

## Lab: Using a Compound Light Microscope

### Background:

Microscopes are very important tools in biology. The term *microscope* can be translated as "to view the tiny," because microscopes are used to study things that are too small to be easily observed by other methods. The type of microscope that we will be using in this lab is a compound light microscope. Light microscopes magnify the image of the specimen using light and lenses. The term *compound* means that this microscope passes light through the specimen and then through two different lenses. The lens closest to the specimen is called the *objective* lens, while the lens nearest to the user's eye is called the *ocular* lens or *eyepiece*.

When you use a compound light microscope, the specimen being studied is placed on a glass slide. The slide may be either a *prepared* slide that is permanent and was purchased from a science supply company, or it may be a *wet mount* that is made for temporary use and is made in the lab room.

### Objectives: In this lab you will:

1. Learn the parts of a compound light microscope and their functions.
2. Learn how to calculate the magnification of a compound light microscope.
3. Learn how to make a wet mount slide.
4. Understand how the orientation and movement of the specimen's image changes when viewed through a compound light microscope.
5. Learn the proper use of the low and high power objective lenses.
6. Learn the proper use of the coarse and fine adjustments for focusing.

### Materials:

microscope	water
slides	pipette
cover slips	scissors
lens paper	magazine picture with various colors
paper towels	hairs of different color
sheets of newspaper/phonebook	other objects for viewing

### Procedure:

#### Part I. Learning about the microscope

1. One member of your lab group should go and get a microscope. Always carry the microscope in an upright position (not tilted) using two hands. One hand should hold the microscope's arm and the other hand should support the base, as shown in **Figure 1**. Set it down away from the edge of the table. *Always remember that a microscope is an expensive, precision instrument that should be handled carefully.*
2. Plug the microscope in at your lab desk. Turn it on and make sure that the light comes on (it may take a second or two to warm up). If the microscope light does not turn on, check with your teacher.
3. Compare your microscope with **Figure 2** on the next page. Identify the parts on your microscope and determine the function of each part.

