LABORATORY EXERCISE #5

INTRODUCTION TO THE COMPOUND LIGHT MICROSCOPE (Nedwidek revised)

INTRODUCTION

One of the biologist's most important laboratory tools is the *compound light microscope* (Greek: micron = small and scopos = aim) which consists of two converging lens systems: the objective and the eyepiece. It is an instrument used to magnify objects that are too small to be seen with the unaided eye.

Cells are the basic units of living things. Today, you will compare the structure of onion cells to the cells of your own inner cheek epithelial tissue. Your observations will show how these specific plant and animal cell structures allow then to survive The onion is an underground, storage structure made up of thickened modified leaves called **scales**. The cells we will observe today come from a thin, translucent tissue which lines the scales' inner curve. The individual cheek cells can be easily scraped off the inner cheek using a toothpick.

STUDENT OBJECTIVES

1. Learn proper handling of the compound microscope.
2. Know and label the names and functions of the parts of the microscope.
3. Prepare a slide.
4. Acquire skills in using low and high-power magnification.
5. Estimate sizes of specimens.
6. Prepare stained, wet-mount slides.
7. Observe structure of plant cells.
8. Observe structure of animal cells.
9. Contrast and compare the structure of plant and animal cells using your microscope examinations and labeled diagrams.

MATERIALS

Microscope, glass slide, cover slips, lens paper, slide with a small clear metric ruler, pieces of newspaper, forceps, onion bulbs, Lugol's iodine solution, paper towels, flat-edged toothpicks, scalpel, scissors

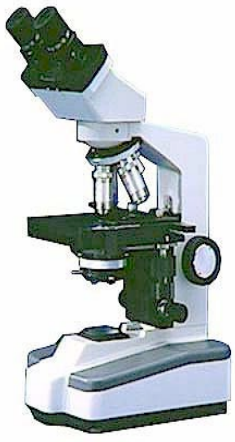
PROCEDURE

Last lab you received a "LAB BENCH NUMBER." This number corresponds to your seat at your lab bench. It is also the number on your microscope and other materials that you will use during the term. You are responsible for items with this number. When instructed, go to the microscope cabinet and grasp your microscope with two hands. Bring it back to your bench and wait for further instructions.

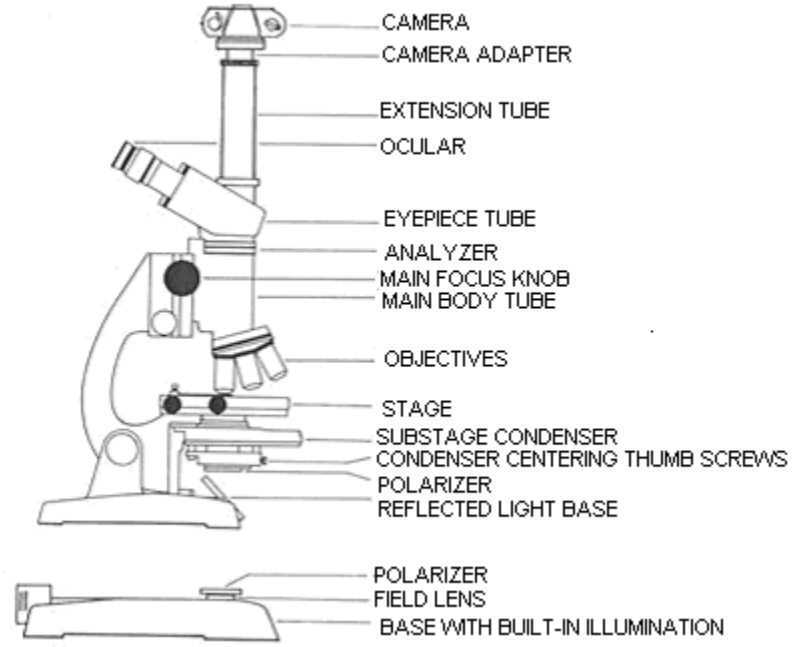
I. Parts of the Microscope

1. As your teacher reviews the parts of the microscope and their uses, you should label the diagram of the microscope.

The following are common compound microscopes in the Biology Department:



You can find the names of the parts of the microscope from the diagram below:



2. The following rules for the proper use of the microscope should be remembered:
   a. Always use two hands when carrying a microscope. One hand should be grasping the arm of the microscope while the other hand should be under the base. Always keep the base parallel to the floor.
   b. Place the microscope on the desk with the arm away from you.
   c. **ONLY use lens paper** to clean the lenses and the mirror before use.

