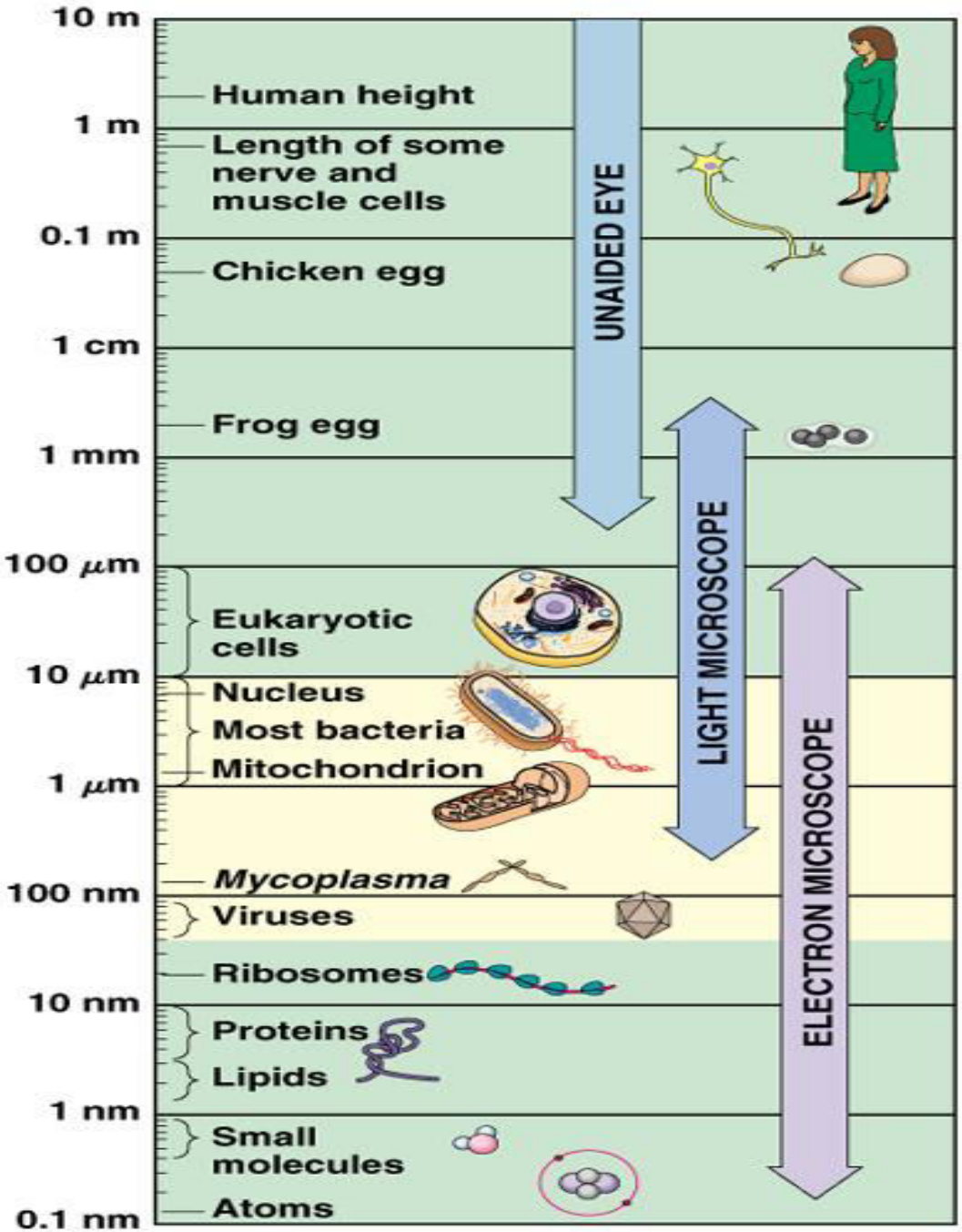
# Microscopy

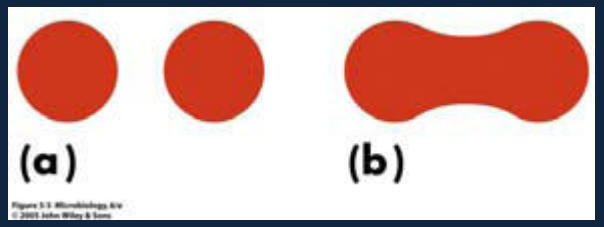
# Microscope

- is an instrument used to see objects that are too small for the naked eye.