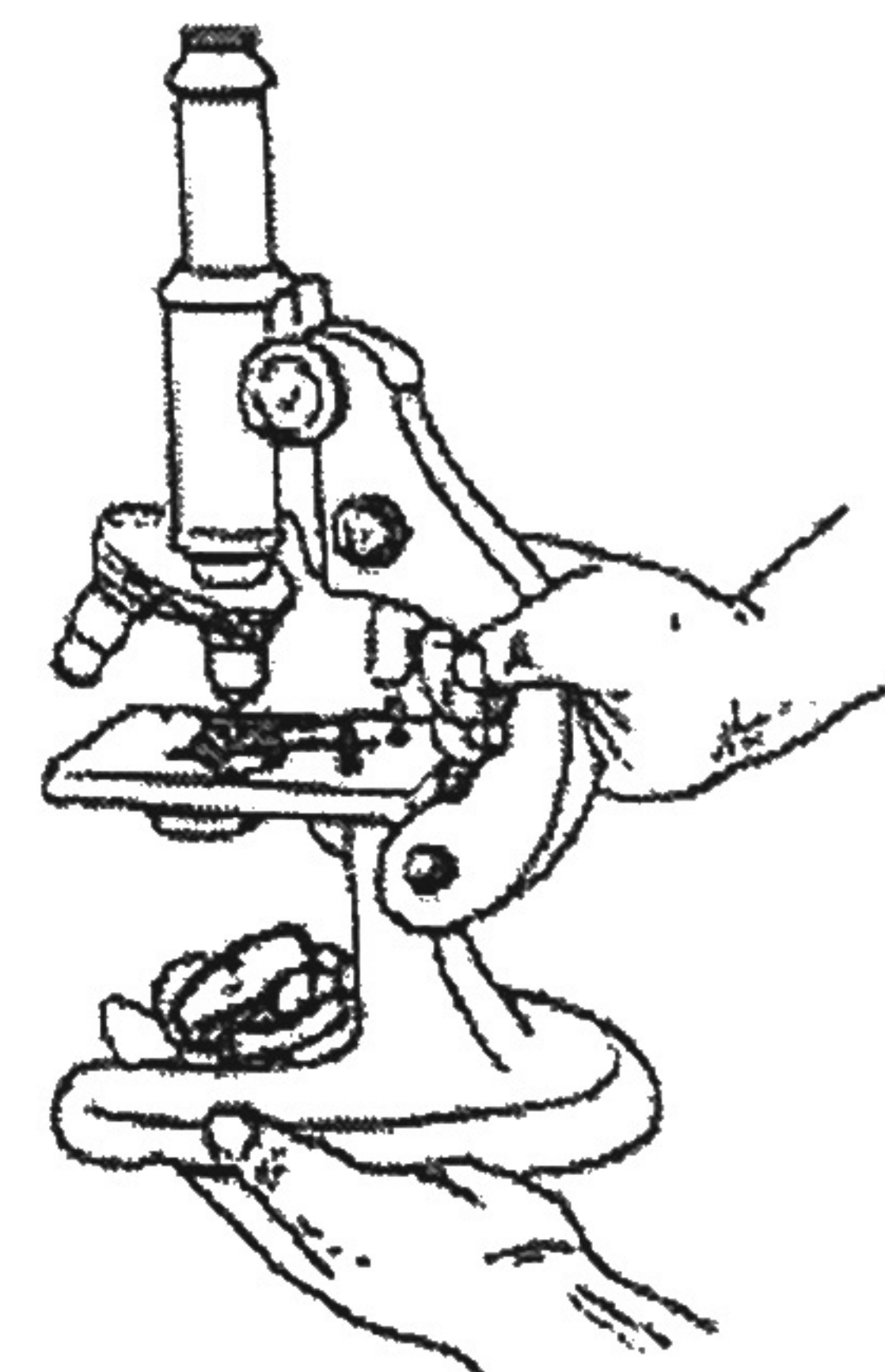
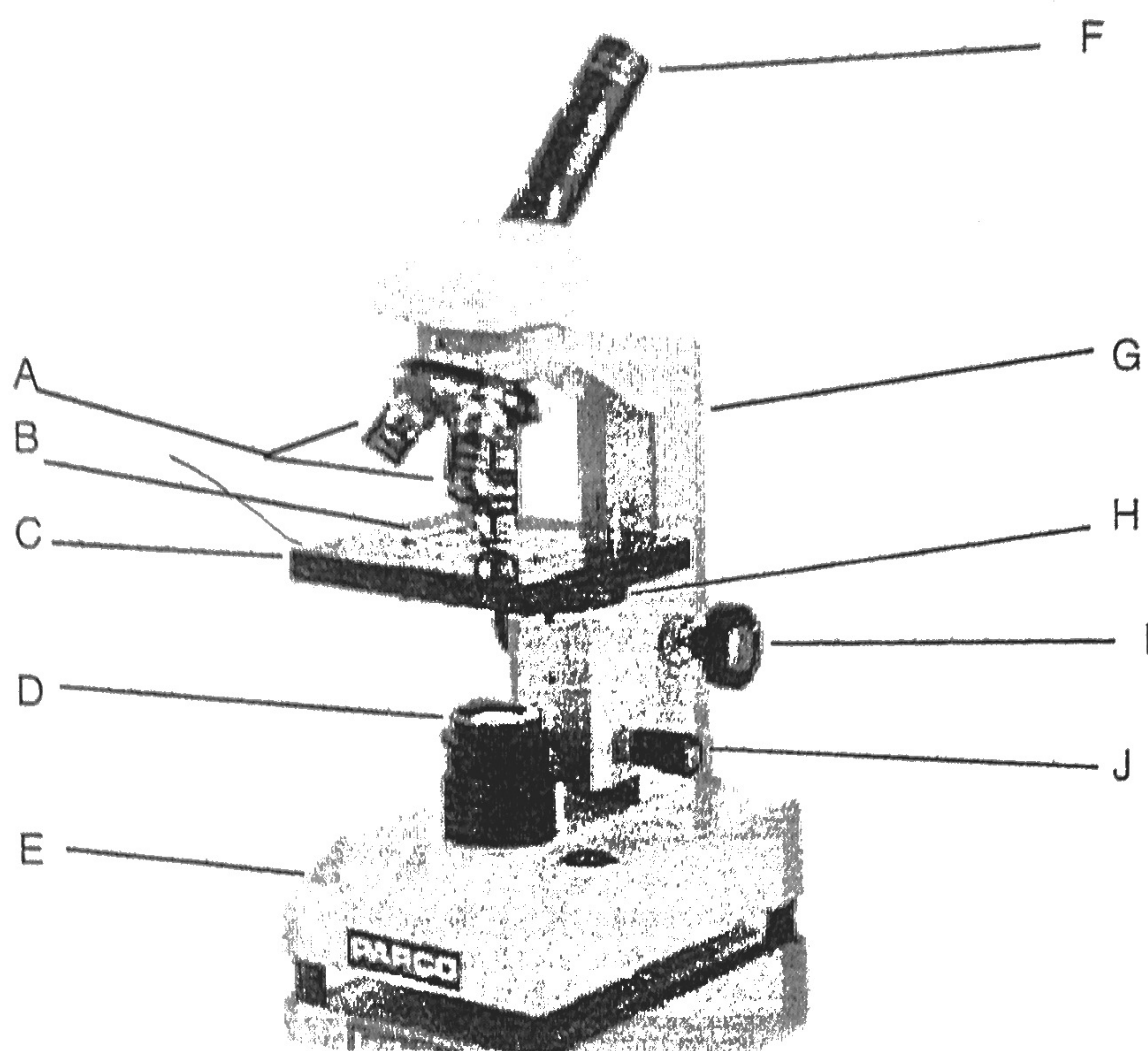


Figure 1



Use the words from the word bank below to fill in the parts names in Table 1 below.

**Word Bank:**

- Arm
- Base
- Coarse adjustment
- Diaphragm
- Fine adjustment
- Objective lenses
- Ocular lens
- Light
- Stage
- Stage clips

**Figure 2**

4. Fill in **Table 1** with the names and functions of each microscope part in **Figure 2**.

**TABLE 1:**

Part	Name	Function
A		
B		
C		
D		
E		
F		
G		
H		
I		
J		

5. The ocular lens is marked with its magnification power. (This is how much larger the lens makes objects appear.)
- What is the magnification power of the ocular lens of your microscope?
6. The three objective lenses are marked with their magnification power. The first number marked on each lens is the magnification power of that lens.
- What is the magnification of the lowest power lens of your microscope?
  - What is the magnification of the high power lens?
7. To find the total magnification of your microscope as you are using it, multiply the ocular lens power times the power of the objective lens that you are using. For example, if the ocular lens of a microscope has a power of 5x and you use an objective that is 10x, then the total magnification of the microscope at that time is 50x ( $5 \times 10 = 50$ ).
- What is the total magnification of your microscope when using low power?
  - What is the total magnification of your microscope when using high power?

