Name: _____

AP Biology

Microscope Skills Review

Objectives

After completing this worksheet, you will be able to

- determine the magnifying power of a microscope.
- determine the size of the field of view of a microscope.
- estimate the size of the specimen you are viewing in micrometers (μm).
- prepare a wet mount slide to microscopically examine plant and animal cells.

Magnifying Power

The microscope is one of the most useful tools in a biology laboratory. In a high school lab, the most common microscope used is the compound light microscope (LM). It contains a combination of lenses that focus and bend light, producing an enlarged (or magnified) image of a specimen. A compound microscope has two sets of lenses. The lens you look through is called the ocular or eyepiece. The lens near the specimen being examined is called the objective.

The objective lens is one of three or four lenses located on a rotating turret above the stage, and they vary in magnifying power. The lowest power is called the low power objective (LP), and the highest power is the high power objective (HP). You can determine the magnifying power of the combination of the two lenses by multiplying the magnifying power of the ocular by the magnifying power of the objective that you are using. For example, if the magnifying power of the ocular is 10X and the magnifying power of an objective is 4X, the magnifying power of that lens combination is 40X. If an object is magnified 40 times, the image you see is 40 times larger than the object would appear if viewed with the unaided eye at a distance of about 25 cm.

Questions:

1. What is the primary difference between a low-power objective and a high-power objective?

2. What is the total magnification of a microscope with a 15X ocular and a 40X objective?

Field of View

The **field of view** is the maximum area visible through the lenses of a microscope, and it is represented by a diameter. To determine the diameter of your field of view, place a transparent metric ruler under the low power (LP) objective of a microscope. Focus the microscope on the scale of the ruler, and measure the diameter of the field of vision in millimeters. Record this number, you will need it later. _____

When you are viewing an object under high power, it is sometimes not possible to determine the field of view directly. The higher the power of magnification, the smaller the field of view. The diameter of the field of view under high power must be calculated using the following equation.

$$\frac{\text{diameter (LP)} \times \text{magnification of LP objective}}{\text{magnification of HP objective}} = \text{diameter (HP)}$$

For example, if you determine that your field of view is 2.5 mm in diameter using a 10X ocular and 4X objective, you will be able to determine what the field of view will be with the high-power objective by using the above formula. For this example, we will designate the high-power objective as 40X.

$$\frac{2.5 \text{ mm} \times (4X)}{(40X)} = .25 \text{ mm} = 250 \text{ µm}$$

Question:

3. A student determines that the field of view with a 10X ocular and a 4X objective is 2.1 mm in diameter. What is the diameter of the field of view with the same ocular and a 40X objective?

Estimating the Size of the Specimen Under Observation

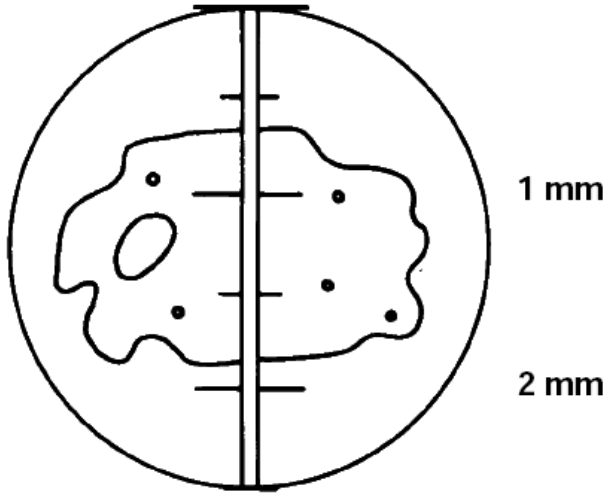
Objects observed with microscopes are often too small to be measured conveniently in millimeters. Because you are using a scale in millimeters, it is necessary to convert your measurement to micrometers. Remember that 1 µm = 0.001 mm & 1000 µm = 1 mm.

To estimate the size of an object seen with a microscope, first estimate what fraction of the diameter of the field of vision that the object occupies. Then multiply the diameter you calculated in micrometers by that fraction. For example, if the field of vision's diameter is 400 µm and the object's estimated length is about one-tenth of that diameter, multiply the diameter by one-tenth to find the object's length.

