

Physics of Medical Devices

Practical Lecture

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Microscope

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Introduction

The microscope is a valuable instrument. There are many small objects or details of objects which cannot be seen by the unaided human eye. The microscope magnifies the image of such objects thus making them visible to the human eye. Microscopes are used to observe the shape of bacteria, fungi, parasites and host cells in various stained and unstained preparations.

History of Microscope

- In the 1st Century AD, the Romans invented the glass and used them to magnify objects.
- In the early 14th Century AD, eyeglasses were made by Italian spectacle makers.
- In 1590, two Dutch spectacle makers, Hans, and Zacharias Jansen created the first microscope. It was a simple tube with 2 lenses system and had 9X magnification.
- In 1670, Robert Hooke, an English Chemist, Mathematician, Physicist, and Inventor, improvise the microscope of that time and developed the compound microscope. He first developed the 3 lenses microscope.
- In 1675, Anton Van Leeuwenhoek ground a glass ball into a convex lens and used it to make a single-lens microscope with 270X magnification. Using this microscope, he first observed the bacterial cells.

- In 1729, Chester Moore Hall first presented and used achromatic lenses in the microscope.
- In 1830, Joseph Jackson Lister suggested the use of multiple low-power lenses to achieve clear magnification.
- In 1878, Ernst Abbe, a German Physicist and Optical Scientist, developed a mathematical theory relating wavelength with image resolution. He was the first to develop and use water and oil immersion lenses.
- In 1903, Richard Zsigmondy invented the ultramicroscope. This could view objects smaller than the wavelength of light.
- In 1932, Frits Zernike invented the phase-contrast microscope.
- In 1938, Max Knoll and Ernst Ruska invented the first electron microscope. It was a transmission electron microscope (TEM). It used a beam of an electron instead of light to make an enlarged image.
- In early 1940, Russian physicist Sergey Y. Sokolov developed concept of ultrasound microscope. But, only in 1970, its working model was developed in America.
- In 1942, Ernst Ruska improved TEM into a scanning electron microscope (SEM). In this type, electron beams are passed across the specimen instead of passing through it as in TEM.
- In 1951, British physicists William Nixon and Ellis Coslett invented X-ray microscope.

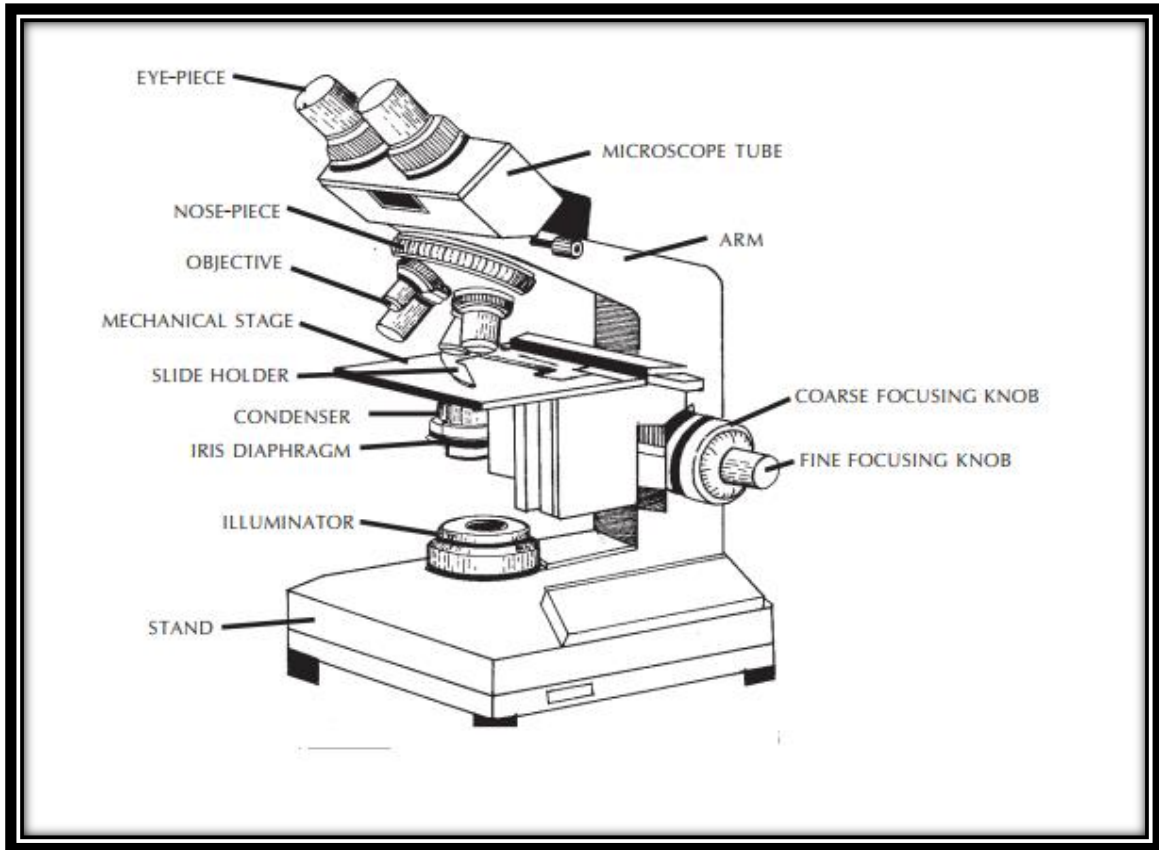
- In 1981, Gerd Binnig and Heinrich Rohrer invented the scanning tunneling microscope. This allowed us to get the 3-D image of an object.

