the space between the object and the objective.

(d) **The scanning objective (X4) (optional).**

The primary magnification (X4, X10, X40 or X100) provided by each objective is engraved on its barrel. The higher power objectives (40X and 100X) of modern microscopes are spring-loaded, that is, the front mount of the objective will be pushed-in rather than pushed-through a specimen if such an objective is accidentally pressed against a specimen when focusing. Spring-loaded objectives help to protect a specimen and the front lens of an objective from being damaged.

The eyepiece

The eyepiece is a drum, which fits loosely into the draw tube. It magnifies the magnified real image formed by the objective to a still greatly magnified virtual image to be seen by the eye.

Usually, each microscope is provided with two types of eyepieces with different magnifying powers (X10 and X15 or X25). Depending upon the required magnification, one of the two eyepieces is inserted into the draw tube before viewing.

Binocular microscopes enable the user to see the image with both eyes at once by dividing the single image between two eyepieces. This reduces the strain on any one single eye particularly if being used for prolonged periods. However, a monocular microscope is suitable when being used for short periods or when daylight is the only available source of light. This is because the prism system in the binocular microscope halves the light, making daylight insufficient when using the 100X oil immersion objective.

Theoretical Principles of Microscopy:

To use the microscope efficiently and with minimal frustration, one should understand the basic principles of microscopy: **magnification and resolution.**

1. Magnification (Magnifying Power):

The total magnification obtained in a compound microscope is the product of objective magnification and ocular magnification.

$$M_t = M_{ob} \times M_{oc}$$

Where,

M_t = Total magnification,

M_{ob} = Objective magnification and

M_{oc} = Ocular magnification

If the magnification obtained by the objective (M_{ob}) is 100 and that by the ocular (M_{oc}) is 10, then total magnification (M_t) = $M_{ob} \times M_{oc} = 100 \times 10 = 1000$. Thus, an object of 1μ will appear as $1,000\mu$. More examples are given below:

Objective magnification	Eyepiece magnification	Total magnification
10X	10X	100X
40X	10X	400X
100X	10X	1000X

2. Resolution (Resolving Power of Objective):

Resolution is the degree to which the detail in the specimen is retained in the magnified image.

The Resolving Power, R of a microscope (also known as the limit of resolution, d of objective) is the closest distance between two structural entities of a specimen at which the entities can still be seen as individual structures in the magnified image.

It may appear that very high magnification can be obtained by using more number of high power lenses. Though possible, the highly magnified image obtained in this way is a blurred one. That means, each point in the object cannot be found as widely spaced distinct and separate point on the image.

Mere increase in size (greater magnification) without the ability to distinguish structural details (greater resolution) is of little value. Therefore, the basic limitation in light microscopes is one not of magnification, but of resolving power (the ability to distinguish two adjacent points as distinct and separate, i.e. to resolve small components in the object into finer details on the image). The smaller the value for R (ie, higher Resolving Power), the smaller the object that can be seen distinctly. Points with their in-between distance less than ' d ' or objects smaller than ' d ' cannot be resolved into separate points on the image.

The best theoretical Resolving Power of a light microscope is about 200nm (0.2µm) or just below the size of many bacterial cells. That is to say that bacterial cell can be observed by light microscope but, for the most part their internal structures cannot be seen.

Resolving power is a function of two factors as given below;

- (a) Numerical aperture (N.A)
- (b) Wavelength of the light (λ)

Therefore, Resolving Power/Limit of resolution of objective (d) = $\lambda/2 \text{ N.A} = 0.5\lambda/\text{N.A}$

Where,

λ = Wave length of light and

N.A = Numerical aperture of the objective

(a) Numerical aperture:

The numerical aperture of an objective is its light gathering capacity, which depends on the size of the angle, θ (half angular aperture) and the refractive index, n (light bending capacity) of the medium existing between the object and the objective.