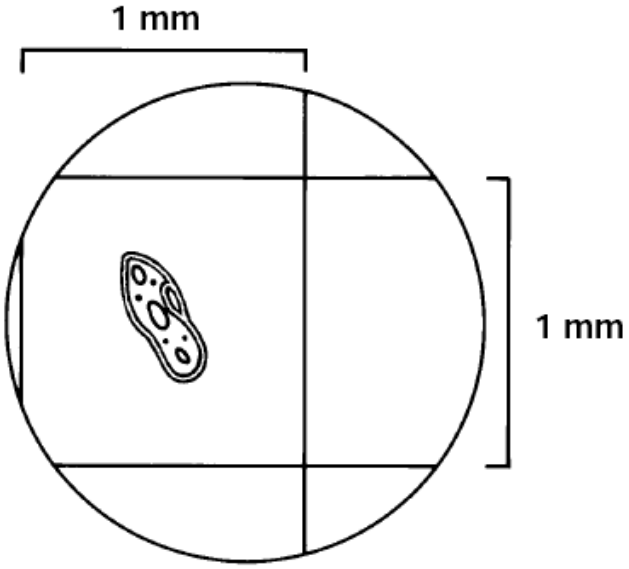
$$400 \text{ µm} \times \frac{1}{10} = 40 \text{ µm}$$

Questions:

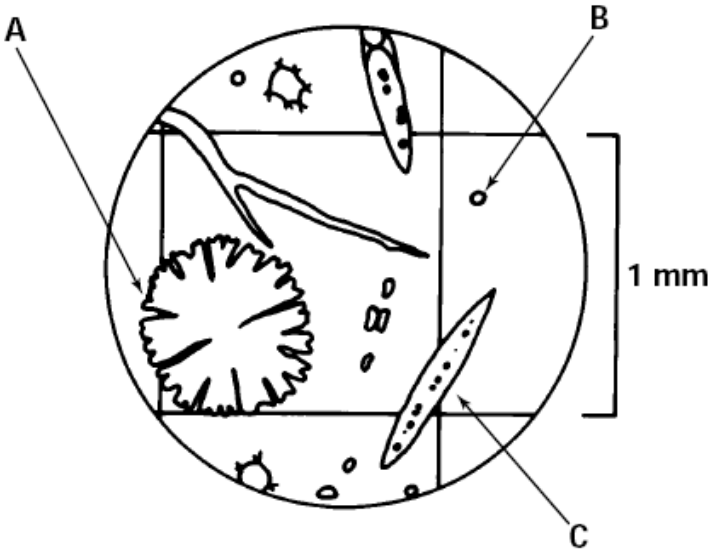
4. If the diameter of the field of view under a microscope is 2.5 mm, what are the approximate dimensions (length x width) of the amoeba in the illustration below? Use the millimeter scale provided to help you answer the question. Express your answer in micrometers (µm).



5. Estimate the length of the organism in the field-of-view illustration below. Express your answer in micrometers (μm).



6. The illustration below is a sample view of the organisms you might see in a drop of lake water, using a 10X ocular and 10X objective. Three of these organisms are indicated by A, B, and C. Using the space below the diagram, describe each organism as completely as you can, including its shape and dimensions, the magnifications used, and the diameter of your field of view. Give all dimensions in micrometers (μm).



Specimen	Shapes and Dimensions (μm)	Magnification	Diameter of Field of View (μm)
A			
B			
C			

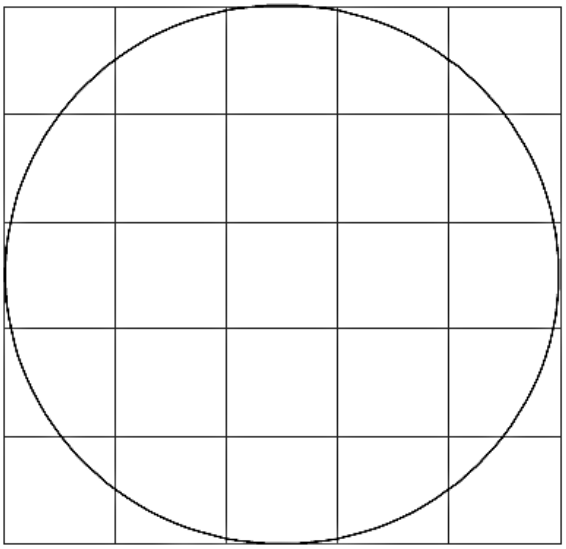
Procedure for Examining Aquatic Plant Cells

1. Take a microscope from the storage area and place it about 10 cm from the edge of the desk.
2. Carefully clean the ocular and objective lenses with lens paper.
3. Place a drop of water in the center of a clean glass slide.
4. With forceps, remove a leaf from the *Elodea* plant and place it on the drop of water on the slide. Make sure that the leaf is flat. If it is folded, straighten it with the forceps.
5. Carefully place a coverslip over the drop of water and the *Elodea* leaf.
6. Place the slide on the stage of the microscope with the leaf directly over the opening in the stage.
7. Using the low-power objective, locate the leaf under the microscope. Turn the coarse adjustment knob (the bigger one) until the leaf comes into focus.
8. Switch to the high-power objective. **CAUTION:** *When turning the high-power objective, you should always look at the objective from the side of your microscope so that the objective does not hit or damage the slide.*
9. Observe the cells of the *Elodea* leaf under high power. In the circle below, draw and label a detailed picture of what you see. Write a brief description of the colors, shapes, dimensions, and cell structures you notice in each of the *Elodea* cells. Do not forget to record the magnification of the microscope and calculate the diameter of the field of view (see explanation above).

