Lab 1 Microscopy & Cells

OBJECTIVES:

- ❏ Learn how to use and care for the compound microscope.
- ❏ Identify and explain the function of the parts of the compound microscopes.
- ❏ Prepare wet mount slides.
- ❏ Understand the terms used in this lab: field of view, working distance, light intensity, depth of field, magnification, etc.
- ❏ Understand of the following terms: prokaryote, eukaryote, cell, cell membrane, cell wall, cytoplasm, membrane-bounded organelle, nucleus, chloroplast, mitochondrion, nucleus, photosynthesis, respiration.
- ❏ Compare and contrast prokaryotic and eukaryotic cells (i.e. be able to describe similarities and differences).
- ❏ Compare and contrast animal and plant cells.
- ❏ Give two general characteristics of prokaryotic cells.
- ❏ Distinguish among the three morphological types of bacteria.
- ❏ Describe the subcellular structure of a typical bacterium and review the function(s) of each structure.
- ❏ Identify the structures of a typical plant cell and review the function(s) of each structure.
- ❏ Identify the structures of a typical animal cell and review the function(s) of each structure.
- ❏ Observe some interesting organisms.

We humans are highly visual creatures, obtaining much of our information about the world around us by using our eyes. To understand many things we need to *visualize* them: we must *see* them. Not surprisingly, then, some of the most useful tools in biology, and indeed in science in general, are those that allow us to visual objects, processes, and phenomena. This is one of the main advantages of the powerful computers of today, because their power allows us to visualize things, and thus explore them, as never before possible. But even that technology cannot replace one of the most fundamental tools of biology, the **microscope**. With the microscope we can see things that are otherwise too small for us to see, and thus study them.

There are many different kinds of microscopes. The kind we will be using will be the light microscope. In the first part of today's lab we explore how to use and care for this most basic of biological tools.

Understanding the nature of cell structure and function is important to an understanding of organisms. All organisms are composed of cells, whether they exist as single cells,

colonies of cells, or in multicellular form. Cells are usually very small, and for this reason, a thorough understanding of subcellular structure and function has been possible only through advances in electron microscopy and molecular biology.

In the second portion of this lab you will explore with your lab partners the different basic kinds of cells that exist. You will be comparing specimens of cells in terms of size, color, and other characteristics. We will then discuss these differences.

GENERAL

1. Work in pairs. However, each student in the pair should have his/her own microscope and do the procedures.
    - If you do not have experience with a compound microscope, pair up with someone who does.
    - If you have experience with a compound microscope, pair up with someone who does not.
    - Please pay careful attention to how you use and care for these instruments. Failure to properly handle the microscopes **may lead to a deduction of points from your lab score**. Microscopes are delicate and expensive optical instruments and must be handled properly and carefully. Prepared slides are also expensive and must be used thoughtfully as well.
2. Do not keep your coats, bags, and other personal belongings (items not used in the lab) on the lab table. You will need room on the lab bench for your lab materials, lab manual, notebook, and microscope.
3. Obtain microscopes from the microscope cabinets. Always carry the microscope in an upright position; the lenses you look through slide into the body tube and can easily fall out (slides left on the microscope by inconsiderate students can also fall to the floor if the microscope is tilted). With one hand, grasp the arm of the microscope and place the other underneath to support the base. Please be patient while you are waiting for students ahead of you to get their microscopes and please do not get in the way of individuals carrying microscopes.
4. Unwrap the cord and plug the microscope into an outlet. Save the rubber band! Arrange the cord so that it does not hang over the edge of the bench or can get snagged by someone, sending it crashing to the floor.
5. Calculating magnification:
    - Multiply the power (magnification) of the eyepiece by the power of the objective lens to obtain the total (total) magnification.
    - The power of the objective lenses is written on them. It usually is followed by an "x", and is an integer, NOT a decimal number.
    - The power of the eye piece is 10x.
    - Always include in any sketches the <u>total</u> power or magnification under which you observed the specimen. Always include the "x" as part of the magnification (e.g., 10x, 400x).

6. Use lens paper ONLY to clean ocular and objective lenses!
7. When finished, be sure that
    - the scope is turned off.
    - all slides are removed and returned to their proper container in the correct orientation.
    - the stage is wiped clean.
    - the scope is set with the lowest power (scanning) objective in place.
    - the stage is lowered (or objectives raised) as much as possible.
    - the cord is properly secured.
    - the scope is returned to its proper place in the microscope cabinet.
8. If your microscope does not work, tell your instructor, or the lab technician, or put a note on it indicating what you think is wrong with it. If you find a slide on your microscope stage please give it to your instructor or place it in the lost slide box.

