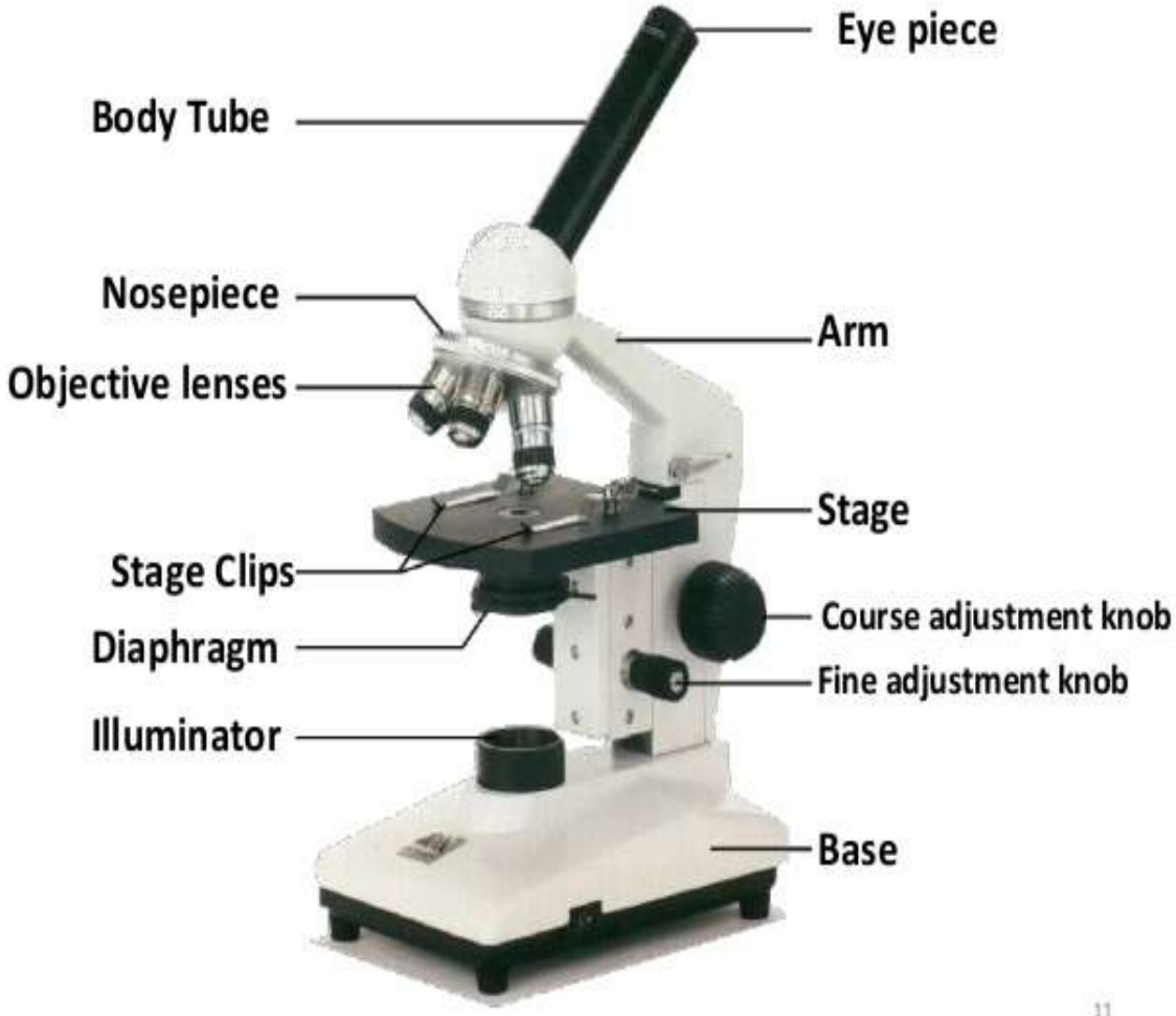
Microscope:

History

- 1590: Two Dutch spectacle-makers father-and-son team, **Hans and Z. Janssen**, create the first microscope.
- Before microscope, no one knew about microbial world
- In 1676, Antonie van Leeuwenhoek observed bacteria, yeast, blood cells and other little living entities.
- improvements in design occurred due to the work by Robert Hooke during 18th century
- named after ancient Greek words; Micros means "small" and Skopeîn means "to see

