

Background Information

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?

Materials (per group)

Compound light microscope	Glass slide
Prepared glass slide	Coverslip
Lens paper	Dissecting needle
Soft cloth (or cheesecloth)	Medicine dropper
Newspaper	Scissors

Procedure**Part A. Introduction to the Light Microscope (LM)**

1. 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. Take a microscope and place it on your worktable or desk at least 10 cm from the edge. Position the microscope with the arm facing you.

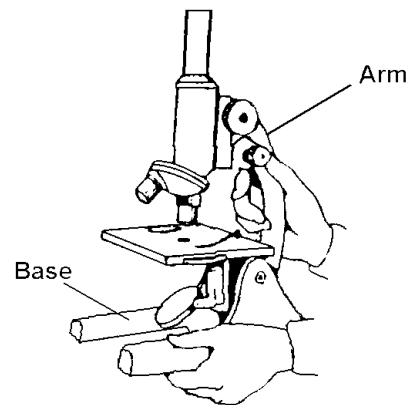
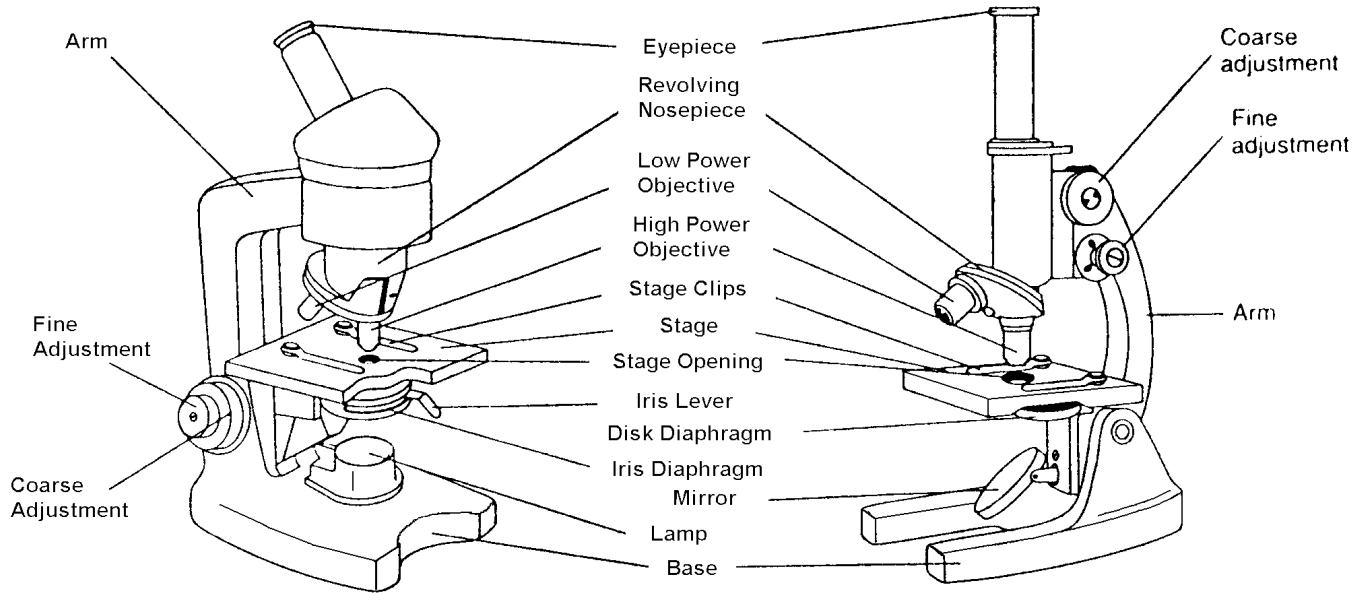


Figure 1

- Study the labeled drawings of the microscopes in Figure 2. Identify the following parts on your microscope: eyepiece, arm, coarse adjustment, fine adjustment, revolving nosepiece, low power objective, high power objective, stage, stage clips, stage opening, diaphragm, light source (mirror or lamp), and base. Review the function of each part of the microscope in Figure 2. Do not proceed with this investigation until you can identify all the parts of your microscope and describe the function of each part. Note: Tell your teacher at once if you find any parts of the microscope missing or damaged.

Figure 2 Parts of the Microscope



- 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 3. 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. Use the following formula to complete the Data Table in Observations.

Magnification of Low Power Objective	x	Magnification of Eyepiece	=	Total Magnification
10X	x	10X	=	100X

- 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.
- Use a soft cloth or a piece of cheesecloth to wipe the stage and the mirror or light.