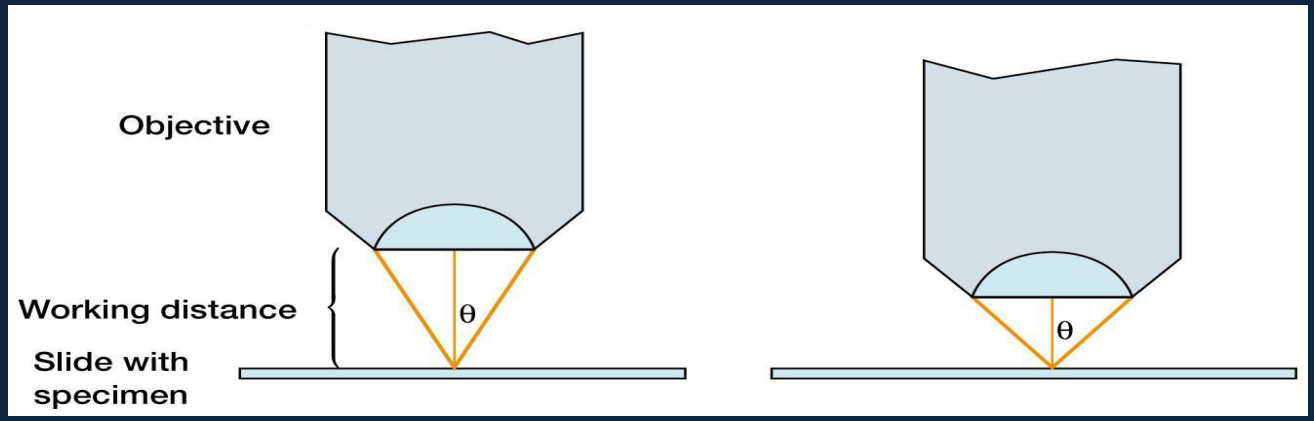
# Function of microscope:

- **Magnification:** to magnify (enlarge) the object being examined.
- **Resolution:** its ability to differentiate two objects when you view them on a specimen slide.



# Terms Related To Microscopes

- **Working distance:** The distance between the objective and the object when the object is in focus.




- **Parfocal:** When move from one objective lens to another you are still in approximate focusing.

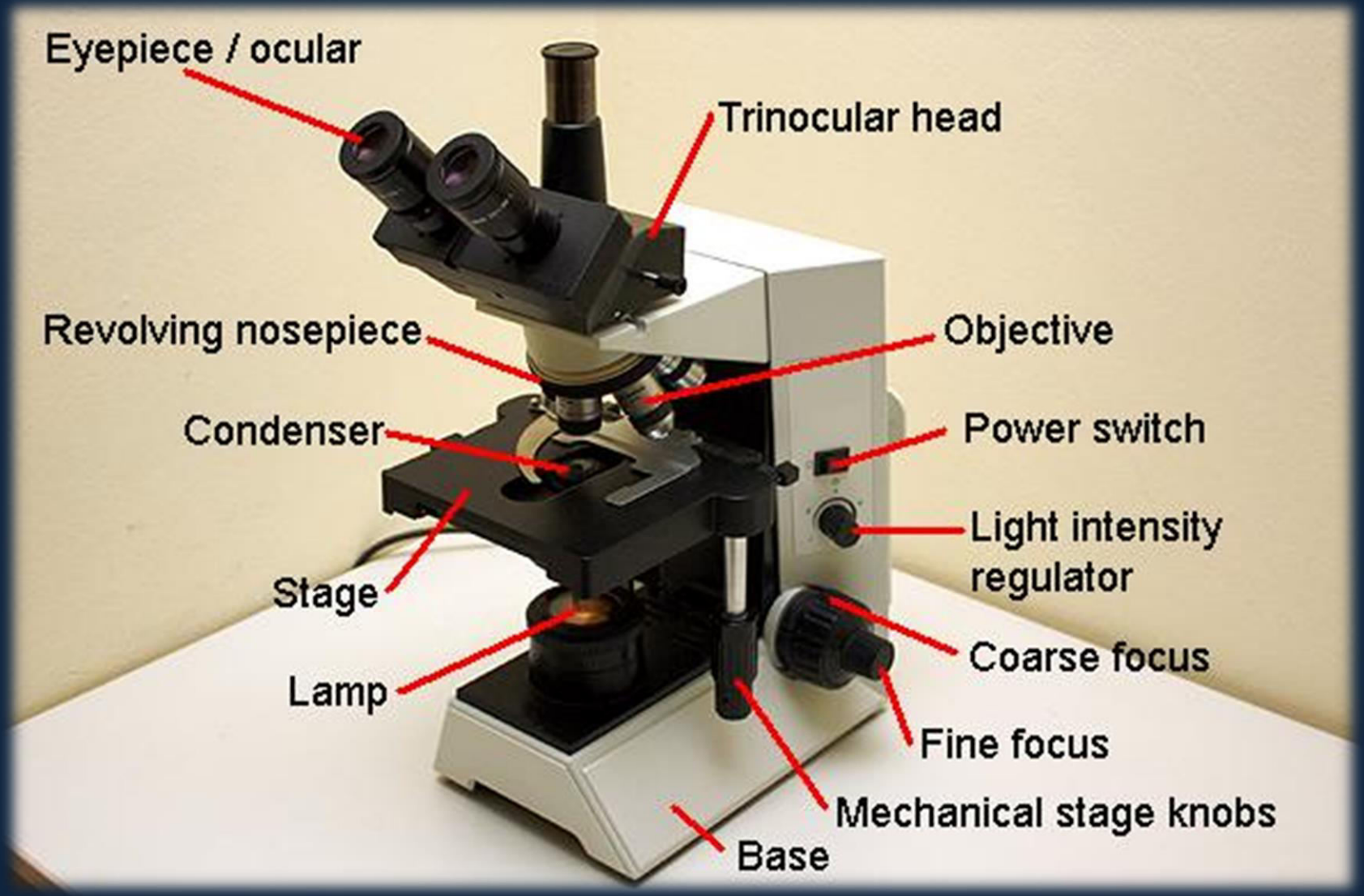
# Types of light microscope:

1. Bright field microscope
2. Dark field microscope
3. Phase contrast microscope
4. Inverted microscope
5. Dissecting microscope

# Bright field microscope

- Produces a dark image against a brighter background
- Has several objective lenses
- Uses ordinary bulb light as source of light.
- Total magnification is 1000x
- The resolution is 0.2µm
- It is mainly used to examine stained preparations.

# Bright field Microscope parts



# Microscope parts

1) Ocular Lens/Eyepiece

Magnifies the specimen image 10x

2) Nose piece

The Nose Piece holds the objective lenses and can be turned to increase the magnification

3) Arm

Used to support the microscope when carried. Holds the body tube, nose piece and objective lenses

**4) Objective lenses:** To produce a magnified image with different magnification. (x4, x10, x20, x40, and x100).

- X100 objective lens: called Oil immersion objective.

To calculate the total magnification of the microscope:

- Total mag. = objective lens mag. X ocular lens mag.
- The lowest mag. = 4 X 10 = X40
- The highest mag. = 100 X 10 = X1000

**5) Slide holder: To hold the slide and prevent it from moving.**

**6) Stage**

Supports the slide/specimen

7) Stage control knobs

It moves stage forwards, backwards.

It moves stage right to left to adjust slide under objective lens.

8) Condenser: To collect the light in a cone shape from the light source to the object.

When using X40 or X100 lens raise the condenser up.

9) Iris diaphragm: Control the intensity of light that goes to the condenser.

When using X100 lens open iris diaphragm.

10) Course adjustment knob: Move the stage up and down rapidly to get approximate focusing.

11) Fine adjustment knob: move the stage slowly to get fine focusing.

# Immersion Oil

Microscope specimen is on a glass slide.