Numerical aperture (N.A) = $n \sin \theta$

Where,

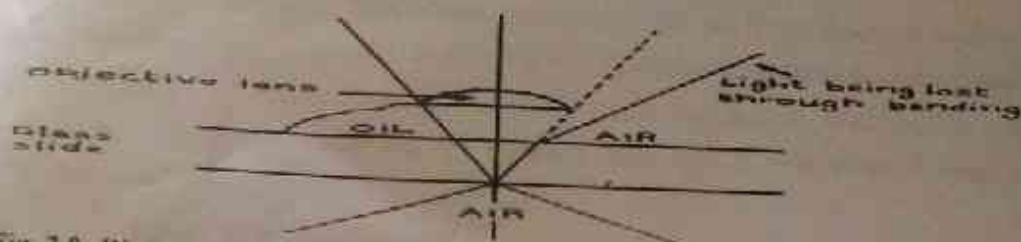
n = Refractive index of the medium between the object and the objective and

θ = Half aperture angle

For air, the value of ' n ' is 1. When the space between the lower tip of the objective and the slide carrying the object is air, the rays emerging through the glass slide into this air are bent or refracted, so that some portion of it do not pass into the objective. Thus, loss of some light rays reduces numerical aperture and decreases the resolving power.

However, when this space is filled with immersion oil, which has greater refractive index ($n=1.56$) than that of air ($n=1.00$), light rays are refracted or bent more towards the objective (fig. 3.0). Thus, more light rays enter into the objective and greater resolution is achieved.

In oil immersion objective, which provides the highest magnification, the size of the aperture is very small. Therefore, it needs bending of more rays into the aperture, so that the object can be distinctly resolved. That is why, immersion oils, such as cedar wood oil and liquid paraffin are used to fill the gap between the object and the objective, while using oil-immersion objective. Oil immersion lens also has a short focal length, a very shallow depth of field (i.e., only thin section of the specimen can be in focus at a time) and a short working distance (i.e., the distance between the specimen and the objective lens). Hence care should be taken when focusing to avoid breaking the slide and scratching the oil immersion lens.



[Fig. 3.0: Diagram showing how immersion oil prevents refraction of a beam of light when passing through glass and air]

(b) Wavelength of light (λ):

The smaller the wavelength of light (λ), the greater is its ability to resolve the points on the object into distinctly visible finer details in the image. Thus, the smaller is the wavelength of light, the greater is its resolving power, n .

Example: For a typical light microscope with $\lambda_{\text{green}} = 0.55 \mu\text{m}$ and $\text{N.A} = 1.30$, calculate the limit of resolution (Resolving Power) of the objective lens.

Solution: $d = \lambda / 2 \text{ N.A} = 0.55 / 2 \times 1.30 = 0.21 \mu\text{m}$.

Therefore, the smallest details that can be seen by a typical light microscope have the dimension of approximately $0.2 \mu\text{m}$. Smaller objects or finer details than this cannot be resolved in a compound microscope.

Useful magnification:

It is the magnification that makes visible the smallest resolvable particle. The useful magnification in a light microscope is 1,000 X the Numerical Aperture (N.A). Any magnification beyond this value makes the image blurred.

ESSENTIAL FEATURES OF VARIOUS MICROSCOPES

1) Brightfield Microscope

This instrument contains two lens systems for magnifying specimens: the ocular lens in the eyepiece and the objective lens located in the nose-piece. The specimen is illuminated by a beam of tungsten light focused on it by a sub-stage lens called a condenser, and the result is that the specimen appears dark against a bright background. A major limitation of this system is the absence of contrast between the specimen and the surrounding medium, which makes it difficult to observe living cells. Therefore, most brightfield observations are performed on nonviable, stained preparations.

2) Darkfield Microscope



This is similar to the ordinary light microscope; however, the condenser system is modified so that the specimen is not illuminated directly. The condenser directs the light obliquely so that the light is deflected or scattered from the specimen, which then appears bright against a dark background. Living specimens may be observed more readily with darkfield than with brightfield microscopy.

The Dark-field Microscopy has been successfully used in the following highly specific investigative studies, such as:

- (a) Examination of unstained bacteria suspended in an appropriate liquid.
- (b) Studies related to the internal structure as observed in eukaryotic microbes, and
- (c) Examination of highly specific and very thin spirochaetes e.g. *Treponema pallidum* - the causative agent of syphilis.

