



S.G.V.C Vidya Prasarak Trust's
MGVC ARTS, COMMERCE AND SCIENCE COLLEGE
MUDDEBIHAL -586212



DEPARTMENT OF ZOOLOGY

A Project Work

CERTIFICATE

Register No: **S1827651**

Class: **BSc 6th Sem**

This is to certify that Mr/Miss **DEVARAJ H HANUMASAGR** of BSc
VIth Semester, MGVC College Muddebihal has satisfactorily completed
the Project work on **Microscopy** under our supervision during the year
2020-2021

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A
REPORT
ON
MICROSCOPY



Introduction :

Microscope is an optical instrument designed for the study of objects. Which can not be seen through naked eye. It makes us see things which are very small and are otherwise invisible. It is an inevitable instrument for clinical and laboratory studies. The very first microscope was designed by JENSSSEN and HANS.

The optical system of the microscope magnifies the objects. The ratio of this magnified image to that formed image to that formed on retina of an unaided normal eye is termed the magnification.

Resolving power :

The resolving power is the capacity of an instrument to show distinct images of points lying very close together. The resolving power of unaided human eye is 0.1mm. thus the human eye can distinguish objects up to a size of 0.1 mm.

Types of microscopes :

The microscopes are classified in to three types based on the source of light. The types are,

1. Light microscopes
2. Electron microscopes, and
3. X-ray microscopes.



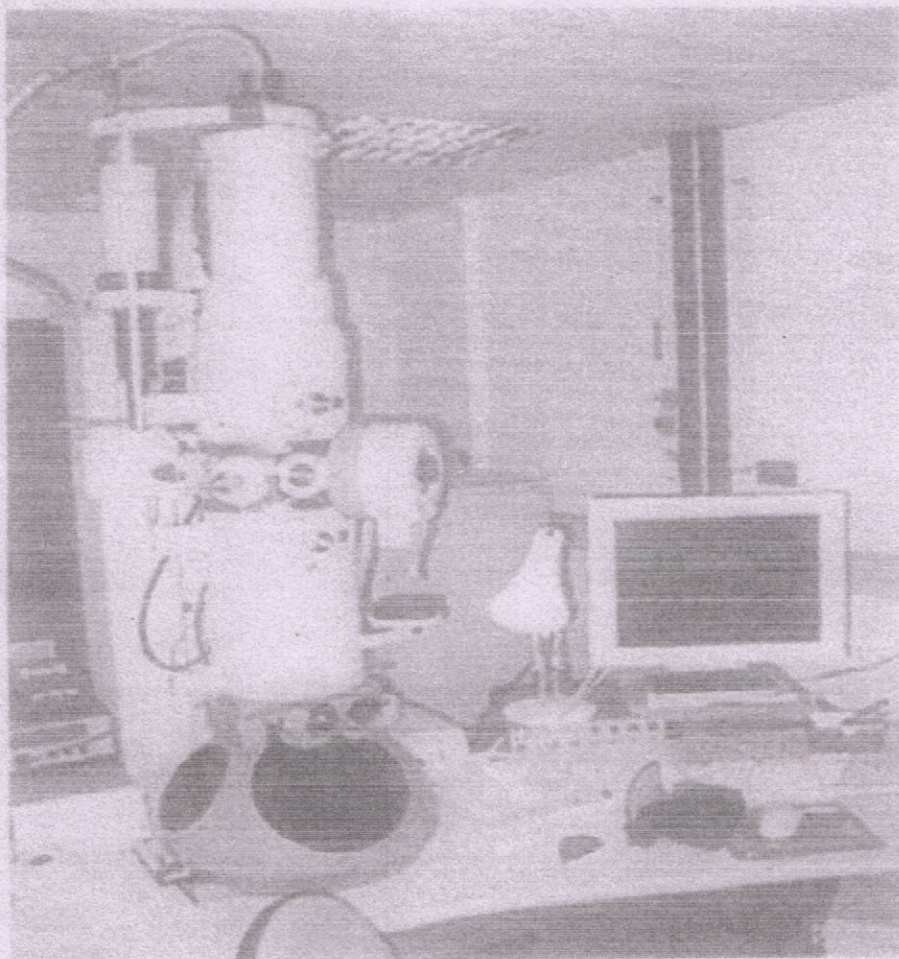
Structure :

The transmission electron microscope comprises the following parts:

01. **Electron gun and anode** . In TEM, source of illumination is electrons which are produced and concentrated into an electron beam by electron gun. Electron gun consists of an electrically heated tungsten filament or cathode that emits electrons. Outside the gun is anode which attracts electrons. Thus, between the filament and anode the electrons attain high velocity with wavelength of 0.005 to 0.003 nm.
02. **Microscope column**. Electrons can travel in straight line vacuum on travel in straight line vacuum only, because in the air, they collide with oxygen or nitrogen atoms. The electron microscope, therefore, is enclosed in a evacuated metal tube.
03. **Condenser lens**. It is the electromagnetic coil which focuses or condenses the electron beam in the plane of object.
04. **Objective lens**. It is the electromagnetic coil which produces first magnified image of the object. It focuses the electrons which are reflected by the first image.
05. **Projector lens**. It is also the electromagnetic coil which magnifies the first image formed by objective lens. It produces the final image.
06. **Fluorescent screen or photographic plate**. Fluorescent screen is used for observing the magnified image of the object. It remains coated with chemical (e.g., zinc sulphide) which on being excited forms the image as on the screen of television. The final image can also be captured on photographic film; such photographs are known as electron micrographs.