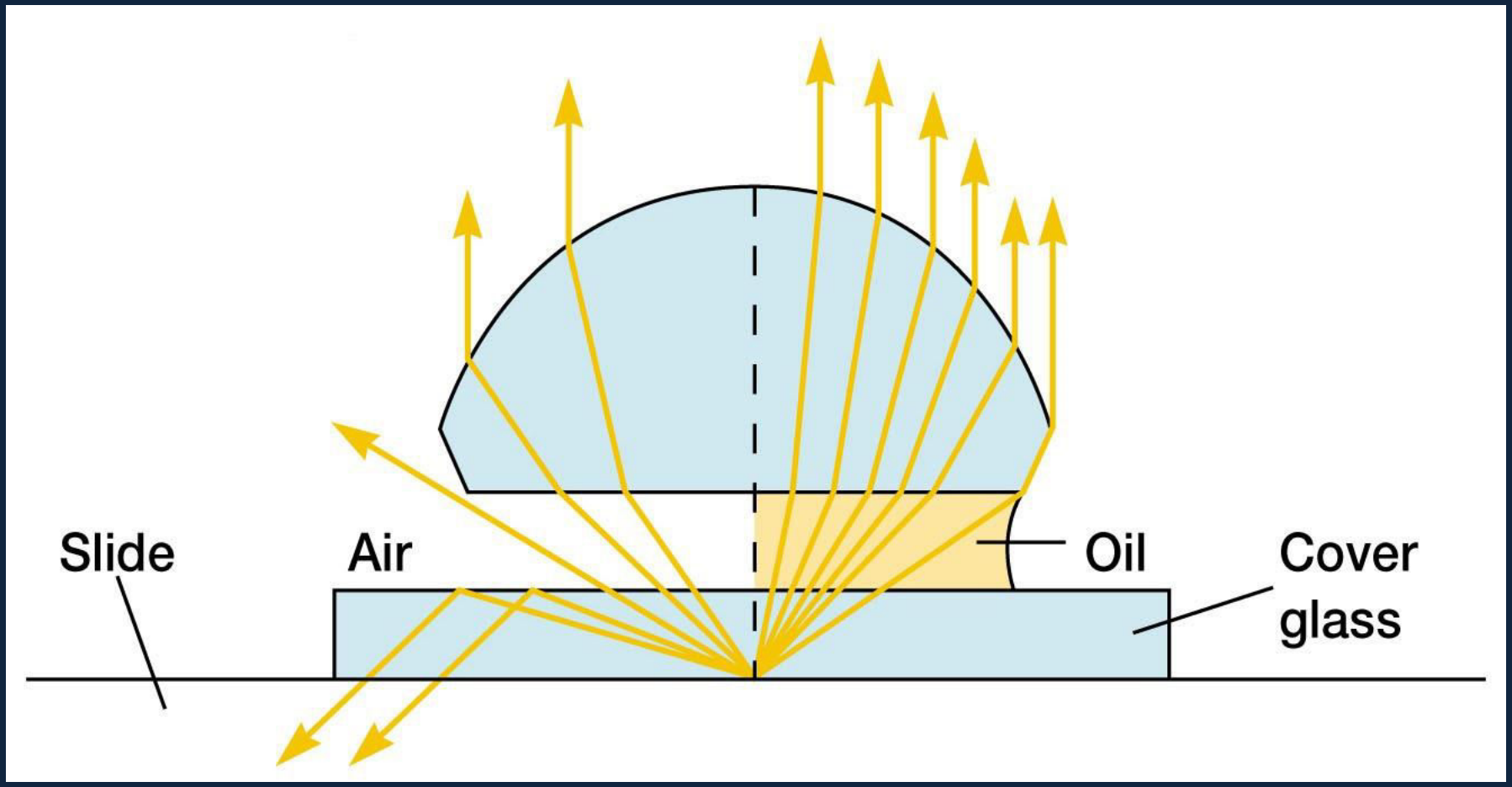
- Light passes through glass slide → air → lens.

  the light gets *refracted*

- At high magnification, this refraction of the light blurs the image
- To eliminate refraction between slide and lens:

  **Eliminate the air, replace with immersion oil**

  *(Immersion oil same index of refraction as glass)*

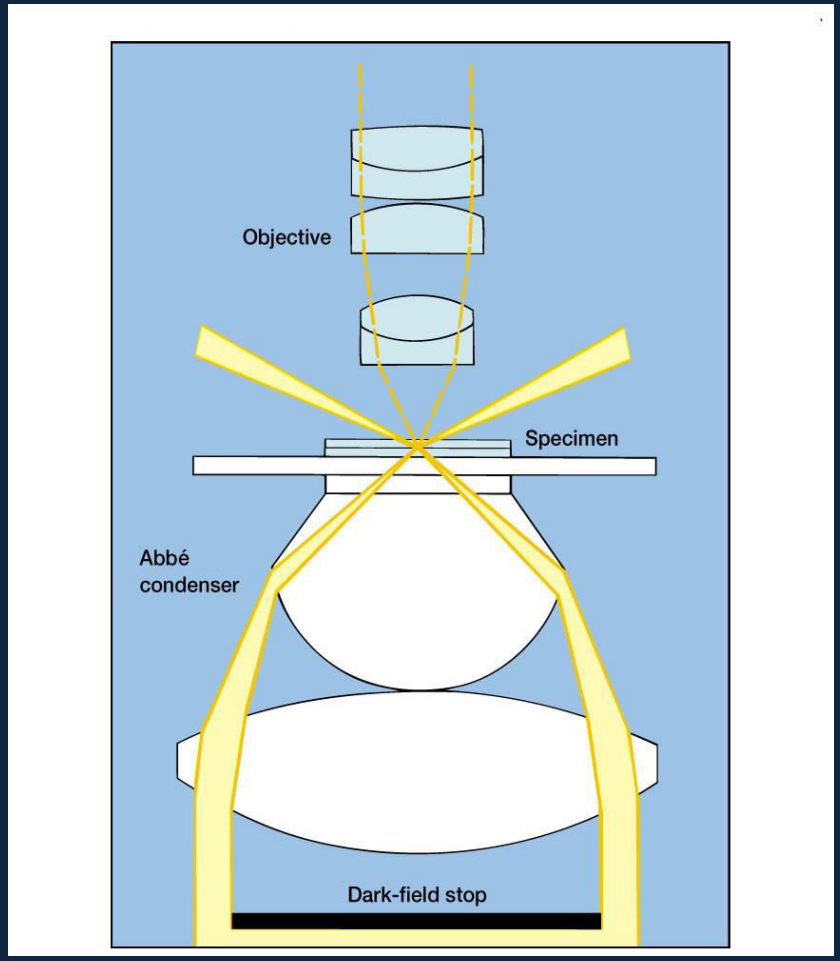

Slide
Air
Oil
Cover
glass

# Dark field microscope

- The condenser condenses the light on the object or specimen but out of the objective.
- The result is dark background and bright object.

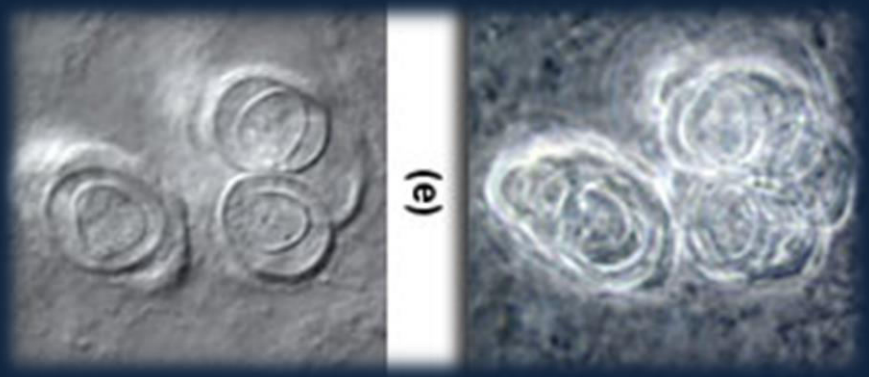
- Used:

  to see the motility of bacteria.


Objective
Specimen
Abbé
condenser
Dark-field stop

# Phase contrast microscope

- Produce contrast between the cell and the background.
- The cells appear darker against a brighter background.




# Phase contrast microscope

- It has special condenser and phase-plate which retards light waves that go through cells in specimen. This makes contrast between cells and background. The cells appear darker against a brighter background.
- **Used:**

  Excellent way to observe living cells in wet preparations.

