

# Lab 2- Microscopes

In today's lab you will learn the proper use and care of a microscope. You will also have an opportunity to examine some prepared slides of cells and tissues as well as your own cells and some items you will collect from outside.

## Activity 1- Dissecting Scope

**1. Find and examine all of the microscope parts on YOUR microscope and familiarize yourself with their functions.**

### PICKING IT UP:

Carefully pick up your scope by grasping the **arm** and supporting the **base** with the other hand held flat, carry to your desk. (Do NOT push fingers up underneath base, you may push out the **stage plate**.) Make a second trip to pick up a **light** with **transformer**. Note where they should be returned, and the way that the cords are wrapped on the light and transformer.



- 1) Ocular lenses (Eyepieces)
- 2) Body w/objective lenses
- 3) Focusing knob
- 4) Arm
- 5) Stage plate
- 6) Stage clips
- 7) Light source
- 8) Mirror and mirror control knob
- 9) Magnification control knob

**Remember: Microscopes have an ocular lens in the eyepiece that has a magnification of 10X.**

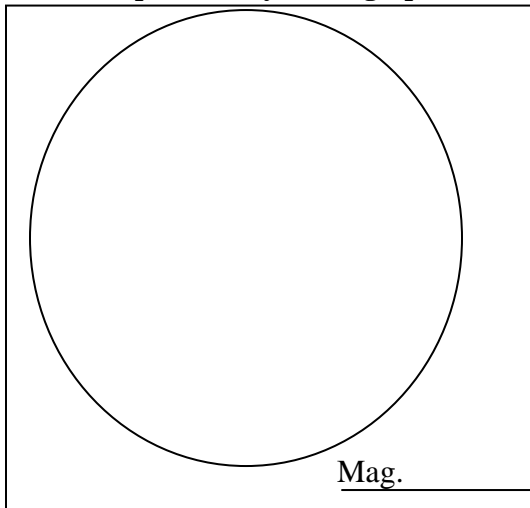
**So, TOTAL MAGNIFICATION is always 10 times the power of the objective lens you have in place.**

**4X objective = 40X total magnification**

## SETTING UP A DISSECTING MICROSCOPE:

1. Remove the dust cover (if there is one), raise **optical head** by rotating **focusing knob** until back of optical head is just above top of **arm**.
2. Unwrap cord from around **light housing**, and, for top lighting, insert light into hole in back of head (or for bottom illumination, into base if equipped with **transilluminator**.)
3. Unwrap cord from around **transformer**, set **voltage selector** to off, plug into electrical outlet.
4. Plug cord from light into back of transformer, turn **voltage selector** on to the #1 setting. Did the light go on? If not, report to your professor.
5. Place specimen in center of field, turn **zoom knob** on top of optical head to 1.0. (This will give an effective magnification of 10X since the oculars are 10X.)
6. Looking only through the **right ocular** with right eye, focus on specimen with the focusing knob. When finely focused, look only through the left ocular with the left eye, and focus by rotating the **adjustable left ocular**. The specimen should now be in focus for both eyes.
7. Adjust for your own **inter-ocular distance** by spreading apart or squeezing together the oculars so that a single three-dimensional image is seen, with no black areas.
8. Adjust the magnification by rotating the zoom knob so that desired detail is clear on the specimen. You may elect to use either the black or white side of the face plate for optimum visibility. Adjust the amount of light on the specimen by adjusting the setting on the transformer. To prolong the life of the bulb, use the lowest transformer setting which yields satisfactory illumination. Use lens paper *only* for cleaning lenses.