II. Using the microscope: Note that our scopes have 10x oculars, and 4x, 10x, and 40x objectives.

1. Setting up the microscope.

Remember that **ocular x objective is total magnification**. Oculars on most of our scopes are 10x.

   a. Revolve the **scanning objective (4X)** until it is centered over the hole in the stage. You should feel a click when it is in place.
   b. Turn on the light (some microscopes may have a concave mirror) and open the diaphragm to produce a bright circle of light. This circle is your field of vision (FOV).
   c. After you have prepared the slide to be viewed you will continue with the following.

2. Focusing the microscope.
   a. Place the slide on the carrier on the stage between the prong and the spring clip so that it nestles against the inside corner of the carrier. DO NOT put the slide under the metal carrier. Using the knobs at the side of the stage, adjust the carrier so that the slide is centered over the hole in the stage.
   b. Using the coarse adjustment knob, bring the stage as close as possible to the objective. For most of our microscopes, the stages rises to meet the objective).
   c. To focus, always start with the coarse adjustment. Focus by moving the stage down slowly. If you have moved the focus a long way and nothing has appeared, you may have to move the stage slowly up until it is a little less than 1 cm from the objective. Move the stage down again until you find the material on your slide. Remember to center your specimen using the knobs at the side of the stage.
   d. To examine the specimen under **low power objective (10X),** gently swivel the objective into place. Focus using the **coarse adjustment knob first, and then the fine adjustment knob.** Move the slide around to get the best view in the center of the FOV.
   e. To examine the specimen under the **high power objective (40X)** gently swivel the objective into place. ONLY FOCUS WITH THE FINE ADJUSTMENT KNOB.
   f. When finished, swivel the nosepiece into the “neutral” position (no objective is in place) and carefully remove the slide from the stage.

Rules for a Biological Drawing

The ability to draw accurately and neatly is a useful skill, especially in science. Most artists follow some basic rules to make their drawings attractive and easy to read. Below are some rules which will improve your drawing skill.

1. Use unlined paper.
2. Draw in pencil.
3. Always print.
4. Leave at least a one-inch margin on all four sides of the paper.
5. Center the title and print it in capital letters.
6. Center the drawing on the page.
7. Never cross lines.
8. Never use the plural form of a word when pointing to a single object or part.
9. Do little or no erasing.
10. When using the scientific name of an organism in places other than the title, remember that the genus or first part of a scientific name is always capitalized. The species or second part of a scientific name is not.
    FOR EXAMPLE: Canis familiaris is the genus and species name for a dog. In a title it would be CANIS FAMILIARIS
11. Print your name and other information as specified by your teacher.

DATA TABLE CHECKLIST

Data tables should be constructed on lined or gridded paper, using ink and a STRAIGHT EDGE.

Title the table in a way that relates the independent variable to the dependent variable. For example: The Effect of Fertilizer Concentration (the independent variable) on Plant Growth (the dependent variable).

Column headings include the dependent and independent variables. They may also include trial or setup numbers or other information.

Column headings need to indicate units of measurement.

The independent variable is typically recorded in increasing order.

The dependent variable is recorded to correspond with the independent variable.

Height of Students in Class 5A1

Student	Gender	Height (cm)
Joe	M	150
Sarah	F	154
Patrick	M	152
John	M	145

PROTOZOA
vacuole
nucleus
cell membrane
Amoeba proteus

Rules for a Biological Drawing

Below is a biological drawing with several errors. Circle all the errors you can find and make a correction next to each error.

Protozoa
nucleus
grooves
gullet
food vacuole

Paramecium Caudatum
Stony Brook
2nd period
Sept 5, 1986
PR. NEDWIDEK

Review the rules for a biological drawing. The next time you do a biological drawing be sure to observe the rules you have learned.

ALWAYS BRING SCOPE, DISSECTION LABS SHEET TO

Stuyvesant High School Name:

Biology Department Section: Teacher: Dr. Nedwidek

Laboratory Exercise #

Title:

Before you proceed, review the rules for a drawing shown above.