Introduction

- Microscopy is the technical field of using microscopes to view samples & objects that cannot be seen with the unaided eye (objects that are not within the resolution range of the normal eye)

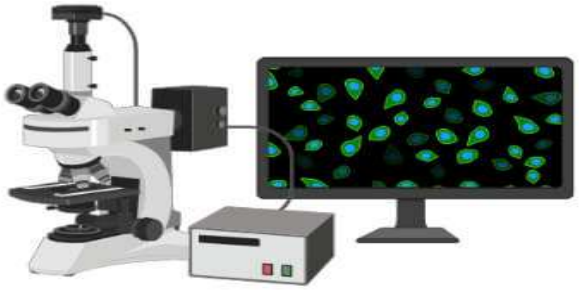


Types Of Microscope

- ✓ **Optical microscope**
- Simple microscope
- Compound microscope
- Dissection or Stereo Microscope
- Confocal Microscope

- ✓ **Electron microscope**
- Scanning Electron microscope
- Transmission Electron microscope
- ✓ **Scanning probe microscope**
- ✓ **X-ray Microscope**

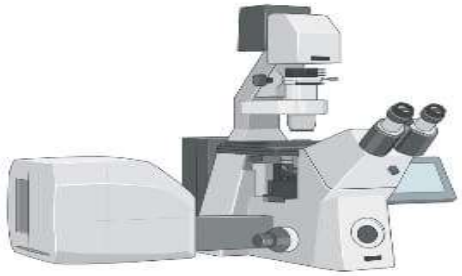
Light Microscope



Fluorescence Microscope

Electon Microscope

Stereo Microscope



Confocal Microscope

Atomic Force Microscope

Inverted Microscope

Retinal Imaging Microscope

Compound Microscope

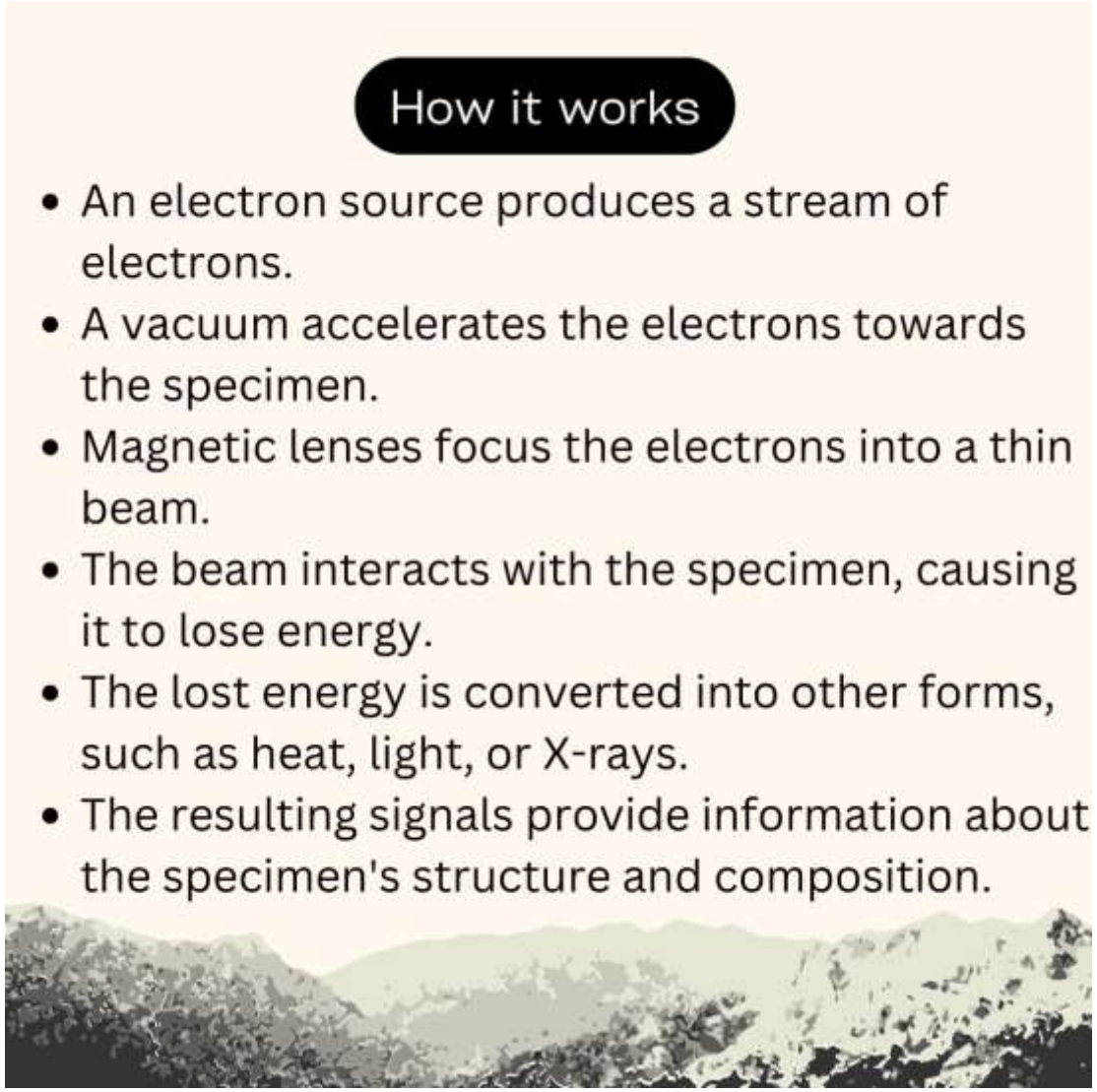
- Compound microscopy is an light microscope which is use in any biological laboratory .
- It is used for passive observation of structural details of a cell, tissue or organ in sections.
- Gives two dimensional images
- Halogen or LED light source is used. Provides high magnification (up to 1000x).

