The human eye cannot distinguish objects much smaller than 0.1 mm. The microscope is a tool that extends vision and allows observation of much smaller objects. The most commonly used compound microscope (Figure PC.1) is monocular (one eyepiece). Light reaches the eye after passing through the objects to be examined.

In this investigation, you will learn how to use and care for a microscope.

Materials (per team of 2)
3 coverslips
dropping pipet
3 microscope slides
compound microscope
newspaper
scissors
transparent metric ruler
prepared slide of colored threads

Procedure

PART A Care of the Microscope

1. The microscope is a precision instrument that requires proper care. Always carry the microscope with both hands—one hand under its base, the other on its arm.
2. When setting the microscope on a table, keep it away from the edge. If a lamp is attached to the microscope, keep its wire out of the way. Keep everything not needed for microscope studies off your lab table.
3. Avoid tilting the microscope when using temporary slides made with water.
4. The lenses of the microscope cost almost as much as all the other parts put together. Never

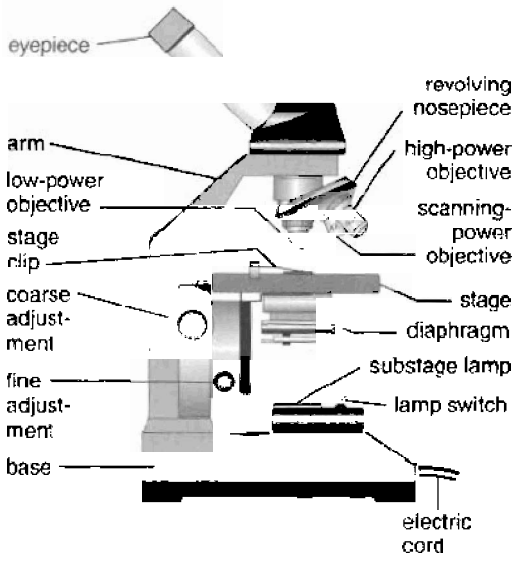

FIGURE PC.1

Parts of a compound microscope.

clean lenses with anything other than the lens paper designed for this task.

5. Before putting away the microscope, *always* return it to the low-power setting. The high-power objective reaches too near the stage to be left in place safely.

PART B Setting Up the Microscope

6. Rotate the low-power objective into place if it is not already there. When you change from one objective to another, you will hear a click as the objective sets into position.
7. Move the mirror so that even illumination is obtained through the opening in the stage, or turn on the substage lamp. Most microscopes are equipped with a diaphragm for regulating light. Some materials are best viewed in dim light; others, in bright light. Remember that direct sunlight can damage eyes. If you use natural light as your light source, do not reflect direct sunlight through the diaphragm.
8. Make sure that the lenses are dry and free of fingerprints and debris. Wipe lenses with lens paper only.

TABLE PC.1
Microscopic Observations

Object being viewed	Observations and comments
Letter *o*	
Letter *c*	
Etc.	
Millimeter ruler	

PART C Using the Microscope

9. In your logbook, prepare a table like Table PC.1
10. Cut a lowercase *o* from a piece of newspaper. Place it right side up on a clean slide. With a dropping pipet, place one drop of water on the letter. This type of slide is called a wet mount.

CAUTION: Scissors are sharp. Handle with care.

11. Wait until the paper is soaked before adding a coverslip. Hold the coverslip at about a 45° angle to the slide, and slowly lower it. Figure PC.2 shows these first steps.

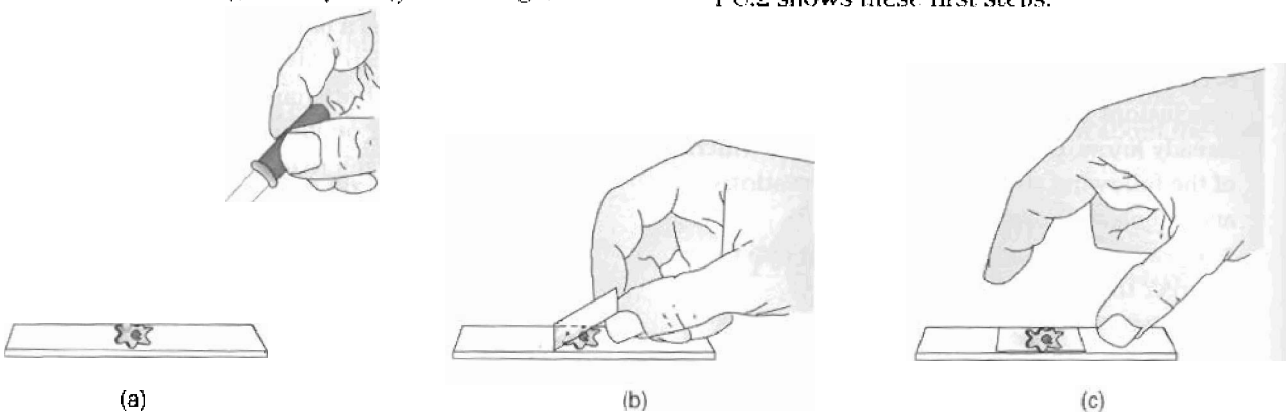

FIGURE PC.2

Preparing a wet mount with a microscope slide and coverslip.