## Part II. Preparing and using a Wet Mount

8. Using a piece of newspaper or phone book, find a small, lowercase letter "e." Cut a 1 cm square with that letter "e" near the middle of the square. (Do **not** just cut out the letter e, or it will be too hard to work with. The piece of paper that you cut out should be about the size of a fingernail.)

9. Place the square of paper in the middle of a clean glass slide. Position the square so that the words are in normal reading position (in other words, don't have the "e" turned sideways or upside-down). With a pipette, put 1 drop of water on the paper square. Drop the water from about 1 cm above the slide; do not touch the pipette to the paper square or the paper will stick to the pipette.

10. Now, cover the water drop with a clean cover slip. The best way to do this is shown in **Figure 3**. Hold the cover slip at a 45° angle to the slide and move it over the drop. As the water touches the cover slip, it will start to spread. Gently lower the angle of the cover slip to allow the water to evenly coat the under surface, then let the slip drop into place.

You should not just drop the cover slip onto the slide or air bubbles will get trapped. This makes the slide very difficult to study. If you do trap several air bubbles, remove the slip and try again. **Never** press on the cover slip to try to remove air bubbles. This will break the cover slip and/or damage your specimen.

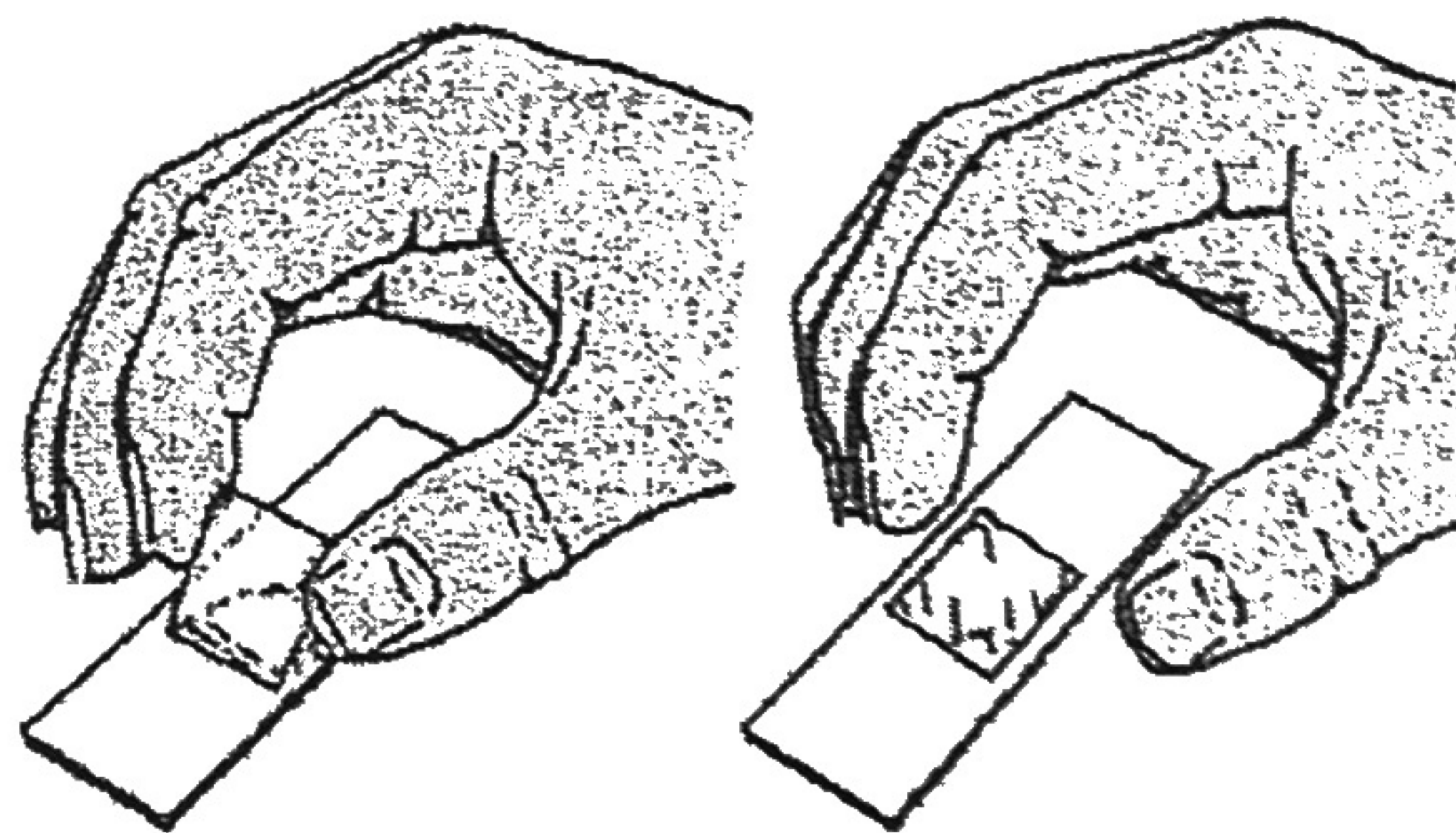
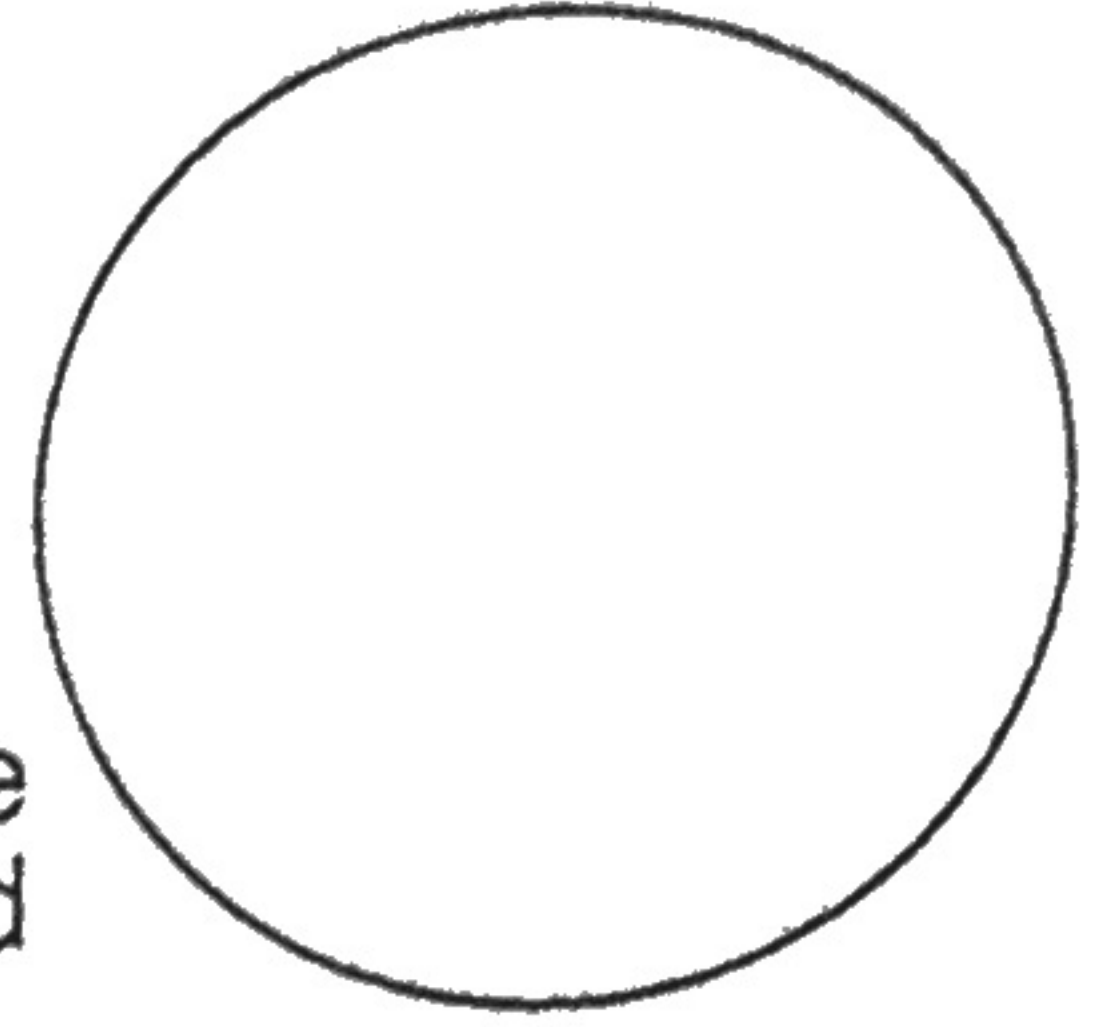