### 1. Draw a picture of your fingerprint.

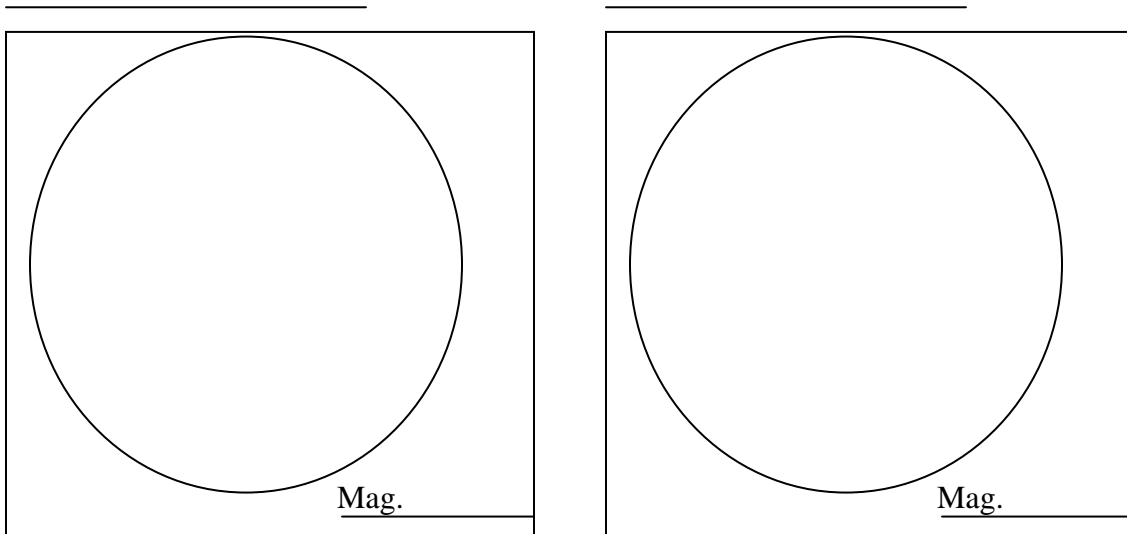


Compare it to others at your table. Are any that are similar?

Move your finger to the right. Which way does the image move in the microscope?

Move your finger towards you. Which way does your finger move in the microscope?

**2. Draw some other biological objects of your choice. Be sure to note the magnification and label your drawings.**



- What is the magnification of the eyepiece lens? \_\_\_\_\_
- What is the magnification range of this microscope? \_\_\_\_\_
- What kind of specimens is it best used for viewing? \_\_\_\_\_

**PUTTING THE DISSECTING SCOPE AWAY:**

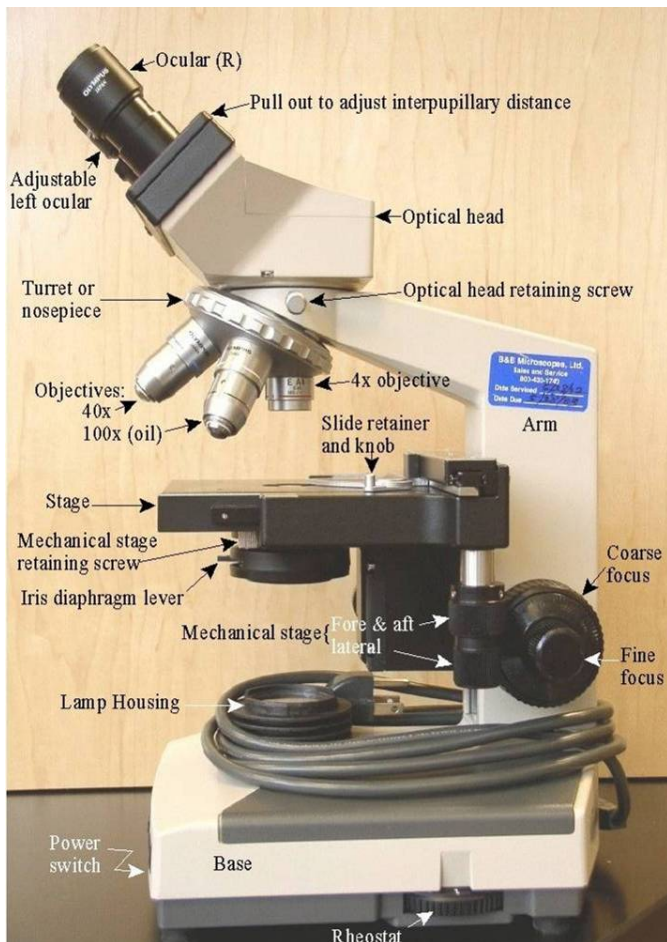
1. Turn off the light, unplug transformer from outlet. Carefully wrap cord snugly around the transformer, tuck in end so that it stays put.
2. Remove light from scope, wrap cord around light body, tuck in. Push into larger side of ring on transformer extension arm, replace in cabinet.
3. Lower optical head of scope until it almost touches base, return to the cabinet.