Difference between Light and Composed Microscope

Feature	Simple Microscope	Compound Microscope
Lenses	Single convex lens	Multiple lenses (eyepiece + objective)
Magnification	Low (up to 10x)	High (40x to 1000x)
Image Clarity	Less detailed	More detailed
Usage	Basic magnification	Advanced biological and medical studies

Electron Microscope

- In the early 20th century a significant alternative to the light microscope was developed.
- First electron microscope develop in 1931
- Used to see viruses or parts of cells smaller than the limits of resolution of the light microscope
- utilizes a beam of electrons instead of visible light and electromagnetic fields in place of optical lenses



Electron Microscope

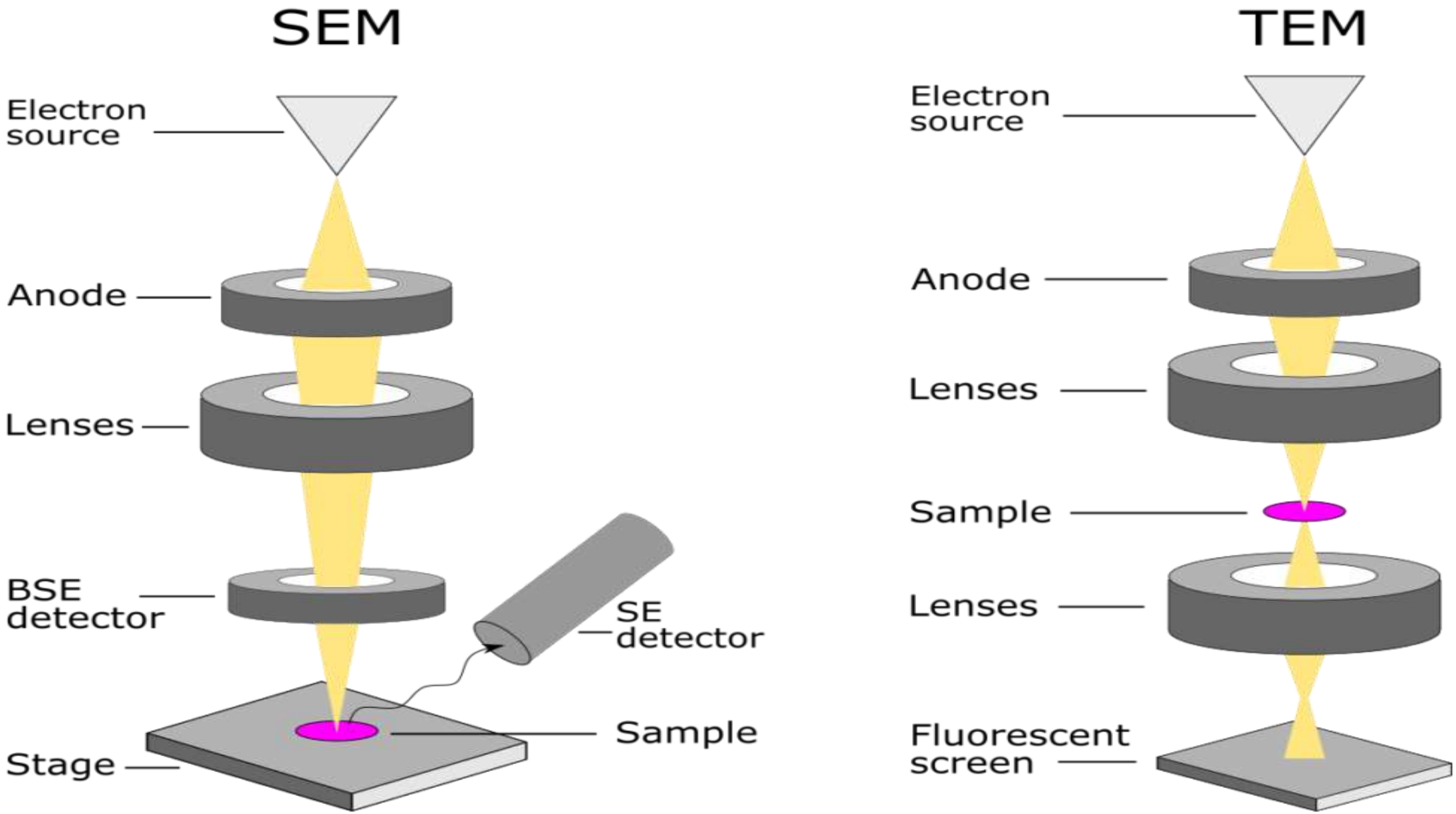
- **Types of Electron Microscopes:**

1. Transmission electron microscope (TEM): Used to view thin specimens, such as tissue sections and molecules

2. Scanning electron microscope (SEM): Used to create images of the surface of a material

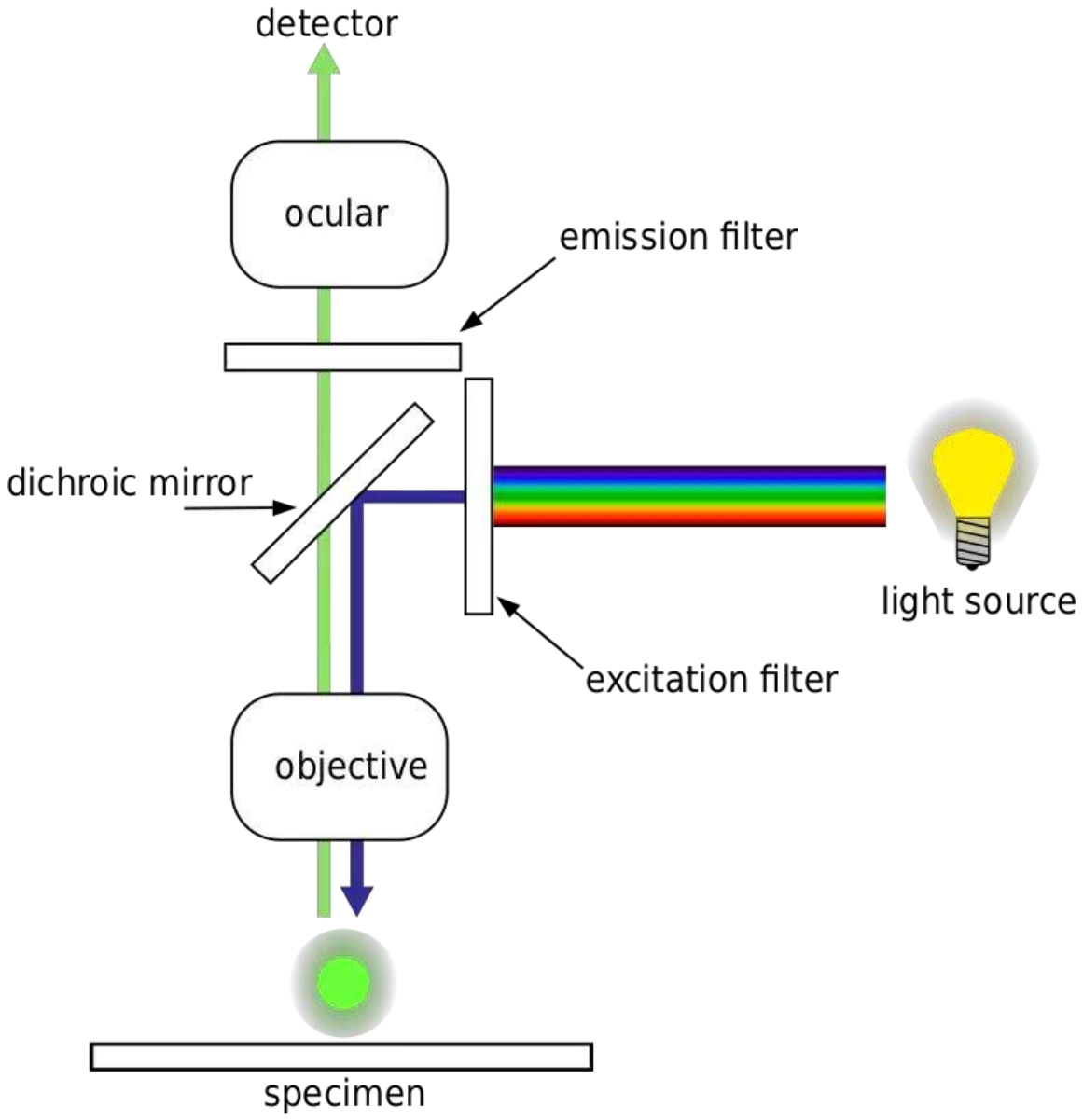
- **What it's Used for:**

- Biomedical research: EM images provide details about the structure of cells, tissues, and organelles.
- Industrial quality control: EM images help identify problems with materials.
- Neuroscience: EM images help researchers study biological structures at the atomic and cellular levels.


SEM
Electron source
Anode
Lenses
BSE detector
SE detector
Sample
Stage
TEM
Electron source
Anode
Lenses
Sample
Lenses
Fluorescent screen

Fluorescence Microscope

- An optical microscope that uses fluorescent light to create an image of a sample previously stained with fluorescing dyes called fluorochromes eg. **Rhodamine** and **Auramine**
- Used ultra - violet light of very short wavelength which is not visible to the eye.
- Dyes transform the invisible short wavelength ultra - violet light in to longer wavelength visible light.
- The fluorescent stained organisms, cells, or particles can be seen glowing (fluorescing) against a dark background



How it works

- A light source, such as a laser, mercury-vapor lamp, or xenon arc lamp, excites the sample with a specific wavelength of light.
- The sample emits light of a longer wavelength.
- An excitation filter selects the light that will excite the sample.
- The emitted light passes through the microscope objective and an emission filter.
- The emission filter blocks out light at the excitation wavelength, allowing only the light emitted by the sample to pass through.
- The image is captured by a detector.

Principle Of Compound Microscope

A microscope enlarges the view of an object by enlarging it twice using the objective and oculus lenses