# Inverted microscope

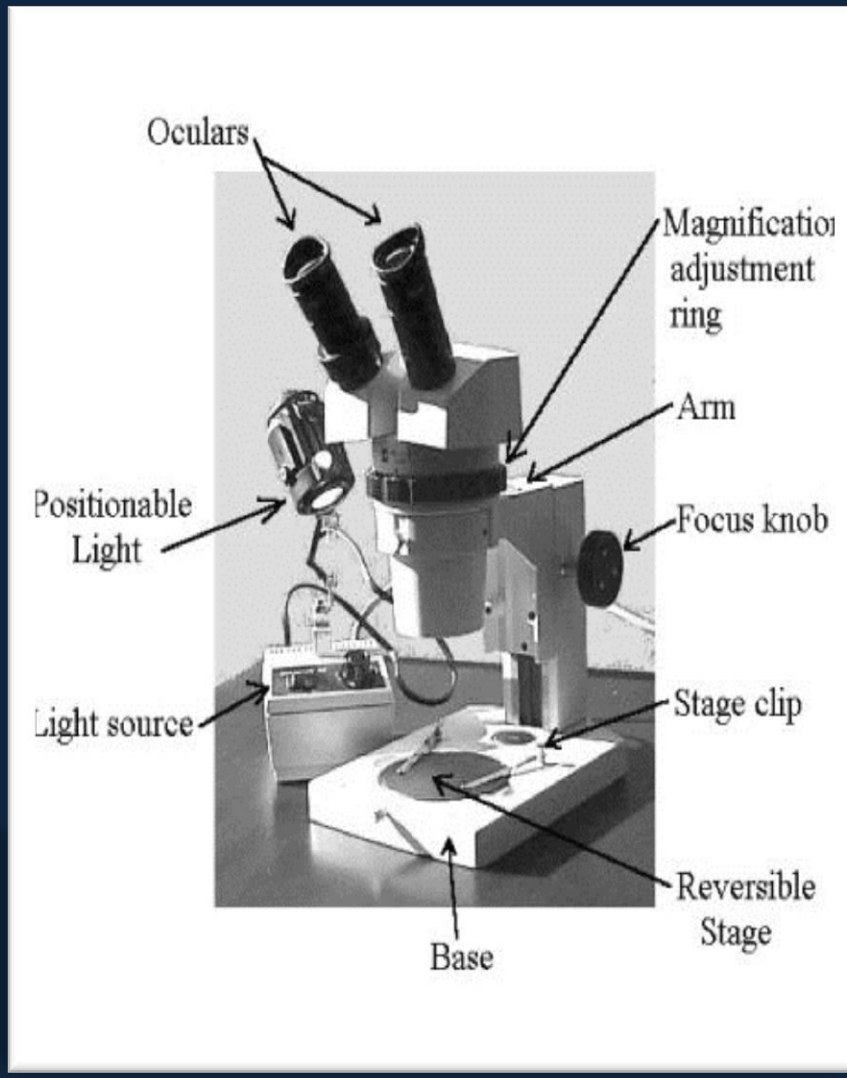
- There is a bigger working distance to allow use of cell culture flasks.
- There is a bigger working distance to allow use of cell culture flasks.
- **Used:**

  **to see the effect of virus on the cells (cell culture flasks).**

# Dissecting microscope

- It is a simple microscope.
- It has  oculars and stage only.
- It magnifies x10 only
- Used:

  in mycology to see the plate of fungi + dissecting of insect.

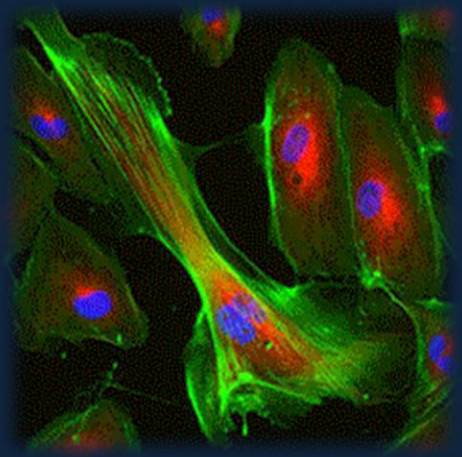
# Non-light microscope

1. Fluorescent microscope

2. Electron microscope (E.M)

# Fluorescent microscope

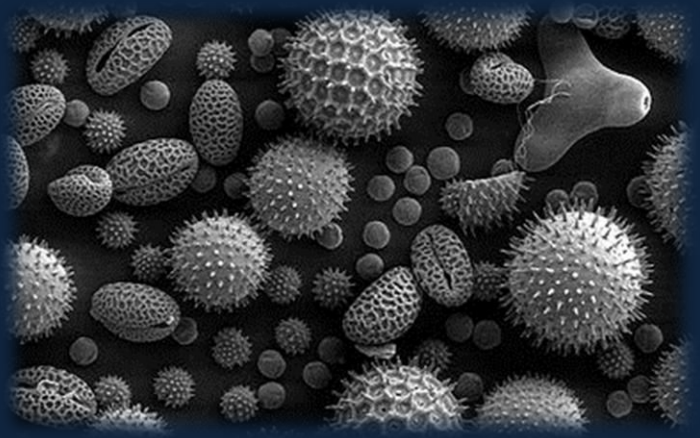
- Produce ultra violet light.
- The slides stained with fluorochrome.
- Used: in immunology.



# Electron microscope (E.M)

- Produce electrons (electron beam).
- Magnification= X100 000- X300 000
- Resolution= 0.0003 µm
- Used:

  to see viruses and the cell ingredients.



# Caring of Microscope

- Clean only with a soft cloth/tissue
- Make sure it's on a flat surface
- Be gentle with the microscope
- Carry it with 2 HANDS…one on the arm and the other on the base

# Thank You