Figure 3

11. On your microscope, move the low-power objective into place. You should always begin studying a slide on low power, because this makes it easiest to find objects on the slide. Position the diaphragm so that the largest opening is used. This will allow the maximum amount of light to be used. Check your wet mount slide to be sure that the bottom of the slide is dry. (A wet slide will stick on the stage of the microscope.) Sit so that the arm of the microscope is closest to you, and place the slide on the stage with the "e" in a normal reading position for you.

12. You may use the stage clips to hold the slide in place if you like. If so, make sure that the clips do not bump into the cover slip or touch the water. Look at the microscope from the side and use the coarse adjustment knob to get the stage as close to the low-power objective as possible.

13. Look through the ocular lens, keeping both eyes open. (It may seem difficult to keep both eyes open, but learning to do so helps to prevent eyestrain or headaches.) Slowly adjust the focus of the microscope using the coarse adjustment knob until the letters become clear. Then, use the fine adjustment to sharpen the focus. Move the slide left or right, forward or backward, until the letter "e" is in the center of the field of view. Do **not** turn the slide or pivot it. In the circle to the right, draw the letter "e" in the same size and position as you see it in the microscope.



f. Describe the position of the image of the letter "e" through the microscope compared to the position that it is placed on the slide.

14. Move the slide to the left.  
g. Which direction did the image move?

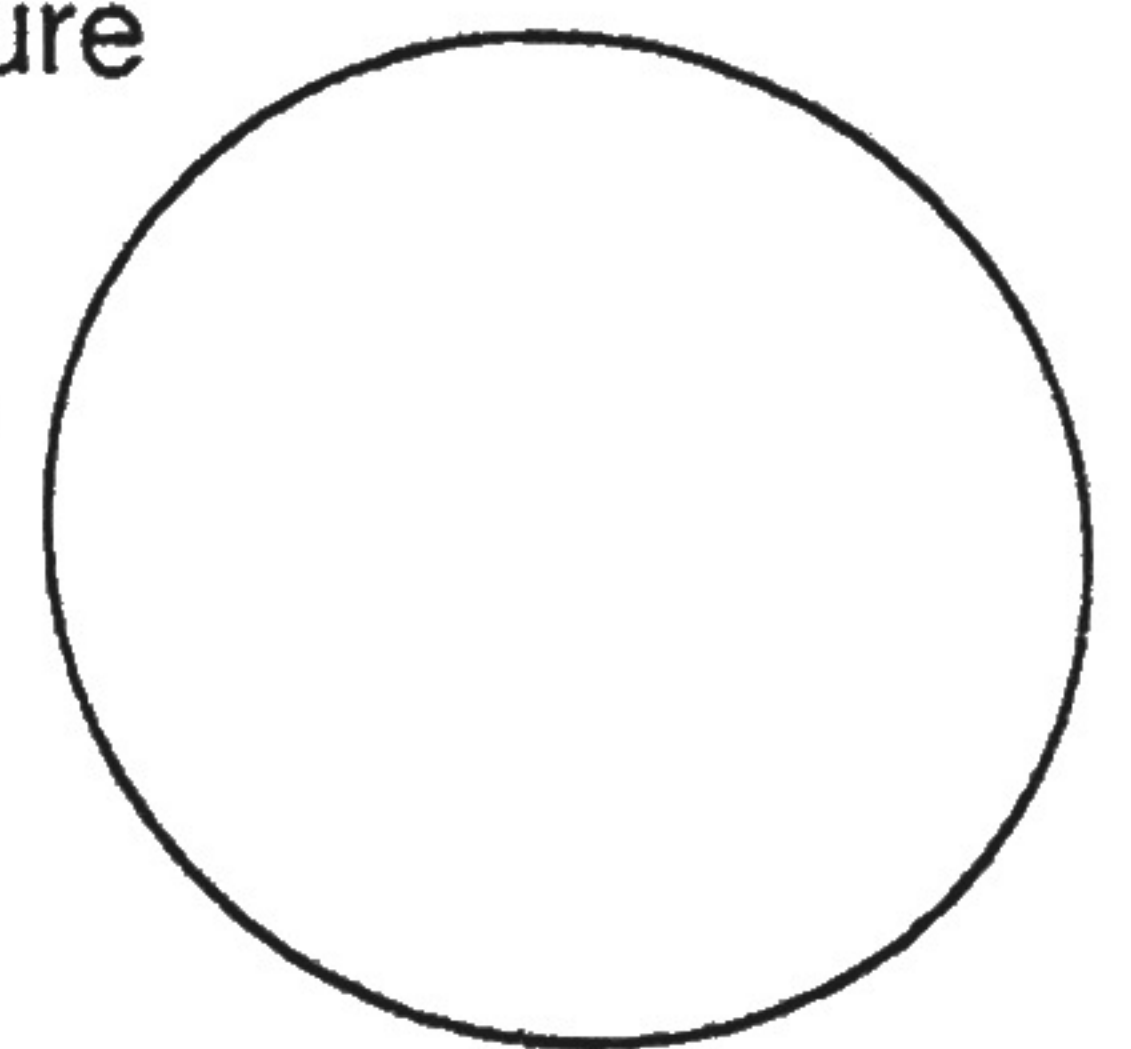
15. Move the slide away from you.  
h. Which way did the image move?

16. Look through the microscope as you change the adjustment of the diaphragm.  
i. What does the diaphragm control?

**Important Note:** Before switching to high power, you should **always** position the specimen in the center of the field of view and use the fine adjust to sharpen the focus of the image. **Never** use the coarse adjustment when using high power. Doing so could break the slide or the microscope!

17. Watching from the side, switch to the high-power objective lens. Make sure that the lens does not hit the slide, but expect it to be very close.

18. Looking through the ocular, use a slight turn of the fine adjustment knob to focus the image of the letter "e". In the circle to the right, draw the letter "e" in the same size and position as you see it in the microscope.



j. Describe the appearance of the image that you see. Are you seeing more or less of the letter "e" than you did at low-power?

k. Is the field of view (the area that you are observing) larger or smaller when you use high power?

19. Look through the microscope (on high power) with the diaphragm at its largest setting. While looking through the ocular, switch the microscope to low-power.
- l. Compare the brightness of the field under high power and low power. Which setting is brighter?