## Activity 2- Compound Light Microscopes

### Microscope Rules: READ FIRST!

- **To carry a microscope:** grasp the arm firmly, lift and support under the base with other hand, set on a cleared desk. Remove and store its dust cover in cabinet under desk.
- **Clean the lenses: use ONLY lens paper.** Polish the objectives and oculars: breathe on them lightly for moisture. (Polish slides with Kimwipes.) If the view is still foggy, ask for help.
- Always begin slide set-up with the **stage lowered** and the **lowest power objective (4x) in place.**
- **Focus initially only by LOWERING the stage** to the focal point using the coarse focus. NEVER raise the stage using the coarse focus during focusing. (The objective may ram the slide, which can damage both.)
- **Use only the fine focus with higher power objectives.** Make only minor changes in focus when necessary with the fine focus knob. If you totally lose focus, return to a lower objective to find the focal point. Do not use the 100x objective unless you have received specific instructions on its use. Carefully follow microscope use instructions.

### 1. Find and examine all of the microscope parts on YOUR microscope and familiarize yourself with their functions.



**Remember: Microscopes have an ocular lens in the eyepiece that has a magnification of 10X.**

**So, TOTAL MAGNIFICATION is always 10 times the power of the objective lens you have in place.**

**4X objective = 40X total magnification**

**Describe the functions of each of the following parts of the microscope:**

- Eyepiece\_\_\_\_\_
- Revolving Nosepiece\_\_\_\_\_
- Stage and stage clip\_\_\_\_\_
- Stage hole\_\_\_\_\_
- Iris diaphragm lever\_\_\_\_\_
- Coarse focus knob\_\_\_\_\_
- Fine focus knob\_\_\_\_\_
- Low power objective lens\_\_\_\_\_
- High power objective lens\_\_\_\_\_

## **VIEWING SPECIMENS UNDER THE MICROSCOPE**

**2. Obtain a letter “e” slide. Place it on your microscope so that it appears right side up to you when you look at it with your naked eye. Using the directions below, focus the slide and draw what you see.**

### **PLACING THE SLIDE ON THE MICROSCOPE:**

- **Turn the coarse focus knob so that there is plenty of space under lenses for you to place the slide the stage.** Rotate the 4X objective into position if it is not already there.
- **Handle all slides by the *edges only*.** Pick up a prepared slide.
- **Open the slide retainer** by pressing the jaw lever to the right. **Place the slide on the stage** with the label to the left. Using the mechanical stage, move the slide until the specimen is over the optic center of the stage. (View from side and front to get a best estimate of center.)
- **The slide should be sitting directly on the stage, held in place by the retainer.**
- **Turn the course focus knob while viewing *from the side*** until the objective almost touches the slide or until the stage stops.

### **FOCUSING:**

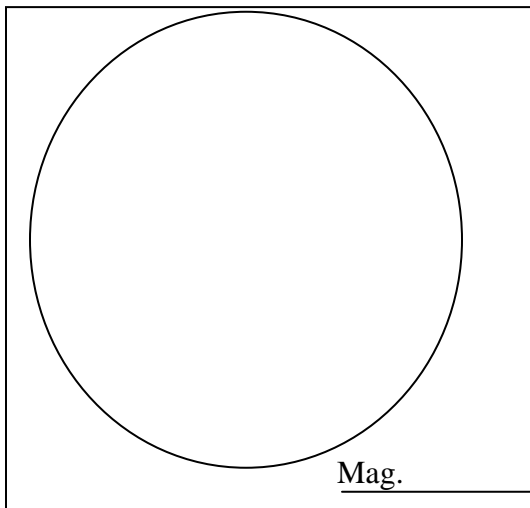
- **Always start with the lowest power objective lens.**
- **Do not increase the magnification until you are focused in the lower power.**