PROCEDURES

1. Your instructor will lead you through this part. Please follow instructions and DO NOT PROCEED FURTHER THAN THE INSTRUCTOR HAS ASKED YOU TO GO!

(A) Prepare a "wet mount" slide with of a newspaper letter "e".

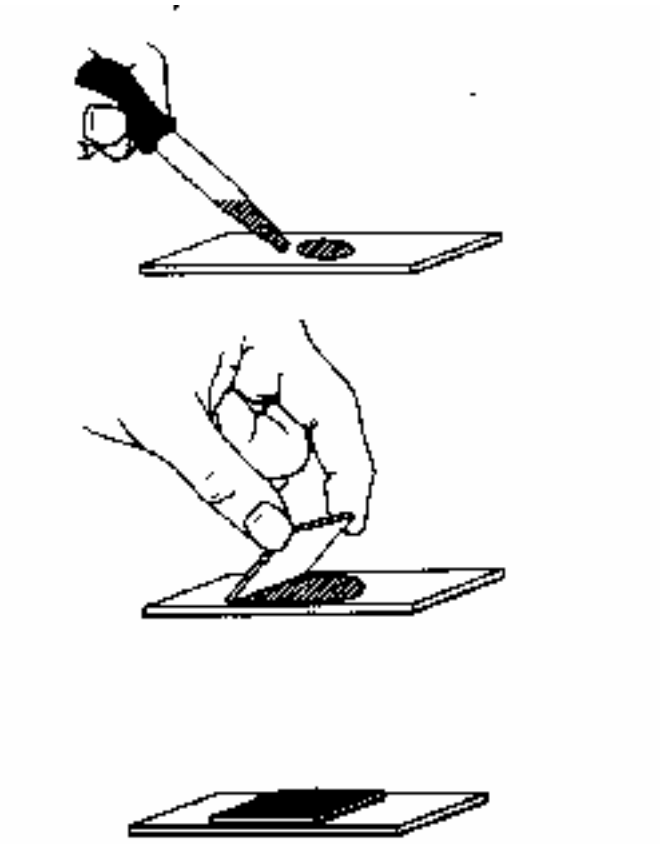
Step 1. Obtain a clean glass slide (you may want to clean it again yourself with a *Kimwipe*). Be careful to hold the slide by the edges to avoid smudging it.

Step 2. Put one drop of water on the slide in the center. It doesn't matter which side of the slide you use.

Step 3. Obtain a letter "e" by wetting a cotton swab and touching it to a single letter "e" in the container, which will stick to it.

Step 4. Touch the "e" in the center of the water droplet on the slide. The "e" should float off and into the water. Be sure the "e" is still right side up—sometimes they flip over. If necessary, use a pair of tweezers (forceps) to turn back over.

Step 5. Obtain a cover slip. Hold the cover slip at an angle and place one edge of the cover slip at the edge of the drop of water (see figure below). Allow the water to "wick" along the edge of the cover slip before slowly lowering the cover slip (to avoid trapping air bubbles).



(B) Determine how to use the microscope.

- Your microscope should already be plugged in, as per the instructions above.
- Leave the microscope on <u>low</u> (scanning) power – that means having the shortest lens in place. It should already be set this way; if not, ask the instructor to help you. What magnification is this? (see instructions above for calculating magnification)
- Your instructor will lead you to answer the following questions one after the other to direct your exploration of the microscope. Again, <u>please do not get ahead of the instructor</u>. Don't worry about the names of the parts of the microscope yet -- for the "where" questions, we want you to find the actual location (be able to point to it). Write your answers on this page.

1) Where do you put the slide?

2) How do you turn the microscope on?

3) What part of the microscope does the light come from?

4) Where do you look into the microscope?

5) How do you change the distance between the two lenses that you look into?

6) How do you focus? (Where are the focus knobs?)

7) What happens on the microscope when you focus (what moves)?

8) What do the two focus knobs do differently?

9) Is the image of the "e" right-side up?

10) How do you increase the amount of light? How do you decrease it?

11) How do you move the slide around without touching it?

13) If you move the slide to the right, which way does the image in the microscope move?

13) If you move the slide to the left, which way does the image move?

14) If you move the slide up (away from you), which way does the image move?