III. Examining the letter "e"

1. Preparation and examination of the LOWER CASE letter "e" slide.
   a. Prepare slide for observation by placing a piece of newspaper with a word containing the letter "e" on a slide (the rectangular piece of glass) in an upright position. Cover it slowly with a coverslip (the small plastic square). (Your teacher may require you to use a drop of water on the specimen to make a WET MOUNT.)
   b. Put the slide in the slide holder and move it so that letter e is over the hole on the stage. Keep the "e" in the upright position.
   c. Follow the instructions in procedure II for focusing under first the **scanning objective (4X)** and then the **low-power objective (10X). Remember to center your letter "e" in the FOV.**
2. Observation of the letter "e".
   a. Under the low-power objective, note the orientation of your letter "e". Move the slide gently to the right and then left, noting the direction in which the letter moves. Move the slide away from you and towards you, noting the direction in which the letter moves.
   b. On your answer paper, draw a circle about 5 cm in diameter to represent the circle of light (field of vision) that you observe in the microscope. In this circle, Observe the way the ink clings to the paper and the texture of the paper. Try to capture these features in your report response.
   c. Switch to the **high power objective (40X)** & think about what has happened to the image. In a circle, **DRAW & LABEL the letter "e" under high power (400x)**. Capture its texture & structure.

IV. Measurement of the size of the field.: TOTAL MAG: 40x (scanning), 100x (low), 400x (high).

1. Switch the nosepiece into the neutral position.
2. Remove the slide of the letter "e".
3. Place a slide of a clear metric ruler onto the stage and adjust it over the light. Carefully switch to the **low-power objective.** Focus with the coarse adjustment knob. Position the ruler's bottom edge along the diameter so you can see at least one mm mark. **ESTIMATE** the size, in micrometers (μm), of the field of vision and record your answer on the answer page.
4. Magnification has an **INVERSE** relationship to the diameter of the field of vision. Determine the actual size, in micrometers (μm), of the scanning, low and high-power fields of vision (FOV) for your answer page. Does the FOV size increase or deacrease with an increase in mag? Answer in your report.
5. In a second circle, **DRAW AND LABEL the "e" with ruler laying over it under low power (100x).**

V. Preparation of the Plant Cell (onion) slide

1. Place several drops of Lugol's solution (iodine) on a clean slide.
2. With the forceps, remove the thin epidermal tissue from the inside of an onion scale. You need a piece approximately 1 cm square. You may need to trim it with your scalpel. **Your instructor will demonstrate this technique**.
3. Place the tissue in the drop of Lugol's solution so that the outer face is up. The surface tension of the fluid will pull the tissue down.
4. If the tissue is wrinkled, use the needle and the forceps to gently flatten it.
5. Place coverslip on slide and be sure the liquid comes to its edges. Use a small piece of paper towel to remove the excess water that may come out from under the coverslip. Remove any air bubbles by pressing gently down on the cover slip with the tip of a toothpick or pencil.

A. Be certain that your microscope is under the scanning objective (4X). Focus. Then switch to the low power (10X) objective. Make sure the field has the appropriate amount of light.
   a. Observe the plant epidermal cells at 40x then 100x.
   b. Look around the edges of the cell and you will find a fine granular substance that has been lightly stained. This is the cytoplasm. In living cells, the cytoplasm will circulate in a process called **cyclosis**. Note whether **cyclosis** is occurring in your onion cells (need 400x for this).
   c. Your instructor will review the recommended method of drawing and labeling your drawings. Always indicate the actual **MAGNIFICATION** the cell drawing represented. Your teacher will remind you what to label for your sketch.

B. Switch to the high power (40X) objective.
   a. Observe the cells under high power, 400x.

b. Carefully using your fine adjustment carefully observe your cell. Try to find the chloroplasts and the nucleus which will be more obvious to you here.
c. In a third circle, **DRAW AND LABEL one onion cell in your field of vision at 400x.** Use the tip of your pencil to capture the texture of the cell components. Do not remove specimen from stage of microscope until you have completed the drawing.

VI. Preparation of the Animal Cell (Stuyvesant Student) slide

1. Place several drops of Lugol's iodine solution on a clean slide.
2. Using the broad, flat end of a clean, newly obtained toothpick, gently scrape the inside of your cheek for epithelial cells.
3. Stir the scrapings into the Lugols solution gently. If the cells are bunched together, carefully spread those apart by tapping the tooth pick gently. Throw the tooth pick into the wastebasket.
4. Place a coverslip on the slide and examine it under the scanning objective, 40x magnification.

a. Switch to low-power and visualize your cheek cells at 100x magnification. The animal cells are much smaller than the plant cells. They will appear more clearly and with greater contrast if you reduce the amount of light in your field of vision.

b. Switch to high power and in a fourth circle, **DRAW AND LABEL two of your cheek epithelial cells at 400x magnification.**

VII. Complete all summary questions as you go along with the 4 drawings, and **hand in the lab on the date assigned.**