3) Phase-Contrast Microscope

Observation of microorganisms in an unstained state is possible with this microscope. Its optics includes special objectives and a condenser that make visible cellular components that differ only slightly in their refractive indexes. As light is transmitted through a specimen with a refractive index different from that of the surrounding medium, a portion of the light is refracted (bent) due to slight variations in density and thickness of the cellular components. The special optics convert the difference between transmitted light and refracted rays, resulting in a significant variation in the intensity of light and thereby producing a discernible image of the structure under study. The image appears dark against a light background.

4) Normarsk Differential Interference Contrast Microscope (NDIC) or (DIC)

This bears a close resemblance to the Phase contrast microscope wherein it produces an image based upon the ensuing differences in the following two fundamental physical parameters: refractive indices and thickness. In practice, two distinct and prominent beams of plane-polarized light strategically held at right angles to each other are produced by means of prisms. The Object beam passes through the specimen, while the reference beam is made to pass through a clear zone in the slide after which the two emerging beam combine meticulously to form an image due to their interference with each other. It produces 3D highly coloured images of live-unstained specimen. It also produces clear and distinct visibility of cellular structures (cell walls, granules, vacuoles, eukaryotic nuclei, and endospores). Note that the resolution of a DIC-Microscope is sufficiently higher than that of phase-contrast microscope due to its production of coloured image.

5) Fluorescent Microscope

This microscope is used most frequently to visualize specimens that are chemically tagged with a fluorescent dye. The source of illumination is an ultraviolet (UV) light obtained from a high-pressure mercury lamp or hydrogen quartz lamp. The ocular lens is fitted with a filter that permits the longer ultraviolet wavelengths to pass, while the shorter wavelengths are blocked or eliminated. Ultraviolet radiations are absorbed by the fluorescent label and the energy is re-emitted in the form of a different wavelength in the visible light range. The fluorescent dyes absorb at wavelengths between 230 and 350 nanometers (nm) and emit orange, yellow, or greenish light. This microscope is used primarily for the detection of antigen-antibody reactions. Antibodies are conjugated with a fluorescent dye that becomes excited in the presence of ultraviolet light, and the fluorescent portion of the dye becomes visible against a black background.

Examples : A few typical examples are as follows :

(a) *Mycobacterium tuberculosis* : **Auramine O** (i.e., a fluorochrome) that usually glows yellow on being exposed to UV-light, gets strongly absorbed by *M. tuberculosis* (a pathogenic 'tuberculosis' causing organism). Therefore, the dye when applied to a specific sample being investigated for this bacterium, its presence may be detected by the distinct visualization of bright yellow microbes against a dark background.

(b) *Bacillus anthracis* : **Fluorescein isothiocyanate (FITC)** (i.e., a fluorochrome) stains *B. anthracis* particularly and appears as 'apple green' distinctly. This organism is a causative agent of anthrax.

6) Electron Microscope

This instrument provides a revolutionary method of microscopy, with magnifications up to one million times. This permits visualization of submicroscopic cellular particles as well as viral agents. In the electron microscope, the specimen is illuminated by a beam of electrons rather than light, and the focusing is carried out by electromagnets instead of a set of optics. These components are sealed in a tube in which a complete vacuum is established. Put simply, an electron microscope scans with electrons rather than visible light, resulting in a very detailed (and awesome looking) image. This works because the wavelength of the electrons is much smaller than the wavelength of light from a bulb or laser, allowing for greater detail when scanning. There are two main types of electron microscope:

- a) **Transmission Electron Microscopes (TEM)**-A TEM works by sending the beam of electrons through a very thin specimen (specimens that are thinly prepared, fixed, and dehydrated for the electron beam to pass freely through them). So rather than scanning over and bouncing off, the electrons pass *through* the sample to create a highly detailed two-dimensional image of the internal cellular structures. Since the TEM allows for such incredible interior detail, they're often used in medical research and nanotechnology.





6) **Scanning Electron Microscopes (SEM)**– They are used for visualizing surface characteristics rather than intracellular structures. A SEM sends a beam of focused electrons to the sample, which bounce off to create a three-dimensional surface image. With this method, you can create a picture with high magnification and high resolution, but it will always be an exterior view. When using a SEM, the sample must be electrically conductive enough so the electrons actually bounce off it to create the image. Thus specimens are often coated in a thin layer of gold or other metal.