15) If you move the slide down (towards you), which way does the image move?

16) Does your microscope have a pointer that you see when you look through it?

STOP HERE until instructed to continue. When everyone is ready, the instructor will go over what you have learned so far, and will tell you the names of the parts of the microscope.

(C) The instructor will next instruct you on how to change the power of magnification on the microscope, and how to focus.

- Increase magnification (power) to the next strongest (this is called "low power" objective). What magnification is this? _____
- Answer the following questions, while still using the "e":

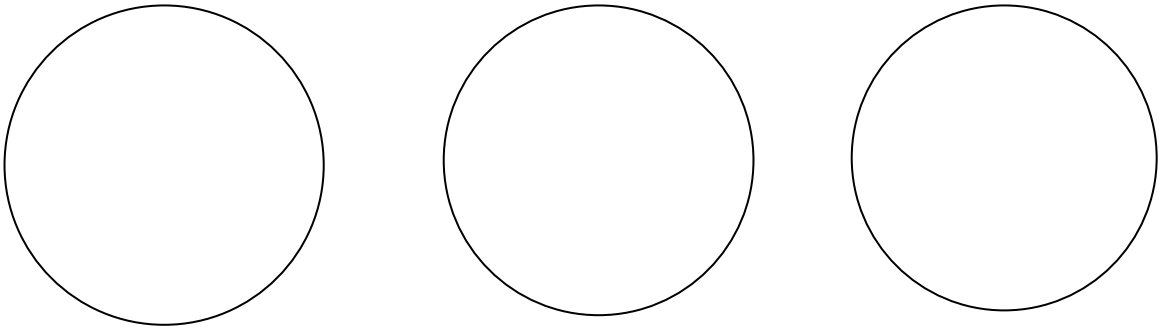
1) What are the names of the parts of the microscope that change the magnification?

2) After you increase magnification, which focus knob should you use? Why?

3) After you increase magnification, is the image still in focus?

4) Is it as bright as it was under lower power?

- Now increase the magnification one more time, to the next strongest (this is called "high power" or "high dry" objective) and observe the "e". Make three sketches of the "e", one at each magnification. Make the sketches in the circles below.



Magnification _____ _____ _____

Wash and dry the "e" slide and return the clean slide to the slide tray from which you got it.

STOP HERE until instructed to continue. When everyone is ready, the instructor will go over what you have learned so far.

(D) LIVING CELLS!

GENERAL PROCEDURE

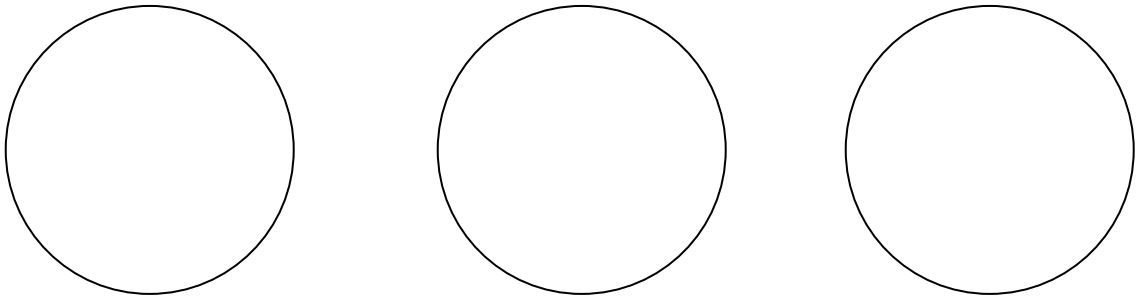
1. Work in pairs. You may set up slides for each other, but each student should complete their own set of drawings.
2. Follow the specific procedures below to prepare specimens, if necessary.
3. When studying the specimens, sketch them in the space provided, note the final total magnification under which you observe them, and note the shapes and colors in the space provided. Also describe any structures you observe within the cells, pointing them out with arrows in your sketches.

SPECIFIC PROCEDURES

Bacterial Cells

1. Examine the oil immersion demonstrations of prepared slides of the bacterial specimens. These are set up at the side of the room. These bacteria are dead and have been stained with a colored dye to make them more visible.

2. **Draw simple sketches of these prokaryotes** focusing on shape of the cells. Make the sketches in the spaces below. For each, note the specimen name, the total magnification, a word or two describing the shape, the color, and a description of any structrues that you can see inside the cells. If you can't see any sturctures, write in "none".