### Part III. Other Observations

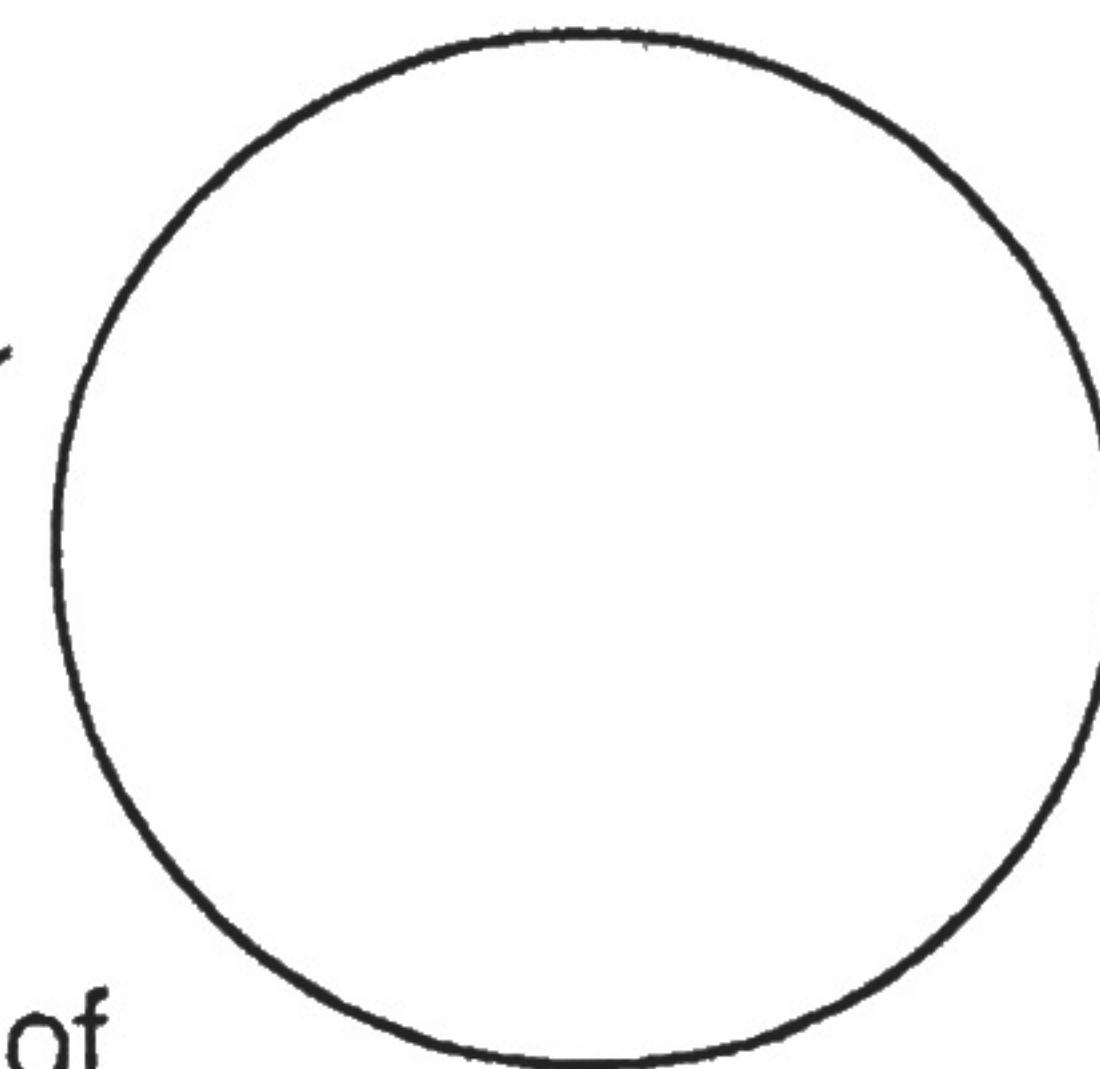
20. Select a picture from a magazine that has several bright colors. Cut out a 1 cm square from the picture that has a variety of colors. Clean off your slide from Part II and make a new wet mount with the magazine picture.

21. Observe the magazine picture, starting on low power and scanning the image. Then switch to high power and observe the colors.

- m. Record the colors seen without the microscope:

Record the colors seen with the microscope:

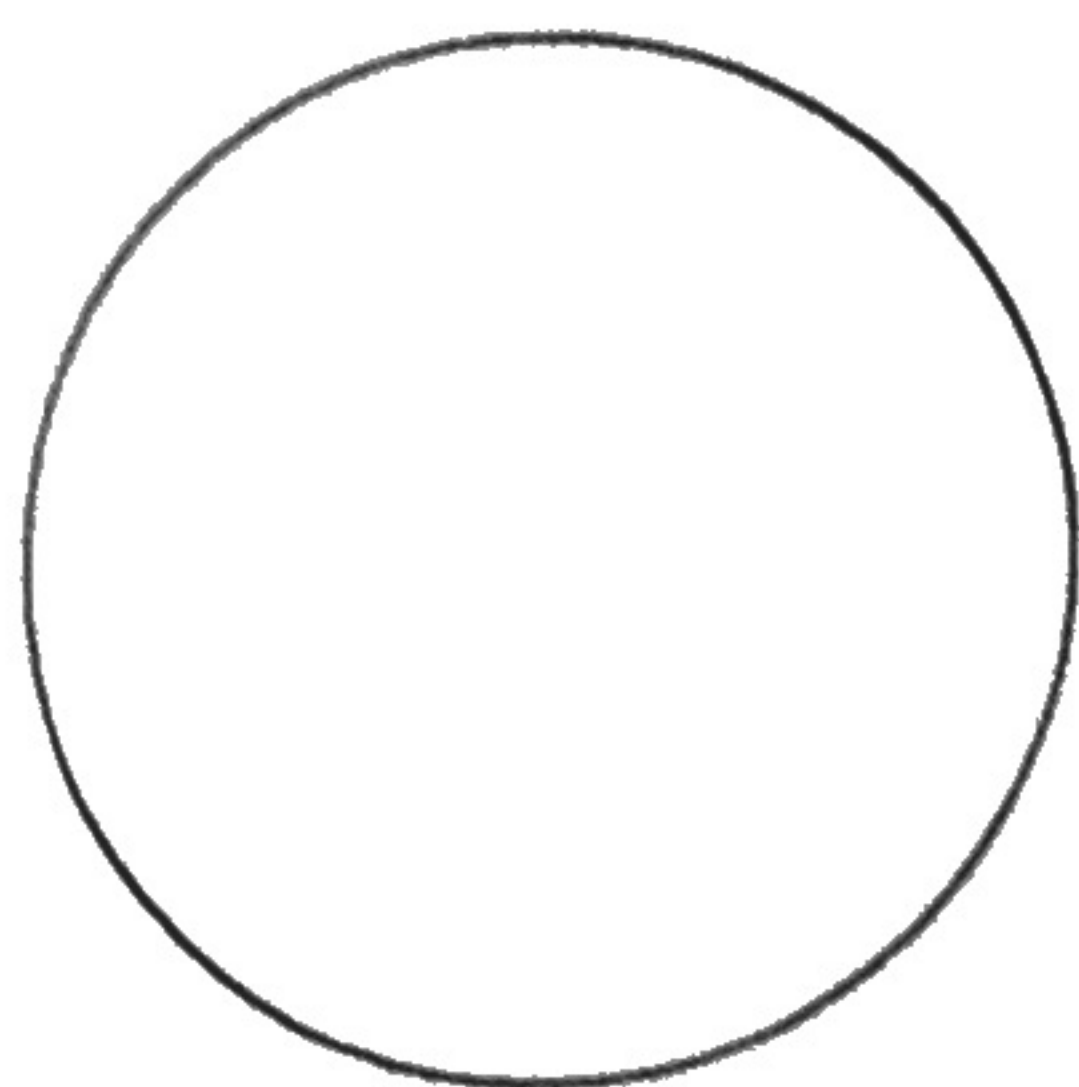
22. Prepare a new wet mount, this time using hair from people that are two different colors. Cross the hairs on the slide (it may be easiest to cut each hair to about a 1 cm length) and cover them where they cross. View the slide under low power and focus on where they cross. Draw the image that you see in the circle to the right.