7) **Confocal Microscopes (Confocal Scanning Laser Microscope, CSLM)**

Unlike other light microscopes that use regular light for image formation, the visible light source in confocal microscope comes from a laser. Because the laser can penetrate a sample deeper than light from a bulb, it creates a three-dimensional (3D) image from a selected depth of the specimen. The laser scans the sample with the help of a series of scanning dichromatic mirrors, assembles the image in a computer, and displays the magnified image on the computer screen. No eyepiece is needed. With this, you can examine the interior structures of a non-opaque specimen, or look at the surface of an opaque specimen as deep as the laser light can penetrate. This results in highly selective, detailed images. Like the compound microscope, these microscopes offer a high degree of magnification, but their resolution is much better. They are commonly used in cell biology and medical applications.

8) **X-ray Microscope**

This type uses electromagnetic radiation in the form of x-rays to produce images of tiny objects. Unlike an electron microscope, it can be used to generate an image of living cells. They're useful in biological research and metallurgy, as the images are highly detailed.

9) **Scanning Probe Microscope**

Scanning Probe Microscopes measure surface features of a sample by moving a sharp physical probe over the object's surfaces. The tip of the probe scans (goes line by line) the specimen many times to generate data, and a computer combines the data to create an image. Unlike an electron microscope, these scan in normal air rather than a vacuum (or partial vacuum). But, the scanning can be slow. It can produce a highly magnified image of about 100,000,000X with a very high resolution of less than 1nm. The major difference between SPMs and other optical microscopes is that, here the user doesn't see the surface directly; rather, the probe feels the surface and creates an image to represent it. There are several types of Scanning Probe Microscopes which include: Scanning Tunneling Microscopes (STMs), Atomic Force Microscopes (AFMs), Magnetic Force Microscopes (MFMs), etc.

SUMMARY OF THE DIFFERENT TYPES OF MICROSCOPES AND THEIR APPLICATIONS

Type of microscope	Maximum useful magnification	Resolution	Description
Bright field	1,500X	100 - 200nm	Extensively used for the virtualization of microorganisms. Usually necessary to stain specimen for viewing.
Dark field	1,500X	100 - 200nm	Used for viewing live microorganisms, especially those with characteristic morphology; staining not required; specimen appears bright on a dark background.
Phase contrast	1,500X	100 - 200nm	Used to examine structures of living microorganisms; does not require staining.
Fluorescence	1,500X	100 - 200nm	Uses fluorescent staining; useful in many diagnostic procedures for identifying microorganisms.
Normal disk differential interference	1,500X	100 - 200nm	Used to examine structures of microorganisms; produces sharp, multicoloured image with 3-dimensional appearance.
Confocal scanning	1,500X	100 - 200nm	Used to examine structures of microorganisms within mixtures of various types of microorganisms; uses fluorescence staining; produces blur-free image; used to produce 3-dimensional images.
Transmission Electron (TEM)	500,000 - 1,000,000X	1-2nm	Used to view ultra-structures of microorganisms including Viruses; much greater resolving power and useful magnification than can be achieved with light microscopy.
Scanning Electron (SEM)	10,000 - 1,000,000X	1-10nm	Used for showing detailed surface structures of microorganism; produces a 3-dimensional image.
Scanning Probe Microscope (SPM)	100,000,000X	<1nm	

These are the main types of microscopes, but there are others. Some have fallen out of fashion, and some (such as the neutron microscope) are still in their experimental stages. As technology and scientific understanding moves forward, the types of microscopes available will change as well.

While scientists have a variety of optical instruments with which to perform routine laboratory procedures and sophisticated research, the compound brightfield microscope is still the "workhorse" and they are commonly found in all biological laboratories. Although you should be familiar with the basic principles of microscopy, you may probably not be exposed to all the diverse, more sophisticated and expensive types in the course of this study. Therefore only the compound brightfield microscope will be discussed in depth and used to examine specimens in your practical sessions.