Specimen _____ _____ _____

Magnification _____ _____ _____

Shape _____ _____ _____

Color _____ _____ _____

Description of any structures you can see inside the cells

_____ _____ _____

_____ _____ _____

_____ _____ _____

Animal Cells

You will examine cheek cells obtained from your mouth (human squamous epithelial cells).

NOTE: Because you will be isolating cells from your cheek, and the cheek cells are considered biohazardous material, please follow the clean-up instructions carefully!

Prepare a "wet mount" slide with of your cheek cells.

1. Obtain a clean glass slide (you may want to clean it again yourself with a *Kim wipe*). Be careful to hold the slide by the edges to avoid smudging it.
2. Place one (1) drop each of water & methylene blue in the center of a clean glass slide.
3. Gently rub the inside of your cheek with the blunt end of a toothpick.
4. Stir the cheek material (you may not be able to see any) into the methylene blue in the center of the slide. If the stain appears very dark, add a drop of water.
5. Apply cover slip. (Touch one edge of the cover slip to the edge of the drop of liquid. Hold the cover slip while the liquid runs along the entire edge. Then gently lower the cover slip over the specimen.
6. Dispose of toothpick in the beaker provided.

Examine your cheek cells.
These cells are flat with irregular shapes or contours. Some of the thin, flat edges of the cells may be folded in your preparation. Many of the cells will probably be clumped together. It is difficult to observe the cells clumped in large masses or groups. Bacteria may be present on some cells; these cells will appear to be covered by small, dot-like or stick-like structures.

7. Place the cheek cell slide on the microscope and examine it under the scanning objective. Focus the cells and adjust the light intensity.
8. Find an area of the slide where the cells are isolated, separate from each other. Select cells for closer examination that have centrally-located, darkly-stained nuclei.
9. Observe these cells using the low power objective. Move the slide so that an individual cell is in the center of the field of view. Then move the high power objective into place and bring the cell into focus using the fine-focus knob.

3. **Draw a single, isolated squamous cheek cell** (at high power) in the space below. Include the name of the organism, magnification, color, a description of the shape, and a **description of any structrues** that you can see inside the cells. If you can't see any sturctures, write in "none".

Specimen_____

Magnification_____

Shape_____

Color_____

Description of any structures you can see inside the cells: _____

_____

_____

Plant Cells #1

1. Prepare a wet-mount slide of an *Elodea* leaf.
2. **Sketch a representative *Elodea* cell** as observed under high power. Include the name of the organism, magnification, color, a description of the shape, and a description of any structrues that you can see inside the cells. If you can't see any sturctures, write in "none".

Specimen_____

Magnification_____

Shape_____

Color_____

Description of any structures you can see inside the cells: _____

_____

_____

3. Do any structures move? ___ Describe their movement.

Plant Cells #2

1. Use a razor blade to slice a piece of tissue, as thin as possible, from a potato. Be careful not to cut your fingers. Prepare a wet-mount slide; use a drop of water.
2. Study the slide at low power (10x objective) and then at high power (40x objective).
3. **Sketch a representative potato cell** as observed under high power on the next page. Include the name of the organism, magnification, color, a description of the shape, and a description of any structrues that you can see inside the cells. If you can't see any sturctures, write in "none".

Specimen_____

Magnification_____

Shape_____

Color_____

Description of any structures you can see inside the cells: _____

_____

_____

4. Plants store excess energy as molecules. Often, the storage molecule is starch, which is a carbohydrate. Add a drop of Lugol's solution (I_2KI) to the side of the cover slip and touch a piece of Kimwipe to the other side of the cover slip to draw the stain solution under the cover slip. The iodine in the Lugol's solution will stain starch blue/purple. Does this solution stain the cells as it reaches them? Are there any structures that turned blue/purple?
5. Sketch a representative potato cell as observed under high powers in the space provided below. Include the name of the organism, magnification, color, a description of the shape, and a description of any structrues that you can see inside the cells. If you can't see any structures, write in "none".

Specimen_____

Magnification_____

Shape_____

Color_____

Description of any structures you can see inside the cells: _____

_____

_____

ANALYSIS

Work with your lab partner to answer the following questions. Be prepared to share your answers with the class!

1. How are ALL the cells the same?

2. How are bacterial cells different from plant and animal cells?

3. What structures are missing from animal cells that are in *Elodea* cells?

4. How are *Elodea* cells different than potato cells? What does this tell you about their function?