- **Turn the coarse focus knob *away from you* to focus** as you look through the oculars until the image becomes sharp (in focus). If it never appears, try to re-adjust the position of the slide nearer the center, and repeat the 4X focusing procedure.
- **BLACK LINE TRICK:** Another trick is to adjust the slide on the stage so that the edge of the slide or cover slip is directly below the lens, then look through the lens for the black line created by the glass edge. It is large and easy to see. Once in focus on the edge of the slide, you can adjust the slide so that the specimen will be in view under the lens.

**INCREASING THE MAGNIFICATION: (only once focused with 4X lens)**

- **Rotate the nosepiece to select 10X objective (100X total mag.).** Center the image again if needed. If you lose the view of the specimen, go back to lower power to find it again and center more carefully. Be sure not to touch the slide or stage when you shift between the lenses. As you go up in magnification you will probably need to increase the amount of light. You may need to make minor adjustments in focus with the fine focus knob.
- **DO NOT USE THE COARSE FOCUS KNOB BEYOND 100X!**
- **The same procedure is repeated to increase the magnification to 400X as needed to view smaller specimens.**

**Draw the letter “e” it at 100X total magnification.**

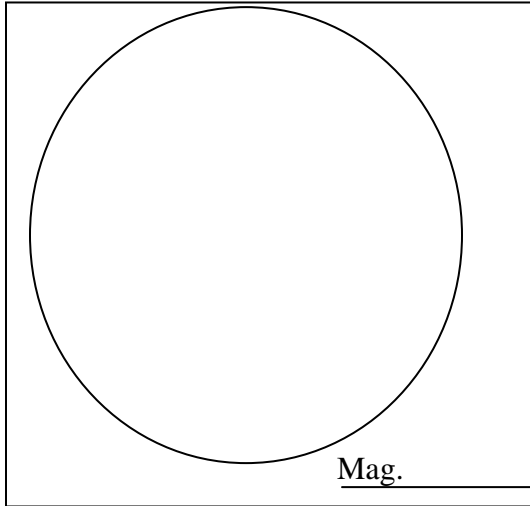


Move the slide to the right. Which way does the image of the “e” move?

Move the slide towards you. Which way does the “e” move?

**3. Obtain a prepared slide of Red Blood Cells. Repeat the focusing procedure as above. Red Blood Cells are VERY small (~8 $\mu$ m). You will not see them at all at low and medium power magnification. You will need to use the BLACK LINE TRICK and view them at 400X total magnification.**

**Draw a picture of the red blood cells as they appear through your microscope at 400X total magnification.**

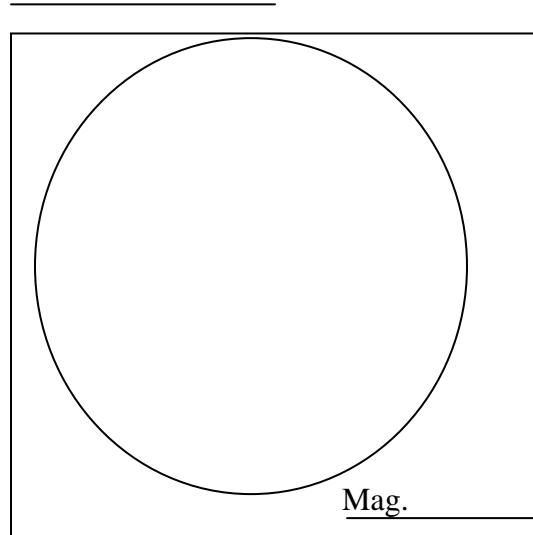
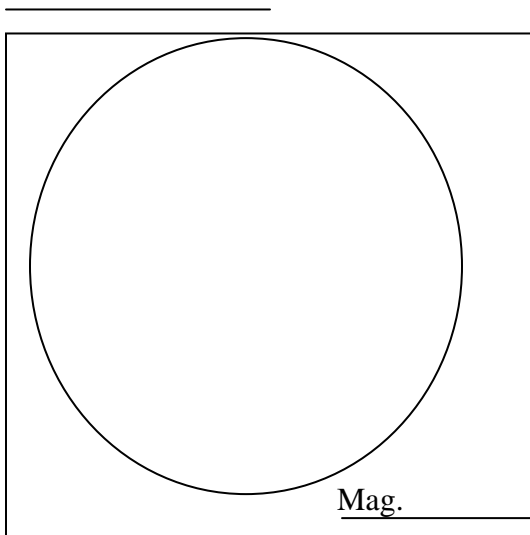