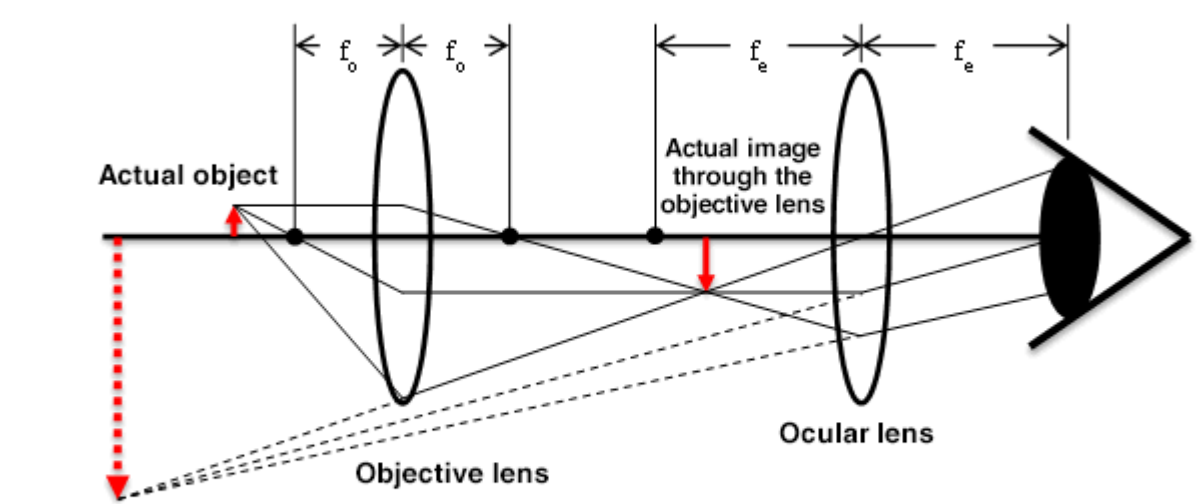


Image observed through the ocular lens
(virtual image magnified by the ocular lens)

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