HOW TO USE A MICROSCOPE

The following routine procedures must be followed to ensure correct and efficient use of the microscope while focusing:

1. Place the microscope slide with the specimen within the stage clips on the mechanical stage. Move the slide to center the specimen over the opening in the stage directly over the light source.
2. Rotate the scanning lens or the low power lens into position. While watching from the side to ensure that the lens doesn't touch the specimen, turn the coarse focus knob to move the stage as close as it can get to the lens without touching the lens. (Always watch from the side whenever you move a specimen towards any objective lens to make sure the lens doesn't crash through the specimen and get damaged!)
3. Now, while looking through the ocular lens, turn the coarse focus knob carefully, and slowly move the stage away from the lens until the specimen comes into vague focus. Then, use the fine focus knob to bring the specimen into sharp focus.
4. Routinely adjust the light source by means of the light source transformer setting, and/or the iris diaphragm, for optimum illumination for each new slide and for each change in magnification.
5. Most microscopes are **parfocal**, which means that when one lens is in focus, other lenses will also have the same focal length and can be rotated into position without further major adjustment. In practice, however, usually a half-turn of the fine-adjustment knob in either direction is necessary for sharp focus.
6. Once you have brought the specimen into sharp focus with a low-powered lens, preparation may be made for visualizing the specimen under oil immersion. Place a drop of oil on the slide directly over the area to be viewed. Rotate the nosepiece until the oil-immersion objective locks into position. The slide is observed from the side as the objective is rotated slowly into position. This will ensure that the objective will be properly immersed in the oil. The fine-adjustment knob is readjusted to bring the image into sharp focus.
7. During microscopic examination of microbial organisms, it is always necessary to observe several areas of the preparation. This is accomplished by scanning the slide without application of additional immersion oil. This will require continuous, very fine adjustments the slow, back-and-forth rotation of the fine adjustment knob only.

CARE AND MAINTENANCE OF A MICROSCOPE

Care before using the microscope

- using a clean cloth, wipe any dust and dirt from the stage and other surfaces of the microscope. Use a small soft brush to clean less accessible places such as the revolving nosepiece.
- Using lens tissue or a piece of clean cotton cloth or cotton bud, clean the surface lenses of the non-oil-immersion objectives, the upper lenses of the eyepieces, condenser lens and illuminator lens.

Care when using the microscope

- Mechanisms in a microscope, for example, focusing controls, mechanical stage controls, etc., are delicate and should never be forced.
- Make sure the surface of the stage and the underside of the specimen is dry and clean before inserting the specimen in the slide holder. Whenever possible, cover wet preparations with cover glasses.
- Always move the 100x objective to one side when inserting and removing specimens to prevent scratching of the front lens of the objective.
- Use non-drying immersion oil whenever possible. When changing to new oil, wipe off all the previous oil from the objective lens to avoid mixing the immersion oils.

Care after using the microscope

- Turn the lamp brightness to its lowest setting, switch off the microscope, and remove the plug from the mains socket.
- Using a piece of soft tissue or a soft piece of clean cotton cloth, wipe the immersion oil from the 100X objective. Do not use organic solvents, these dissolve the glue holding the lens.
- Do not leave an objective or eyepiece opening (socket) empty. If the plugs are not available, cover the openings with adhesive tape to prevent dirt from entering into the microscope.
- Clean the microscope stage using a piece of cloth or cotton swab dampened in 70% ethanol, then dry the surface.

- Cover the microscope with its dust cover.
- The correct way of carrying the microscope is by using both hands. Hold the limb using one hand while the other supports the base.
- If the microscope is to be stored for a number of days in hot humid conditions, it should be sealed in an airtight plastic bag containing dry silica gel which absorbs moisture from the bag.

Summary

1. The basic function of the microscope is to magnify the images of small objects so that they can be clearly seen.
2. The main parts of a microscope are the illuminating apparatus, the stage, the tube, the objectives and the eyepieces.
3. The total magnification of a microscope is the product of the magnification of the objective and that of the eyepiece.
4. Steps involved in the correct use of a microscope are:
 - i. Position microscope correctly
 - ii. Align the 10X objective
 - iii. Insert specimen (on a slide) onto the stage
 - iv. Focus image of object
 - v. Focus the condenser
 - vi. Adjust illumination
 - vii. Examine under the 40X and then 100X

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