

Using a Compound Light Microscope

Introduction

Many objects are too small to be seen by the eye alone. They can be seen, however, with the use of an instrument that magnifies, or visually enlarges, the object. One such instrument, which is of great importance to biologists and other scientists, is the compound light microscope. A compound light microscope consists of a light source or mirror that illuminates the object to be observed, an objective lens that magnifies the image of the object, and an eyepiece (ocular lens) that further magnifies the image of the object and projects it into the viewer's eye.

Objects, or specimens, to be observed under a microscope are generally prepared in one of two ways. Prepared or permanent slides are made to last a long time. They are usually purchased from biological supply houses. Temporary or wet-mount slides are made to last only a short time—usually one laboratory period.

The microscope is an expensive precision instrument that requires special care and handling. In this investigation, you will learn the parts of a compound light microscope, the functions of those parts, and the proper use and care of the microscope. You will also learn the technique of preparing wet-mount slides.

Problem

What is the proper use of a compound light microscope?

Pre-Lab Discussion

Read the entire investigation. Then, work with a partner to answer the following questions.

1. Why might it be a good idea to keep your microscope at least 10 cm from the edge of the table?

2. Why should a microscope slide and coverslip be held by their edges?

3. Why do scientists use microscopes?

4. Why should you use lens paper only once?

5. Why is it important to eliminate air bubbles from the slide?

Materials (per group)

compound light microscope
prepared slide
lens paper
soft cloth (or cheesecloth)
newspaper

microscope slide
coverslip
dissecting probe
dropper pipette
scissors

Safety

Put on a laboratory apron. Always handle the microscope with extreme care. You are responsible for its proper care and use. Use caution when handling microscope slides, as they can break easily and cut you. Never use direct sunlight as a light source for a compound light microscope. The sunlight reflecting through the microscope could damage your eye. Be careful when handling sharp instruments. Observe proper laboratory procedures when using electrical equipment. Note all safety alert symbols next to the steps in the Procedure and review the meanings of each symbol by referring to Safety Symbols on page 8.

Procedure

Part A. Care of the Compound Light Microscope

- Figure 1 shows the proper way to carry a microscope. Always carry the microscope with both hands. Grasp the arm of the microscope with one hand and place your other hand under the base. Always hold the microscope in an upright position so that the eyepiece cannot fall out. Place a microscope on your worktable or desk at least 10 cm from the edge. Position the microscope with the arm facing you.
- Notice the numbers etched on the objectives and on the eyepiece. Each number is followed by an "X" that means "times." For example, the low-power objective may have the number "10X" on its side, as shown in Figure 2. That objective magnifies an object 10 times its normal size. Record the magnifications of your microscope in the Data Table. The total magnification of a microscope is calculated by multiplying the magnification of the objective by the magnification of the eyepiece. For example:

magnification of objective	X	magnification of eyepiece	=	total magnification
10X	X	10X	=	100X

Use the formula to complete the Data Table.

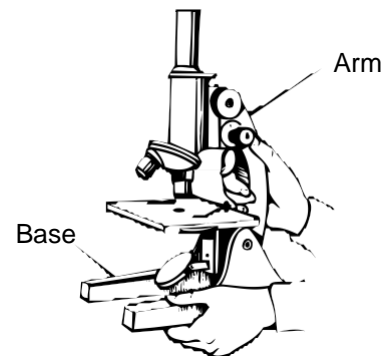


Figure 1

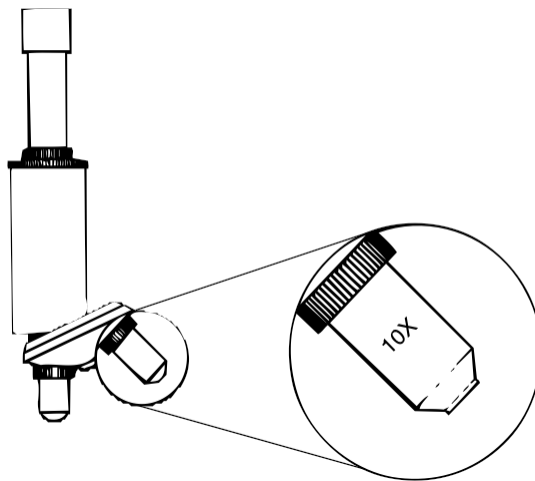


Figure 2

Data Table

Objective	Magnification of Objective	Magnification of Eyepiece	Total Magnification
Low power			
High power			
Other			