Magnification: _____ X

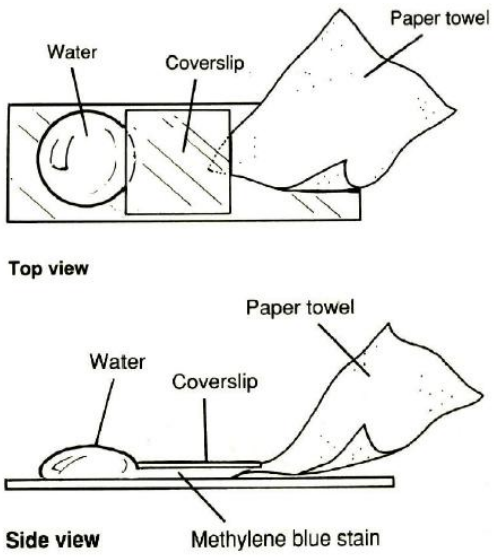
Diameter of Field of View (µm): _____

Description of Cells:



Procedure for Examining Human Cheek Cells

1. Carefully clean and dry your slide and coverslip using the lens paper.
2. Place a drop of water in the center of your glass slide.
3. Using the flat end of a toothpick, gently scrape the inside of your cheek. **CAUTION:** *Do not use force when scraping the inside of your cheek. Only a few cells are needed.* The end of the toothpick will have several cheek cells stuck to it even though you may see nothing but a drop of saliva.
4. Stir the water on the slide with the end of the toothpick to mix the cheek cells with the water. Dispose of the toothpick.
5. Put one drop of methylene blue stain on top of the drop of water containing cheek cells. **CAUTION:** *Use care when working with methylene blue to avoid staining hands and clothing.*
6. Wait one minute, and then carefully place a coverslip over the stained cheek cells.
7. To remove the stain from under the coverslip and replace it with clear water, place a piece of paper towel at the edge of one side of the coverslip. Then place a drop of water at the edge of the coverslip of the opposite side. The stained water under the coverslip will be absorbed by the paper towel. As the stain is removed, the clear water next to the coverslip on the opposite side will be drawn under the coverslip. Discard the paper towel after it has absorbed the stained water. See diagram on the right for help.

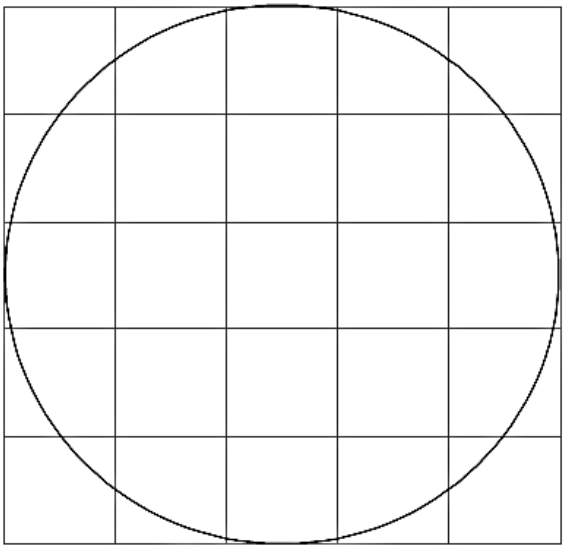


8. Place the slide on the stage of the microscope with the center of the coverslip directly over the opening in the stage.
9. Using the low-power objective, locate a few cheek cells under the microscope. **NOTE:** *You will need to reduce the amount of light coming through the slide in order to see the cells more clearly. Adjust the diaphragm or the light intensity dial as necessary.*
10. Switch to the high-power objective and observe some cheek cells. In the circle below, draw and label a detailed picture of what you see. Write a brief description of the colors, shapes, dimensions and other patterns you notice in each of the cheek cells. Do not forget to record the magnification of the microscope and calculate the diameter of the field of view.

Magnification: _____ X

Diameter of Field of View (µm): _____

Description of Cells:



Summary Questions:

1. When do you use the coarse focus adjustment knob on a microscope?

2. Suppose you are looking at protists under the microscope and cannot see anything on low power. What adjustment could you make to the microscope that might help you see the protists, without switching to a higher magnification?

3. Suppose you focused on an organism using medium power, but then cannot see the organism after switching to high power. What should you do?

4. When putting together a wet-mount slide, why is it important to make sure that there are no air bubbles under the cover slip? What should you do if there are air bubbles?