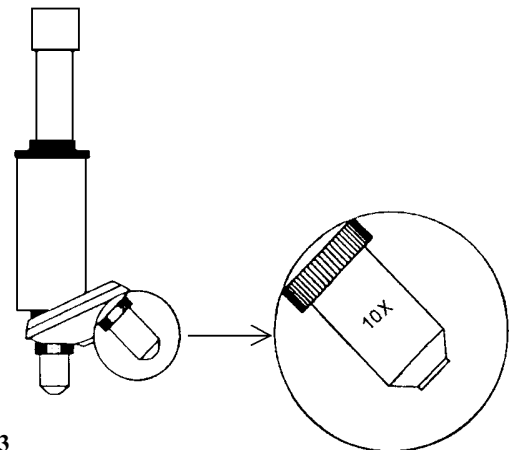
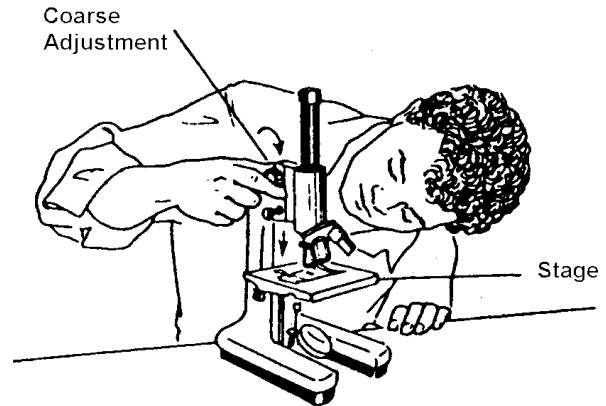


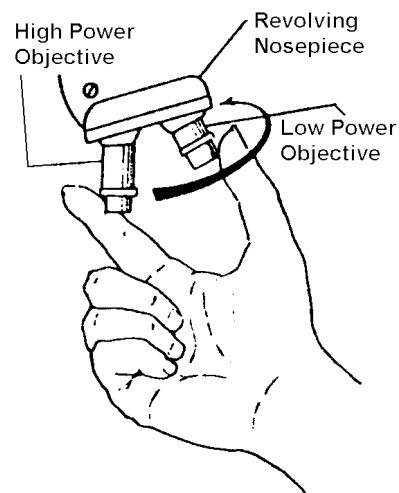
Figure 3

Part B. Using a Compound Microscope

1. Look at the microscope from the side as shown in Figure 4. Locate the coarse adjustment knob which moves the objectives up and down. Practice moving the coarse adjustment knob to see how it moves its objectives with each turn.



2. 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 5. 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.



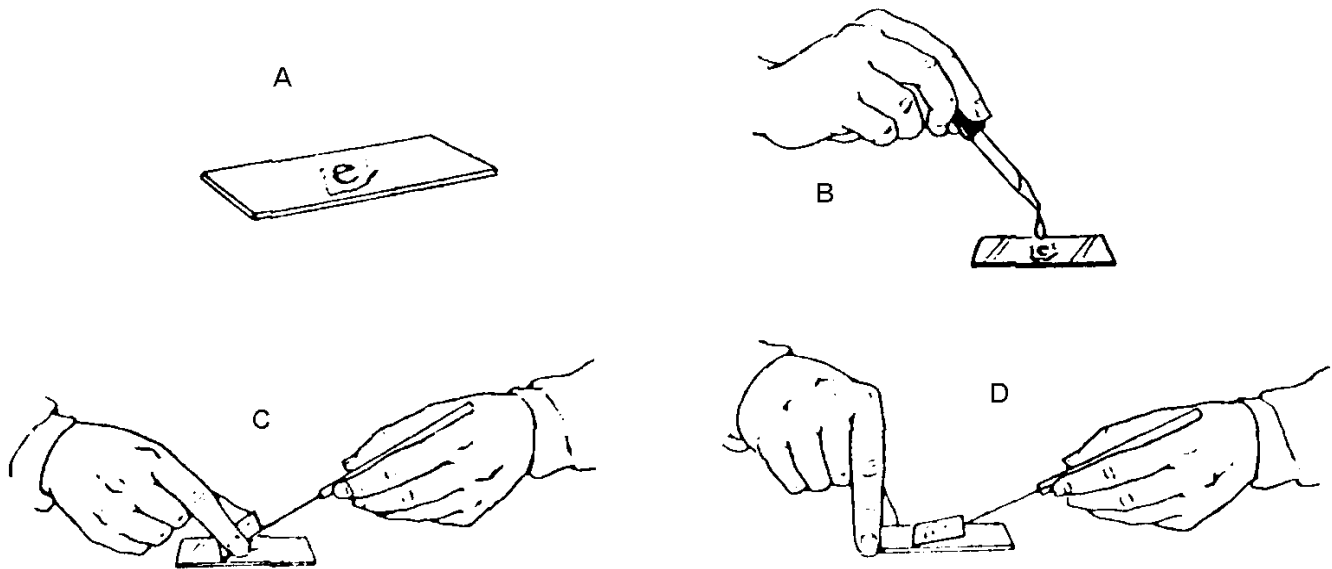
3. If your microscope has an electric light source, plug in the cord and turn on the light. If your microscope has a mirror, turn the mirror toward a light source such as a desk lamp or window. **CAUTION:** Never use the sun as a direct source of light. Look through the eyepiece. Adjust the diaphragm 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 towards the stage while looking through the eyepiece.
6. Turn the fine adjustment to bring the object into sharp focus. You may wish to move the diaphragm to adjust the amount of light so that you can see the object more clearly. In the appropriate place in Observations, draw what you see through the microscope. Note the magnification