23. Center the crossing point and switch to high power. Focus on the lighter of the two hairs, using the fine adjustment knob.

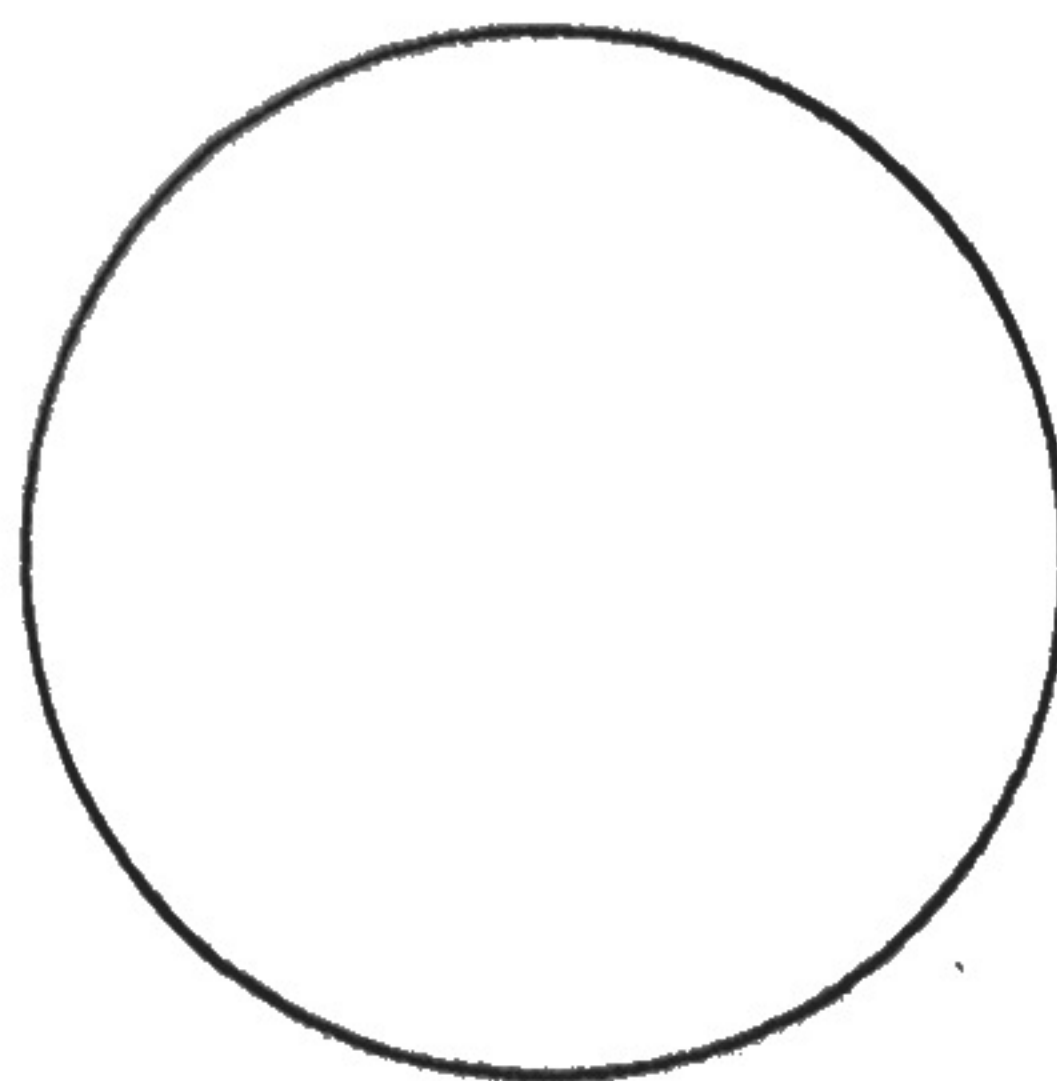
- n. Is the darker hair in focus at the same time as the lighter hair? Explain.