NON-OPTICAL COMPONENTS

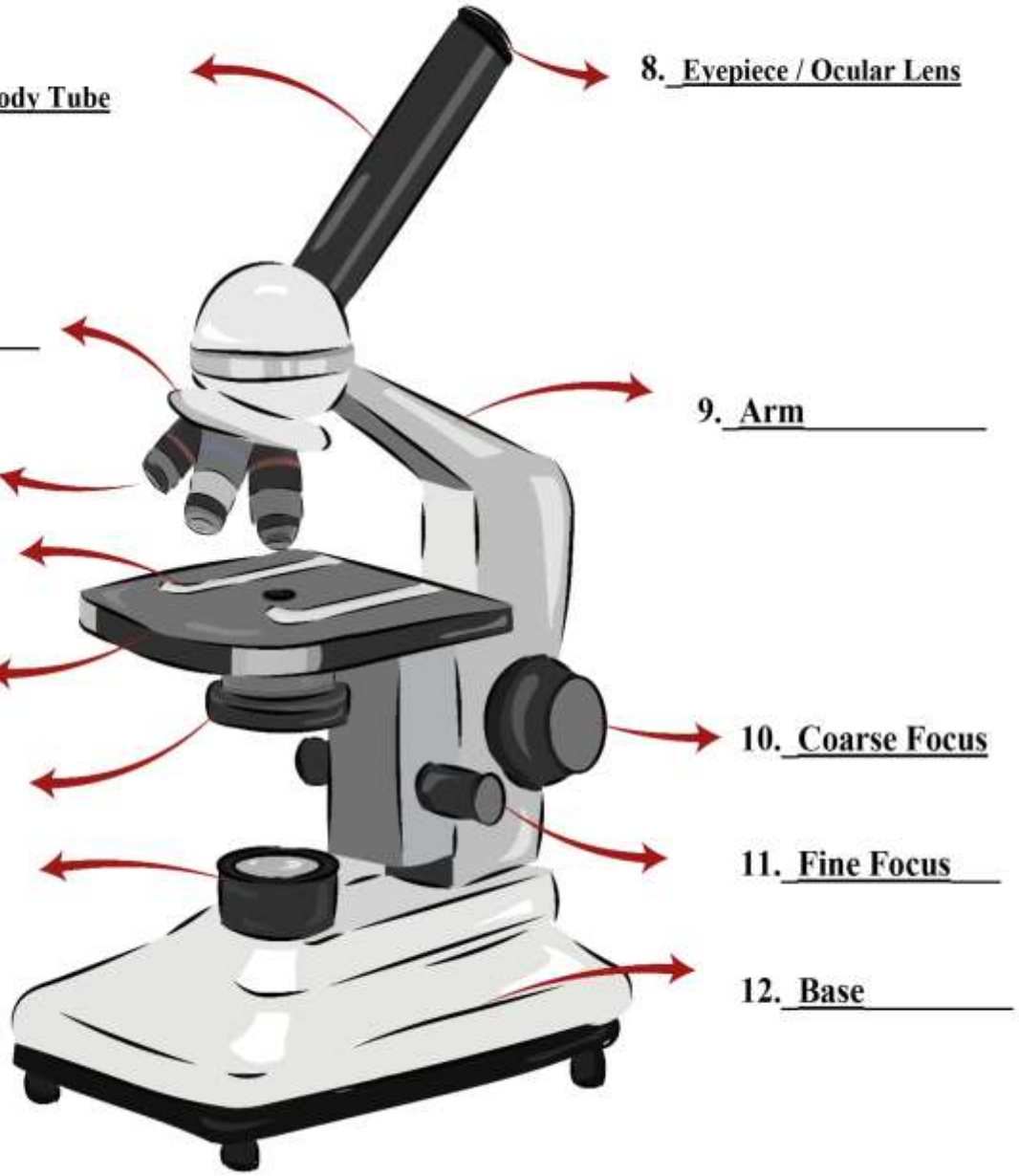
- Base
- Pillers
- Arm
- Inclination joint
- Stage
- Body tube
- Coarse Adjustment
- Automatic stop
- Adjustment screws
- Fine Adjustment