What is the magnification of the ocular lens?

What is the magnification of the low power objective? The high power?

What does "total" magnification mean?

**3. Obtain prepared slides of other tissues and cells and practice bringing them into focus at different magnifications. Draw what you observe. Be sure to label your drawings and write the magnification. Draw to scale.**

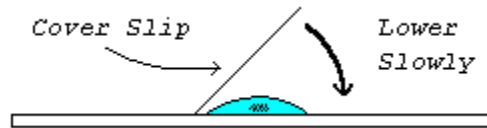


## Activity 3- Pond Water Slides

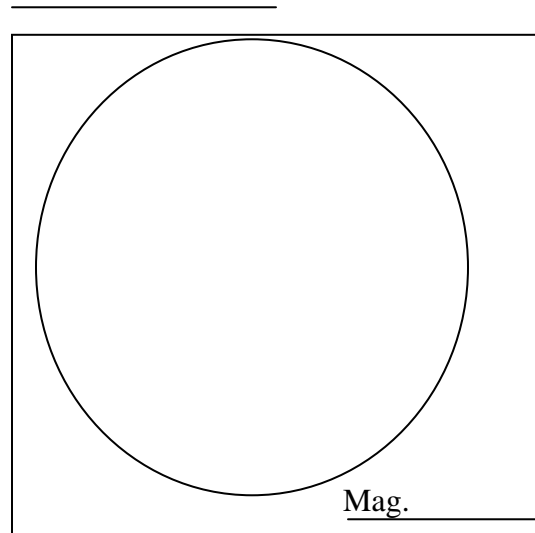
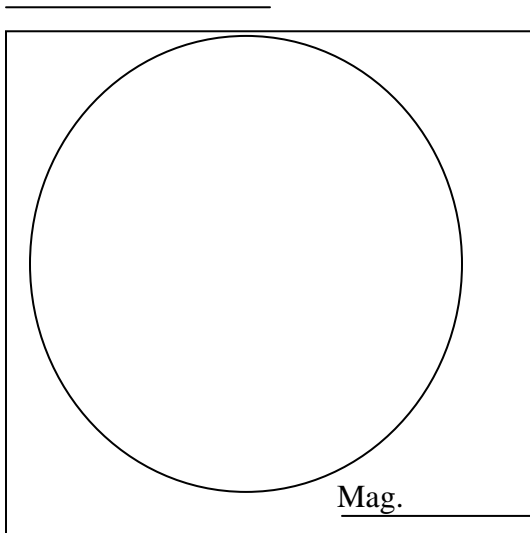
In this activity you will make a slide of the pond water and observe it with the microscope. It's a good idea to stir up the jar or get your sample from the bottom of the jar (where all the organisms are located). Use **Detain/Protoslo** if available to slow the critters down.

### HOW TO MAKE A WET MOUNT

- Obtain a depression slide and a cover slip- *Always hold these items by the edges.*
- Check the depression slide to make sure the concave side is facing upwards, so it forms a little "bowl". Clean the slide off with a soft cloth or tissue after touching it with your fingers.
- Set the slide down on the countertop and place two or three small drops of pond water in the depression.
- Hold the cover slip (by the edges) at an angle, so that the bottom edge of it touches the liquid on the slide. Then slowly lower it over the specimen. See diagram below. This technique is used to help avoid trapping air bubbles under the cover slip.