- Before you use the microscope, clean the lenses of the objectives and eyepiece with lens paper. **Note:** *To avoid scratching the lenses, never clean or wipe them with anything other than lens paper. Use a new piece of lens paper on each lens you clean. Never touch a lens with your finger. The oils on your skin may attract dust or lint that could scratch the lens.*

Part B. Use of a Compound Light Microscope

- Look at the microscope from the side. Locate the coarse adjustment knob that moves the objectives up and down. Practice moving the coarse adjustment knob to see how it moves the objectives with each turn.
- Turn the coarse adjustment so that the low-power objective is positioned about 3 cm from the stage. Locate the revolving nosepiece. Turn the nosepiece until you hear the high-power objective click into position. See Figure 3. When an objective clicks into position, it is in the proper alignment for light to pass from the light source through the objective into the viewer's eye. Now turn the nosepiece until the low-power objective clicks back into position. **Note:** *Always look at the microscope from the side when moving an objective so that the microscope does not hit or damage the slide.*

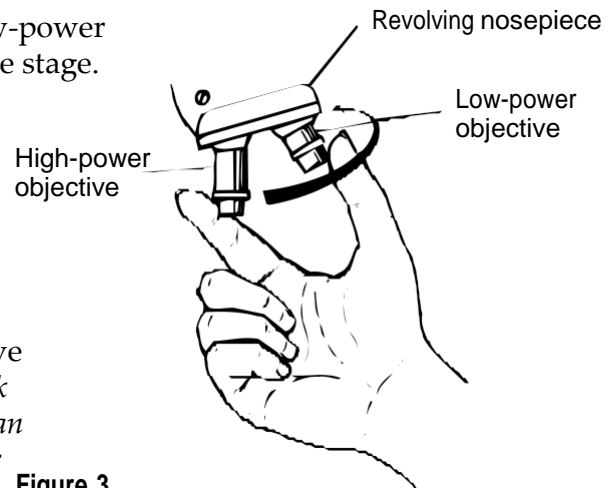
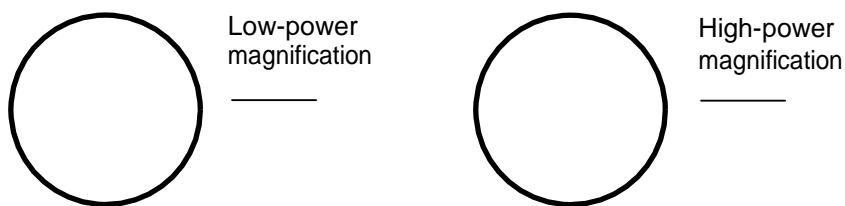


Figure 3

3. If your microscope has an electric light source, plug in the cord and turn on the light. If your microscope has a mirror, turn the mirror toward a light source such as a desk lamp or window. **CAUTION:** *Never use the sun as a direct source of light.* Look through the eyepiece. Adjust the diaphragm to permit sufficient light to enter the microscope. The white circle of light you see is the field of view. If your microscope has a mirror, move the mirror until the field of view is evenly illuminated.
4. Place a prepared slide on the stage so that it is centered over the stage opening. Use the stage clips to hold the slide in position. Turn the low-power objective into place. Look at the microscope from the side and turn the coarse adjustment so that the low-power objective is as close as possible to the stage without touching it.
5. Look through the eyepiece and turn the coarse adjustment to move the low-power objective away from the stage until the object comes into focus. To avoid eyestrain, keep both eyes open while looking through a microscope. **CAUTION:** *To avoid moving the objective into the slide, never lower the objective toward the stage while looking through the eyepiece.*
6. Turn the fine adjustment to bring the object into sharp focus. You may wish to adjust the diaphragm so that you can see the object more clearly. In the appropriate space below, draw what you see through the microscope. Record the magnification.
7. Look at the microscope from the side and rotate the nosepiece until the high-power objective clicks into position. Look through the eyepiece. Turn the fine adjustment to bring the object on the slide into focus. **CAUTION:** *Never use the coarse adjustment when focusing the high-power objective lens. This could break your slide or damage the lens.* In the appropriate space below, draw what you see through the microscope. Record the magnification.



8. Remove the slide. Move the low-power objective into position.

Part C. Preparing a Wet Mount

1. Use a pair of scissors to cut a letter "e" from a piece of newspaper. Cut out the smallest letter "e" you can find. Position the "e" on the center of a clean glass slide.
2. Use a dropper pipette to place one drop of water on the cut piece of newspaper. See Figure 4B.