24. Prepare wet mounts of various other things, such as pieces of cloth, a fly's wing, cotton, or anything else that is thin enough to make into a wet mount. Draw your observations in the circles below, labeling each with a brief description of what the specimen is and what magnification was used for the drawing.



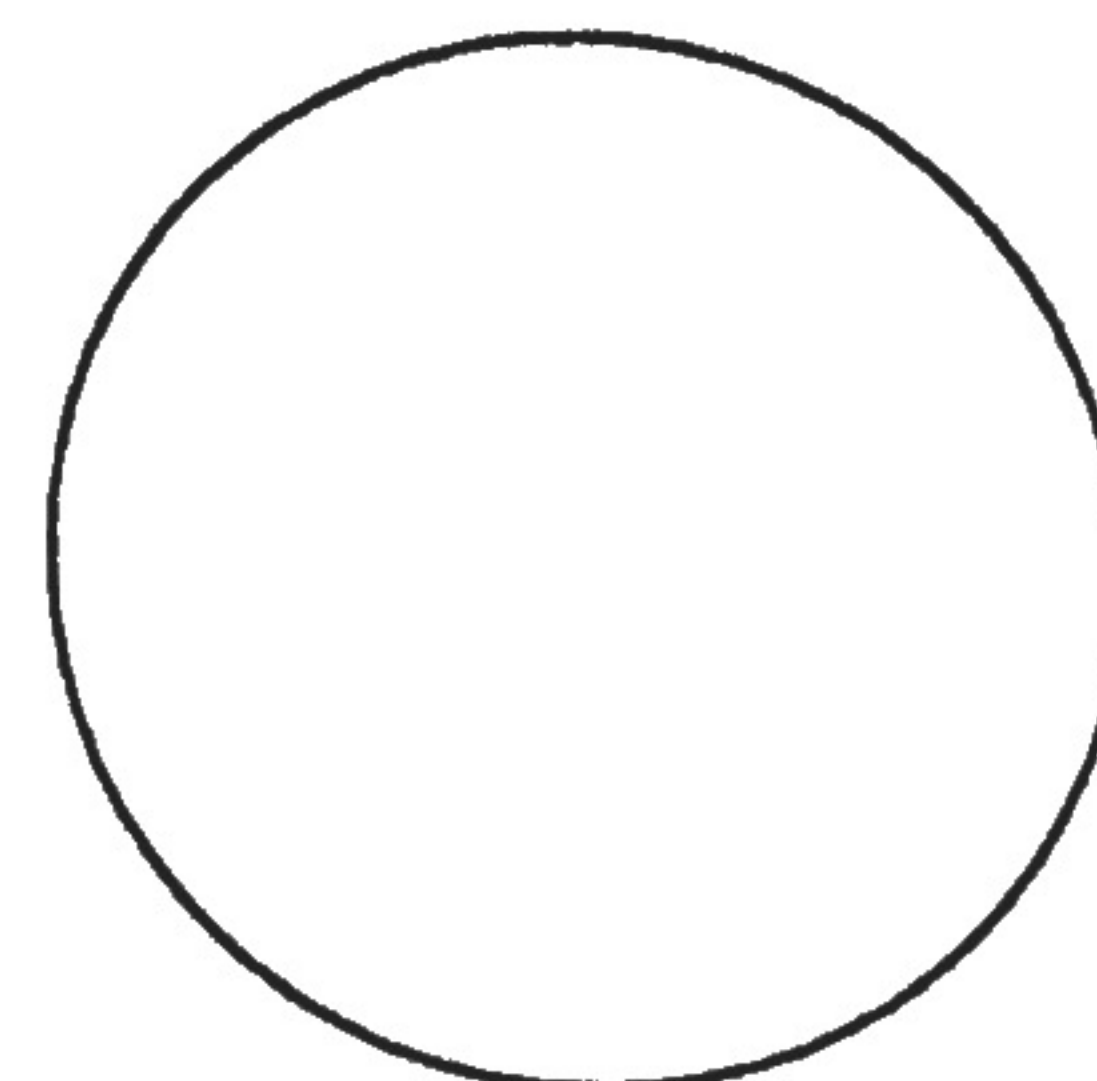
Object:

Magnification:



Object:

Magnification:



Object:

Magnification:

**Final Analysis:**

1. Why should you always begin to use a microscope with the low-power objective?
  
  
  
  
  
  
  
  
  
  
2. Why should you only use the fine adjust when the high-power objective is in position?
  
  
  
  
  
  
  
  
  
  
3. Why must the specimen be centered before switching to high power?
  
  
  
  
  
  
  
  
  
  
4. If you placed a letter "g" under the microscope, how would the image look in the field of view?
  
  
  
  
  
  
  
  
  
  
5. If a microscope has an ocular with a 5x power, and has objectives with powers of 10x and 50x, what is the total magnification of: (Show your math for full credit!)
  - a. low power?
  
  
  
  
  
  
  
  
  
  
  - b. high power?
  
  
  
  
  
  
  
  
  
  
6. If you are looking through a microscope at a freshly prepared wet mount and you see several perfect circles that are completely clear surrounding your specimen, what is the most likely explanation?