Principle. In the electron microscope, an electron beam passes through the specimen and is focused by electrostatic and/ or magnetic lenses. The object is viewed on a fluorescent screen or is photographed. Lens which works as projection lens. It further magnifies the image and projects it on a zinc sulphide or fluorescent screen at the bottom of TEM. This screen is similar to screen of a television tube. It is coated with a layer of crystals that respond to electrons by emitting visible light. The final image can also be captured on the photographic film.

Electrons that are not transmitted by the specimen leave correspondingly dark regions on the viewing plate. Thus, image from a TEM is a pattern of bright and dark areas corresponding to the area of greater or lesser electron density.



ELECTRON MICROSCOPE

Electron microscope was developed 1930s. It is a powerful tool for studying the ultrastructure of cells because it has much greater resolving power than the light microscope.

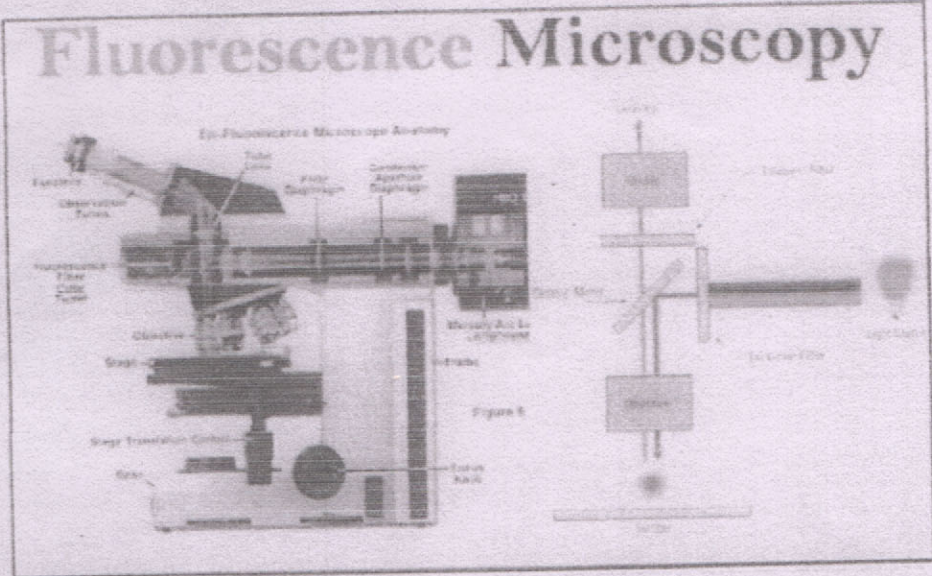
Basically there are two types of electron microscope such as Transmission electron microscope (TEMs) form images from electrons that hand bounded off the surface of the specimen. A transmission electron microscope has a very high magnification (500,000times). The resolution TEM was designed by Knoll and Ruska of Germany in 1932. It permits direct study of biological ultrastructures of cell organelles. The practical limit of resolution of electron microscope is about 3 to 5A.

they fall directly on to a photographic film , producing an micrograph.



Unlike the compound light microscope, in which image formation depends primarily upon differences in light absorption, the electron microscope forms images as a result of difference in the way electrons are scattered by various regions of the object.

In TEM, the illuminating agent is not light but the electrons of short wavelength (0.500 Å), the wavelength of electrons is determined by the voltage at which these are generated. For example, at 50,000 volts, the electrons have 0.50 Å wavelength and resolution power of electron microscope can be $0.50 \text{ Å} / 2 = 0.25 \text{ Å}$. Similarly, in an electron microscope with a voltage of 100,000 volts, the wavelength of an electron is 0.004 nm (or 0.04 Å). Theoretically the resolution of such a microscope should be 0.02 Å. But practical resolving power of most modern electron microscope is 0.5 nm or 5 Å. This is 400 times greater than that of a light microscope.



FLUORESCENT MICROSCOPE

This microscope is based on the principle of fluorescence.

Fluorescence: The substance which emit visible light when illuminated by ultraviolet rays. The fluorescence is of two types. They are autofluorescence and secondary fluorescence. The fluorescence emitted by the substances themselves is called autofluorescence, Eg., chlorophyll, porphyrin, riboflavin, Vitamin A etc. The fluorescence emitted by nonfluorescent substances is called secondary fluorescence. The non fluorescence substances emit fluorescence when they are combined with fluorescent dyes called fluorochromes. The fluorochromes are fluorescein emitting yellow-green light and rhodamine emitting orange red light. The increased resolution of the electron microscope is possible because the path of electrons can be resolved to much smaller distance than light. In practice, a resolution of 10 Å is common. And greater resolution (to 2Å) is possible with special technique.

Instead of using visible to illuminate the object (as used in light microscope), the electron microscope uses a beam of accelerated electrons

and it focusses the electron beam with electromagnets (magnetic lenses). An image is formed when electrons strike a fluorescent screen or when