- 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 place in Observations, draw what you see through the microscope. Note the magnification.
- When the drawing is finished, move the low power objective into position. Now you may remove the slide and return it to its storage tray.

Part C. Preparing a Wet Mount Slide

- Select a word from a newspaper article which has a small case letter “e” in it. Using a scissors, cut out the word and position it on the center of a microscope slide. Place the word on the slide so that you can read it in a normal fashion - right side up.
- Place a clean slide on a paper towel. Use a medicine dropper to add a few drops of water on the piece of newspaper. See Figure 6.

Figure 6

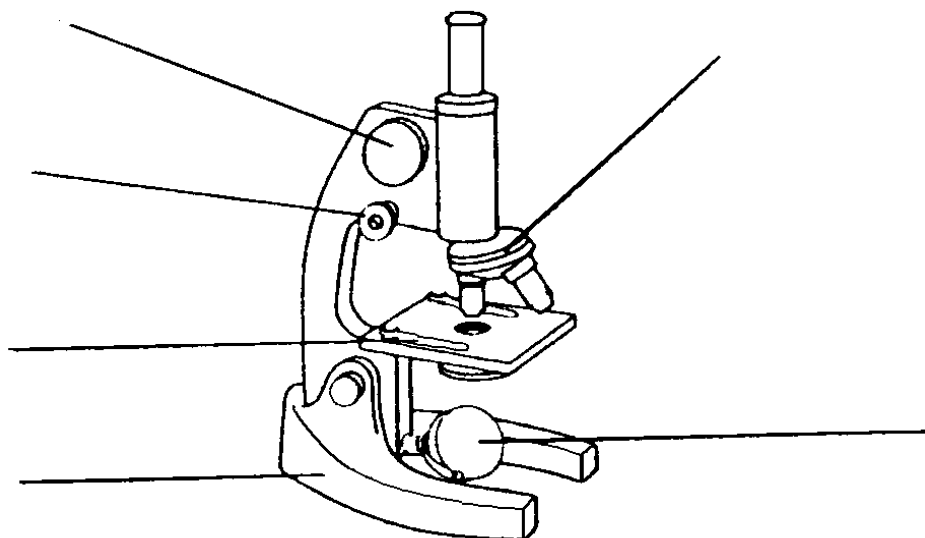


- Hold a clean coverslip in your fingers as shown in Figure 6. Make sure the bottom edge of the coverslip is in the drop of water. Use a dissecting needle to slowly lower the coverslip onto the wet newspaper. Slowly lowering the coverslip prevents air bubbles from being trapped between the slide and the coverslip. The type of slide you have just made is called a wet mount slide. Practice the technique of making a wet mount slide until you can do so without trapping air bubbles on the slide.

- Center the slide on the stage with the “word” in its **normal upright position**. **Note:** Make sure the bottom of the slide is dry before you place it on the stage. Use the scanning power objective and bring the letter “e” of the word into focus. In the appropriate place in Observations, draw the letter “e” as seen through the microscope. Note the magnification.
- Turn the low power objective into position and bring the letter “e” into focus. In the appropriate place in Observations, draw the letter “e” as seen through the microscope. Note the magnification.
- Rotate the nosepiece back to the low power objective and bring the letter “e” into focus again. While looking through the eyepiece, move the slide to left. Notice the way the letters seems to move. Now move the slide to the right. Again notice the way the letters seem to move. Move the slide up and observe the direction the letters move. Move the slide down and observe the direction the letters move.
- Rotate the nosepiece back to the scanning power objective and carefully remove the slide. Turn off the light source and your teacher will give you instructions about putting the microscope away in a proper manner.
- Take apart the wet mount slide. Clean the slide and coverslip with water. Carefully dry the slide and coverslip with paper towels and return them to the proper area.

