What you should have learned from the microscope labs....



- Directionality
 - Items appear backwards and inverted

On Stage



In Microscope



NOT!!!!



- More Directionality
 - Items move opposite of the way you think they should
 - Left = Right
 - Right = Left
 - Up = Down
 - Down = Up

- How magnification affects the object you see...
 - You see less of the object, but greater detail.



- How magnifications affect your field of view...
 - The area of your slide that you see is reduced as you increase magnification.

• Determining the total magnification.

Total magnification = (Magnification of the objective lens) Magnification of the ocular lens) ie:

Total magnification = (Magnification of the ocular lens) Magnification of the objective lens)

Total magnification = (10X)(40X)Total magnification = 400X







 ie: if the lowest powered lens was a 60X objective lens and the higher powered objective lens was 180X:

of times greater the field of view is at the lower objective lens = <u>The higher powered objective lens magnification</u> The lower powered objective lens magnification

of times greater the field of view is at the lower objective lens = 180 X

60X

of times greater the field of view is at the lower objective lens = 3

High resolution

• Resolution:





The ability to tell two points
apart as separate items



- The shortest distance between two points that can still be distinguished.
- It is the capacity to show details clearly. Resolution allows the viewer to see two objects that are very close together as two objects rather than as one.

- Using the Diaphragm
 - Which setting shows specimen best?
- Depth of field
 - Sometimes you have objects on top of each other or swimming in a pool of water...because there is depth (the object is not flat) you will see the object 3-Dimensionally and will need to adjust the focus to view specimen at different depths
 - You can not focus on all depths clearly at the same time.





Figure 1

- Staining:
- How?
 - Make a wet mount slide.
 - Add stain to slide to one side of cover slip.
 - Use a filter paper on the other side of the cover slip to draw the stain through the organism.
- Why?
 - Helps make specimen or structures of specimen more visible.





- Measuring your field of view
- Measured in micrometers
 - Micron (μ)
 - So...1000 μ m = 1 mm
- # of micrometers = field of view in mm(1000)

ie: if your field of view was 12mm: # of micrometers = field of view in mm(1000) # of micrometers = 12 mm (1000) # of micrometers = 12000 μm

• High power will only see a portion of one milimeter, so you can not measure it exact enough. You must use a proportion to determine the number of micrometers.

High power field diameter = scanning power field diameter in micrometers x <u>scanning power objective</u> high power objective

• if we again use our 12 mm or 12000 μm example:

High power field diameter = scanning power field diameter x <u>scanning power objective</u> high power objective

High power field diameter = $12000 \ \mu m \times \frac{4X}{40X}$

High power field diameter = $1200 \ \mu m$

- Measuring Specimen (Version 1)
 - Determine what portion of the field of view it covers. That fraction times the field of view diameter in micrometers is the approximate length of the organism in micrometers.

approximate length of the organism in micrometers = portion if the field of view (size of field of view) approximate length of the organism in micrometers = $0.25 \times 12000 \mu m$ approximate length of the organism in micrometers = $3000 \mu m$



- Measuring Specimen (Version 2)
- Estimate the number of the specimen that would fit across the diameter of the field of view and divide the field of view diameter in micrometers by your estimate.
- ie: if you estimate that 5 cells would fit across the diameter of your field of view, and your field of view was 12000µm:

approximate length of the organism in micrometers = $\frac{\text{field of view diameter in micrometers}}{\text{estimate of # of specimen that would fit}}$ approximate length of the organism in micrometers = $\frac{12000 \text{ micrometers}}{5}$ approximate length of the organism in micrometers = $2400\mu\text{m}$

- Determine the diameter of your circle representing the field of view and divide your field of view measured in lab 5 by this number.
- This needs to be done for each field of view and each circle.
- Draw a line in your circle representing the field of view for your drawing that is 1cm in length. Mark that it is equal to the µm you calculated it to represent.

