#### Cell Size



#### Microscope Measurement



# How big is that object in the microscope?

#### Lesson Objectives

- Calculate the magnification using different objective lens.
- Differentiate between eyepiece graticule and the stage micrometer.
- Convert mm to micrometers.
- Calculate the cell length and breadth using the relationship between the size of the image, actual size and magnification.
- The structure and function of different parts of the microscope
- The difference between a light microscope and an electron microscope.



- Give the name and function of each structure labeled.
- A <u>Ocular lens/eyepiece</u>: used to look at specimen
- B <u>Fine adjustment</u>: to focus specimen under high power
- C Arm: to hold microscope
- D <u>Objective lens</u>: used to magnify image
- E <u>Coarse adjustment</u>: to focus specimen under low power
- F <u>Diaphragm</u> adjust amount of light



#### Light Microscope



A light microscope (also, optical microscope) is an optical instrument used to make objects larger in order to view their details. It uses light to illuminate the objects under view

#### **Electron Microscope**



An electron microscope is an optical instrument that uses a beam of electrons to make objects larger for a detailed view



#### Light microscope vs Electron microscope

- What is the difference between a light microscope and an electron microscope? A number of differences such as the source of light they use, their magnification level, cost, resolving power, among other factors sets these two types of microscopes apart from each other.
- VIDEO

# What is happening to the image as you increase the power of the objective lens?

#### Calculating total magnification

- If two lenses are always magnifying the specimen, how do you figure out the total magnification being used ?
- Total Magnification = ocular x objective
  = 10 x 4 (low power)
  = 40 (low power)

# How do we find the overall magnification of a light microscope?





<u>Nanometre</u>	<u>Micrometre</u>	<u>Millimetre</u>
5	0.005	0.000005
1	0.001	0.000001
1000	1	0.001
1 000 000	1000	1
3000	3	0.003
7	0.007	0.00007
500 000	500	0.5



The diagram below is a drawing of an organelle from a ciliated cell as seen with an electron microscope.



Answer = ...... 
$$A = \frac{102 \text{mm}}{\text{M}} = \frac{102000 \text{\mu}\text{m}}{20000} = \frac{102000 \text{\mu}\text{m}}{20000}$$
  
5.1

Calculating actual size:



#### Size of the magnified image > actual size

# To accurately measure the size of cellular structures we need a suitable scale:



### Field of View

- When you look into a microscope, the "field of view" is the visible circular area.
- What happens to your field of view when you increase the power of the objective lens?
- By knowing the size of the field of view (diameter), you can measure the size of objects in the microscope.
- The size of objects in the field of view is different at each magnification you have to calculate the diameters of the fields of view at each magnification.
- This process is called "calibrating your microscope"

## Estimating Specimen Size

- The area of the slide that you see when you look through a microscope is called the "Field of View".
- If you know how wide your field of view is, you can estimate the size of things you see in the field of view.



# Ideally, we need a scale we can see directly alongside the cells we are observing:



#### Eye piece graticule or reticule





- It is a glass or plactic disc with 8 divisions etched on to its surface and fitted into one eyepiece.
- The size of the eyepiece reticule is constant despite the change in magnification of the object.
- The value of each division varies with the change in magnification.

#### **Stage Micrometer**



- simply a microscope slide with a finely divided scale marked on the surface.
- 1 division= 0.01 mm
- 10 divisions= 0.1 mm
- 100 divisions = 1 mm
- 1 mm = 1000 micrometers.

#### Instructions

- Take sample of onion cell (peel of the onion)
- Add a drop of water
- Cover the subject glass with cover slip
- Fix it with the stage clips.
- Focus the specimen on low power objective lens.
- Now change to medium power objective lens and observe.
- Change to high power objective lens and observe.

# IMPORTANT FORMULA!Object Size = $\frac{field of view (in mm)}{number of "fits"} \cdot 1000$ Object Size = ? $\mu$ m

\*\* Remember that the field of view changes with each objective!.