Observations

- Label the parts of the microscope shown below.

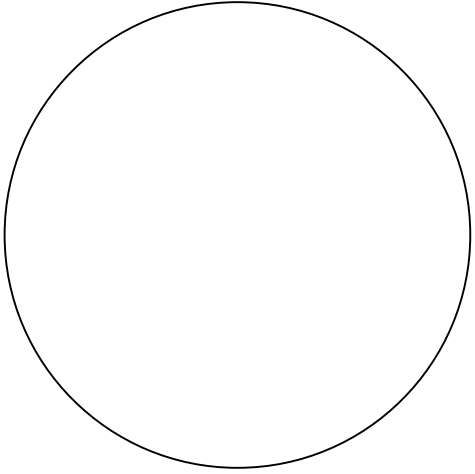
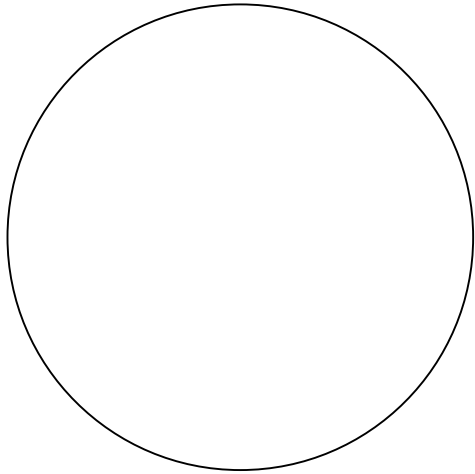


- Fill in the magnification of each objective and the eyepiece of your microscope. To determine the total magnification, multiply the magnification of each objective by the magnification of the eyepiece.

Data Table

Objective	Magnification of Objective	Magnification of Eyepiece	Total Magnification
Scanning Power			
Low Power			
High Power			
Oil Immersion			

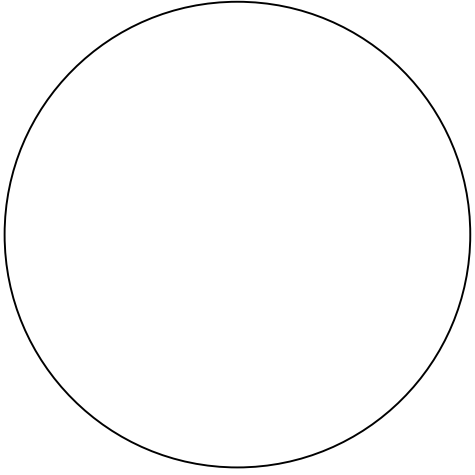
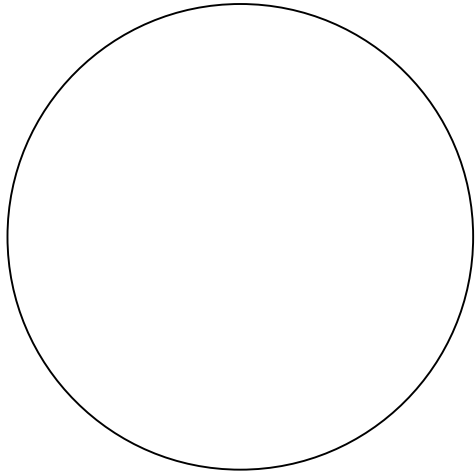
3. **Optional activity.** Make a drawing of the object on your prepared slide as seen under low power and high power.



Low Power Magnification _____

High Power Magnification _____

4. Make a detailed drawing of the letter “e” as seen under scanning and low power.



Scanning Power Magnification _____

Low Power Magnification _____

Analysis and Conclusions

1. Why do you place one hand under the base of the microscope as you carry it?
2. Does your microscope have a mirror or lamp for illumination?
3. How is the image of an object seen through the high power objective different from the image seen through the low power objective?

4. How does the letter “e” as seen through the microscope differ from the way an “e” normally appears?
5. When you move the slide to the left, in what direction does the image appear to move?
6. When you move the slide up, in what direction does the image appear to move?

Critical Thinking and Application

1. Explain why a specimen to be viewed under the microscope must be thin.
2. Why should a glass slide and a coverslip be held by their edges?
3. Why should you use a piece of lens paper only once?
4. 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?
5. List the steps for putting away your light microscope at the end of the period.
 - a)
 - b)
 - c)
 - d)
 - e)
6. If possible, you should always focus on the edge of the _____ when you first begin.
7. Complete the table below.

Type of Microscope	Power	Disadvantage
	2000	Limited ability to magnify objects.
	100,000	Cannot view living specimens.
	200,000	Cannot view living specimens.