12. Place the slide on the microscope stage, and clamp it down. Move the slide so the letter is in the middle of the hole in the stage. Use the coarse-adjustment knob to lower the low-power objective to the lowest position.
13. Look through the eyepiece, and use the coarse-adjustment knob to raise the objective slowly until the letter *o* is in view. Use the fine-adjustment knob to sharpen the focus. Position the diaphragm for the best light. Compare the way the letter looks through the microscope with the way it looks to the naked eye.
14. To determine how greatly magnified the view is, multiply the number inscribed on the eyepiece by the number on the objective being used. For example, eyepiece (10×) × objective (10×) = total magnification (100×).
15. Follow the same procedure with a lowercase *e*. In your logbook, describe how the letter looks when viewed through a microscope.
16. Make a wet mount of the letter *e* or the letter *r*. Describe how the letter looks when viewed through the microscope. What new information (not revealed by the letter *c*) is revealed by the *e* or *r*?
17. Look through the eyepiece at the letter as you use your thumbs and forefingers to move the slide *away* from you. Which way does your view of the letter move? Move the slide to the right. In which direction does the image move?
18. Make a pencil sketch of the letter as you see it under the microscope. Label the changes in image and movement that occur under the microscope.
19. Make a wet mount of two different-colored hairs, one light and one dark. Cross one hair over the other. Position the slide so that the hairs cross in the center of the field. Sketch the hairs under low power; then go to Part D.

PART D Using High Power

20. With the crossed hairs centered under low power, adjust the diaphragm for the best light.
21. Turn the high-power objective into viewing position. Do *not* change the focus.
22. Sharpen the focus with the *fine-adjustment knob only. Do not focus under high power with the course-adjustment knob.*
23. Readjust the diaphragm to get the best light. If you are not successful in finding the object under high power the first time, return to step 20 and repeat the whole procedure carefully.
24. Using the fine-adjustment knob, focus on the hairs at the point where they cross. Can you see both hairs sharply at the same focus level? How can you use the fine-adjustment knob to determine which hair crosses over the other? Sketch the hairs under high power.
25. Remove the wet mount of the hairs, and replace it with the prepared slide of the colored threads. The prepared slide contains three colored threads that overlap in a specific order.
26. Focus the threads under low power, and adjust the diaphragm for best light.
27. Turn the high-power objective into viewing position. Do *not* change the focus.
28. Sharpen the focus with the fine-adjustment knob only.
29. Readjust the diaphragm to get the best light. If you are not successful in finding the threads under high power, return to step 26 and repeat the procedure.
30. Using the fine-adjustment knob, focus on an area where the threads overlap. Use the fine-adjustment knob to determine the order in which the colored threads lie on the slide.

PART E Measuring with a Microscope

31. Because objects examined with a microscope usually are small, biologists use units of length smaller than centimeters or millimeters for microscopic measurement. One such unit is the micrometer, which is 1/1,000 of a millimeter. The symbol for micrometer is μm, the Greek letter μ (called mu) followed by m.
32. You can estimate the size of a microscopic object by comparing it with the size of the circular field of view. To determine the size of the field, place a plastic metric ruler on the stage. Use the low-power objective to obtain a clear image of the divisions on the ruler. Carefully move the ruler until its marked edge passes through the exact center of the field of view. Count the number of divisions that you can see in the field of view. The marks on the

ruler will appear quite wide; 1 mm is the distance from the center of one mark to the center of the next. Record the diameter, in millimeters, of the low-power field of your microscope.

33. Remove the plastic ruler, and replace it with the wet mount of the letter *e*. (If the wet mount has dried, lift the coverslip and add water.) Using low power, compare the height of the letter with the diameter of the field of view. Estimate as accurately as possible the actual height of the letter in millimeters.

Analysis

1. Summarize the differences between an image viewed through a microscope and the same image viewed with the naked eye.
2. When viewing an object through the high-power objective, not all of the object may be in focus. Explain your answer.
3. What was the order of the overlapping colored threads in step 30?
4. What is the relationship between magnification and the diameter of the field of view?
5. What is the diameter in micrometers of the low-power field of view of your microscope?
6. Calculate the diameter in micrometers of your high-power field. Use the following equations:

$$\frac{\text{magnification number of high-power objective}}{\text{magnification number of low-power objective}} = A$$

$$\frac{\text{diameter of low-power field of view}}{A} = \text{diameter of high-power field of view}$$

For example, if the magnification of your low-power objective is 12× and that of your high-power objective is 48×, $A = 4$. If the diameter of the low-power field of view is 1,600 μm, the diameter of the high-power field of view is 1,600 μm ÷ 4, or 400 μm.

7. Use your sketch of the hairs under high power and the diameter of your high-power field calculated above to estimate the diameter of your human hair.