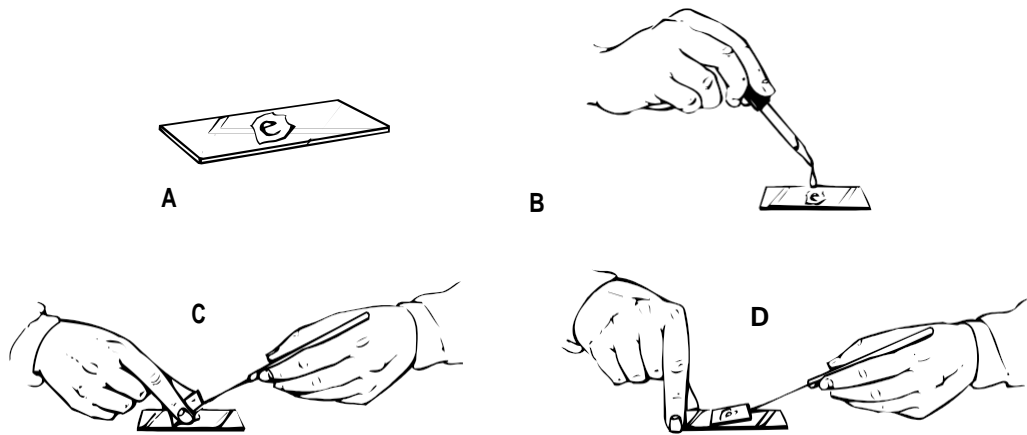
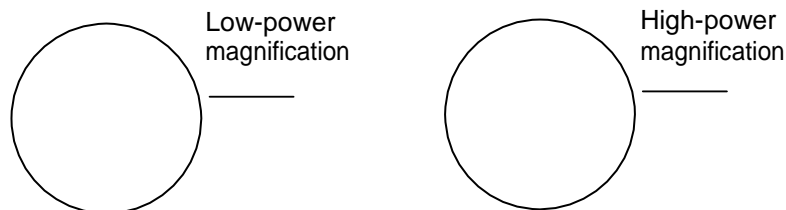


Figure 4



3. Hold a clean coverslip in your fingers as shown in Figure 4C. Make sure the bottom edge of the coverslip is in the drop of water. Use a dissecting probe to slowly lower the coverslip onto the wet newspaper. Slowly lowering the coverslip prevents air bubbles from being trapped between the slide and the coverslip. The type of slide you have just made is called a wet mount. Practice making a wet mount until you can do so without trapping air bubbles on the slide.
4. Center the wet mount on the stage with the letter “e” in its normal upright position. **Note:** *Make sure the bottom of the slide is dry before you place it on the stage.* Turn the low-power objective into position and bring the “e” into focus. In the appropriate place below, draw the letter “e” as seen through the microscope. Record the magnification.
5. While looking through the eyepiece, move the slide to the left. Notice the way the letter seems to move. Now move the slide to the right. Again notice the way the letter seems to move. Move the slide up and down and observe the direction the letter moves.
6. Turn the high-power objective into position and bring the letter “e” into focus. In the appropriate place below, draw the letter “e” as seen through the microscope. Record the magnification.



7. Take apart the wet mount. Clean the slide and coverslip with soap and water. Carefully dry the slide and coverslip with paper towels and return them to their boxes.
8. Rotate the low-power objective into position and use the coarse adjustment to place it as close to the stage as possible without touching. Carefully pick up the microscope and return it to its storage area.

Analysis and Conclusions

1. **Inferring** Why do you place one hand under the base of the microscope as you carry it?

2. **Observing** How is the image of an object seen through the high-power objective different from the image seen through the low-power objective?

3. **Observing** How does the letter “e” as seen through the microscope differ from the way an “e” normally appears?

4. **Inferring** Explain why a specimen to be viewed under the microscope must be thin.

5. **Inferring** Why should you never use coarse adjustment when focusing the high-power objective lens?

6. **Drawing Conclusions** Images under a microscope are reversed and inverted. Suppose you were observing an organism through the microscope and noticed that it moved toward the bottom of the slide and then it moved to the right. What does this tell you about the actual movement of the organism?

7. Describe the changes in the field of view and the amount of available light when going from low to high power on a compound light microscope.