Types of Microscopy

Microscopes used in clinical practice are light microscopes. They are called light microscopes because they use a beam of light to view specimens. A compound light microscope is the most common microscope used in microbiology. It consists of two lens systems (combination of lenses) to magnify the image. Each lens has a different magnifying power. A compound light microscope with a single eye-piece is called monocular; one with two eye-pieces is said to be binocular. Microscopes that use a beam of electrons (instead of a beam of light) and electromagnets (instead of glass lenses) for focusing are called electron microscopes. These microscopes provide a higher magnification and are used for observing extremely small microorganisms such as viruses.

Parts of the Microscope

The main parts of the microscope are the eye-pieces, microscope tube, nosepiece, objective, mechanical stage, condenser, coarse and fine focusing knobs, and light source



What is Refractive Index?

Refractive Index can be defined as velocity of light in a vacuum to velocity of light in a medium (substance). Simply it is the measure of bending of a light ray when passing from one medium to another.

Mathematically it can be defined as;

$$n = c/v$$

where,

n = refractive index

c = speed of light in vacuum

v = velocity of light in a medium

Resolution

in microscopy, this refers to the ability of the lens system to separate the image into small parts OR the ability to separate two elements in the viewing field from each other. As the light increases, resolution should increase or as the light decreases, resolution in general decreases. Since resolution is a function of diffraction the very best optical microscopes are limited to a resolution of 0.2 micrometers. NOTE – in modern usage, the resolution of a projector or a video monitor or any other display unit, has a different meaning, as noted below.

What is Resolution?

Resolution can be defined as the shortest distance between two points on a specimen that can be distinguished by a microscope in its image. It is the ability of a microscope to distinguish details on a specimen.

Mathematically it is given as;

$$r = \lambda / 2NA$$

where,

r = resolution

λ = imaging wavelength

NA = numerical aperture

Monocular

A single eyepiece, is set at 45° to the head for viewing by one observer. Inexperienced microscopists will tend to close the eye not looking through the eyepiece, which may result in headaches and eye strain to the intense light in one eye and darkness in the other.

Binocular

Two eyepieces, side by side, one for each of the Viewer's eyes. The two eyepieces are generally set at a comfortable 45° angle. The inter-ocular or inter-pupillary distance may be adjusted to match the distance between an individual's eyes.

Polarizing Microscope

Polarizing Microscope is a special type of light microscope that uses polarized light to illuminate a specimen and develop its magnified image. It is similar to a regular optical microscope but uses polarized light instead of normal natural light. It enhances image quality and image contrast. They are also called petrographic microscopes.

Polarizing Microscope Principle

Normal light produced from illuminator is passed through polarizer which converts the normal light into plane-polarized light. The polarizing microscope focuses the plane-polarized light on anisotropic (substance having multiple refractive indexes) specimen. When the polarized light waves strike such anisotropic specimen, birefringence (double reflection) occurs generating two waves, ordinary and extraordinary waves, which are perpendicular to each other. These two waves get transmitted in different phases. An analyzer combines these two waves and passes through the ocular lens to develop an enlarged image.

Uses of Polarizing Microscope

- Used in geological studies to study rocks, minerals and soil components.
- Used in studying internal structures of transparent planktons, diatoms, protozoans, etc.