**Observe the organisms that live in the pond water. Draw a couple of these organisms. Are they prokaryotes or eukaryotes? How can you tell?**



**9. Why didn't we need to stain the organisms we saw in the pond water? Hint: what color were they and why?**

**10. Were these prokaryotes or eukaryotes? How do you know?**

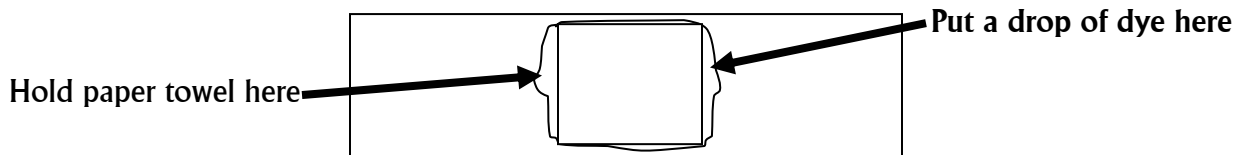
### **Activity 4- CHEEK CELLS**

To prepare a wet mount using your own cheek cells, you will proceed as above with the following exceptions:

Obtain the following items:

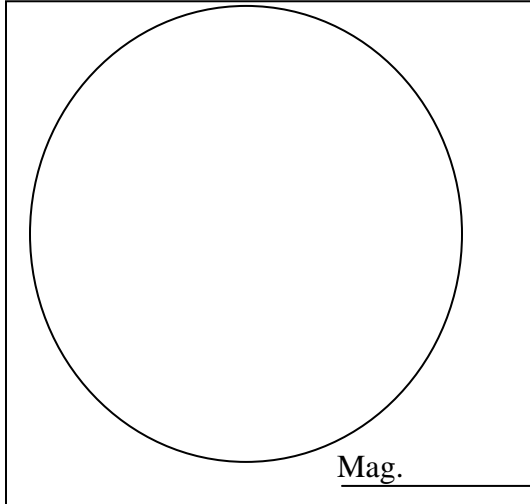
- A regular flat slide and a cover slip.
  - A couple tooth picks
  - A bottle of methylene blue dye
  - A bottle of sterile saline
  - A small 2-3 inch square piece of paper towel
- 
- Place the slide on your lab table and put a very small drop of saline on the slide.
  - Using your toothpick, scrape the inside of your cheek lightly.
  - Tap the toothpick into the saline on the slide to get some cells to fall into the drop.
  - Place the cover slip over the drop of saline as you did for the pond water.

**Your slide should look like this:**



- To stain the cells, place a small drop of Methylene blue dye on the slide **NEXT TO THE COVERSLIP**. Hold the paper towel on the slide touching the liquid on the other side of the cover slip. This will cause the dye to “wick” under the cover slip and stain the cells.

**Draw your cheek cells. Be sure to label your drawing and note the magnification. Also, draw any “other” cells you see in this preparation.**



Are your cheek cells eukaryotic or prokaryotic?  
How can you tell?

Are there any other cell types in your sample?  
Do you think they are eukaryotic or prokaryotic?  
Why?

#### **HOW TO RETURN MICROSCOPE TO STORAGE:**

- Lower stage fully
- Return the 4x objective to the viewing position
- Turn light all the way down to lowest setting, turn off power switch
- Wrap cord snugly, no sharp bends, tuck in plug
- Return to storage cabinet, arm towards you.
- Report any problem with the instrument immediately.

#### **SLIDE DISPOSAL:**

- Prepared slides must be returned to the correct trays
- Wet mounted slides should be placed into the slide disposal beaker near the sink

### **CLEAN UP YOUR WORK AREA!**



**PUT AWAY ALL THE LAB EQUIPMENT YOU HAVE USED IN THE SAME MANNER YOU FOUND IT.**