OPTICAL COMPONENTS

- Diaphragm
- Condenser
- Reflector mirror/light source
- Eye piece
- Objective piece

Parts of a Microscope Worksheet

1. Evepiece Tube or Body Tube
2. Nosepiece
3. Objective Lenses
4. Stage Clips
5. Stage
6. Diaphragm
7. Illuminator
8. Evepiece / Ocular Lens
9. Arm
10. Coarse Focus
11. Fine Focus
12. Base



Non optical components

- **Base:**

  It is U-shaped metallic structure that supports whole microscope.

- **Pillars:**

  It is short upright path that connect with arm and base.

- **Arm:**

  It is a curve metallic handle that connect with arm by inclination joint. It supports the stage and body tube.

- **Automatic Stop:**

  It is a small screw fitted at the lower of rack and pinion. It is meant stopping for downward sliding of the body tube.

- **Body Tube:**

  It is meant for holding ocular and objective lenses at its two end.

- **Inclination joint:**

  It is used for tilting the microscopy.

- **Stage:**

  It is a metallic platform with central hole fitted to lower part of arm. Microscopy sides are held on stage.

- **Fine Adjustment:**

  Stage moves extremely short distance. It is required to obtain sharp image.

- **Adjustment Screws:**

  There are two pair of screws for moving the body tube in relation to the stage

- **Draw tube :**

  It is a small tube that remain fixed at upper and of body tube. It hold eye piece.

- **Rack & Pinions:**

  It is attached with body tube or stage for begging object under focus.

- **Nose piece:**

  A revolving nose piece hold multiple lenses of different magnification level.

- **Coarse Adjustment:**

  Body tube for stage moves up and distance. It is meant for briefly objective lens at a proper distance from object.

Optical Components

- **Diaphragm:**

  It is fitted just below the stage for regulating the amount of light filling on object.

- **Condense:**

  It is attached below diaphragm. It can be move up and down to focus light on the object.

- **Reflector /mirror/light source:**

  It is attached to the base . It may be a bulb or a mirror both of its surfaces bears mirrors:

  Plane and concave mirror on either side. Plane side is used in strong light and concave side is use in weak light, it direct light on the object.

- **Eye Piece:**

  It is a lens through which image of the mirror of microscopy object is observed. usually provides a magnification power of 10x to 15x

- **Objective piece:**

  These are fitted over nose piece. It has 3 – 4 in number.

  - Lower power(10x or 4x)
  - Higher power(40x)
  - Oil immersion (100x or more)

Routine use of the microscope

1. Place on a firm bench so that it does not vibrate.
2. Not be exposed to direct sun light.
3. User must be seated at the correct height for the convenient use of the microscope.
4. Select the appropriate source of light.
5. Place the specimen on the stage, making sure that the underside of the slide is completely dry.
6. Select the objective to be used. • It is better to begin examination with 10x objective.

7. Adjust the light source until the illumination of image is at its brightest.
8. Adjust the aperture (opening) of the condenser iris according to the specimen being examined.
9. Examine the specimen by systematically moving the slide with the mechanical stage.
10. For a higher magnification, swing the 40x objective into place. Focus the 40x objective, using the fine adjustment.
11. For the highest magnification, add a drop of immersion 80 oil to the specimen and swing the 100x oil immersion objective into place.

Care of Microscope

1. Always carry a microscope using both hands.
2. When not in use, a microscope should be protected from dust, moisture, direct sunlight and put in microscope case.
3. Keep it standing in place ready for use, but protected by light cover.
4. In humid climate it is necessary to cover the microscope in a plastic bag with a drying agent (silica gel) over night to avoid molds growing on the lenses.
5. At the end of each day's work, the surface lenses of the objectives, eyepieces, and condenser should be cleaned using lens tissue.