Microscope Lab

Introduction to the Microscope Lab Activity

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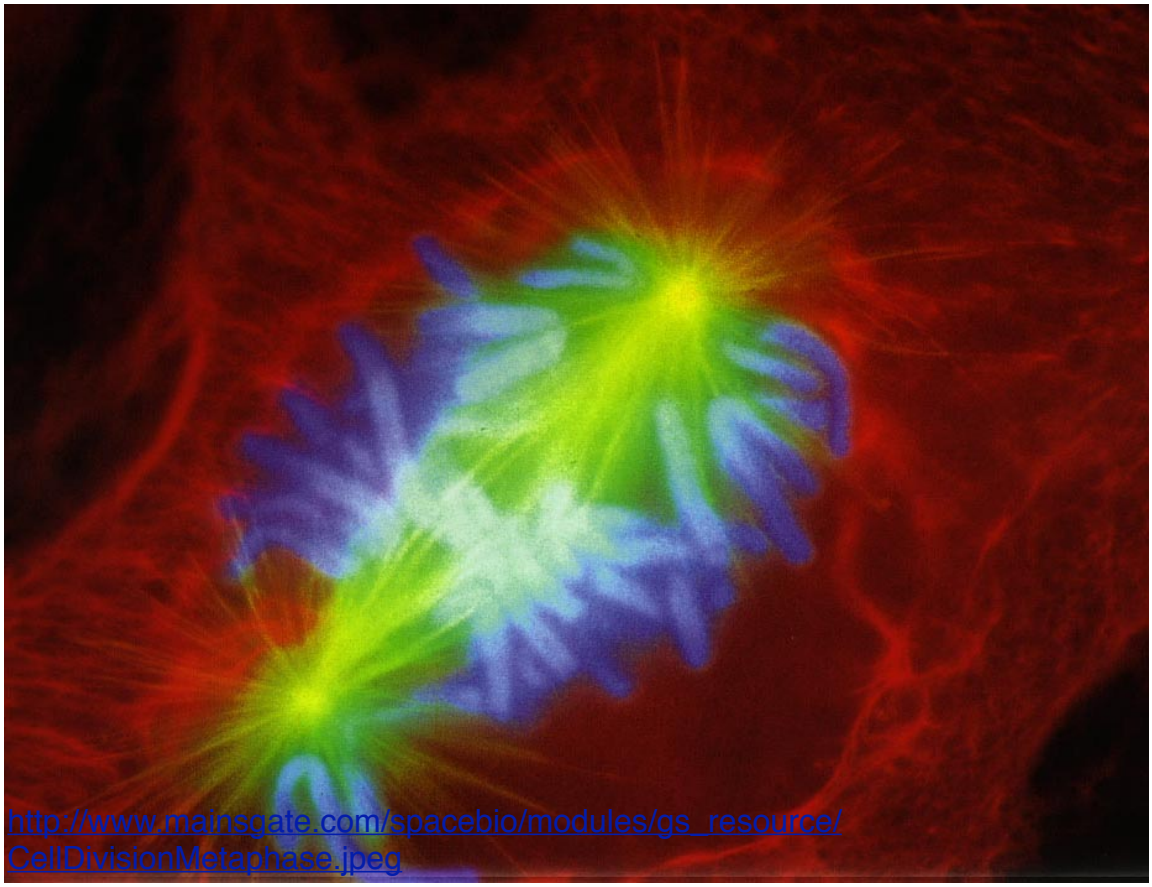


http://www.mainsgate.com/spacebio/modules/gs_resource/CellDivisionMetaphase.jpeg

Introduction

Microscope is a tool used to enlarge images of small objects that are hard to study with bare eyes. The compound light microscope, which is going to be used in this lab activity, is an instrument with two lenses and various knobs to focus the image. In this lab, we will learn about the proper use and handling of the microscope.

Objectives:

- Demonstrate the appropriate procedures used while using the compound light microscope correctly.
- Make and use a wet mount.
- Calculate the total magnification of the microscope.
- Explain how to handle the microscope properly.
- Describe changes in the filed of view and the amount of light when going from low to high-power objectives using the compound light microscope.
- Explain why objects must be centered in the field of view before changing from low to high-power objective.
- Explain how to control the light intensity when changing the power of objectives.
- Explain the proper process for focusing under low and high-power using the compound light microscope.

Procedures

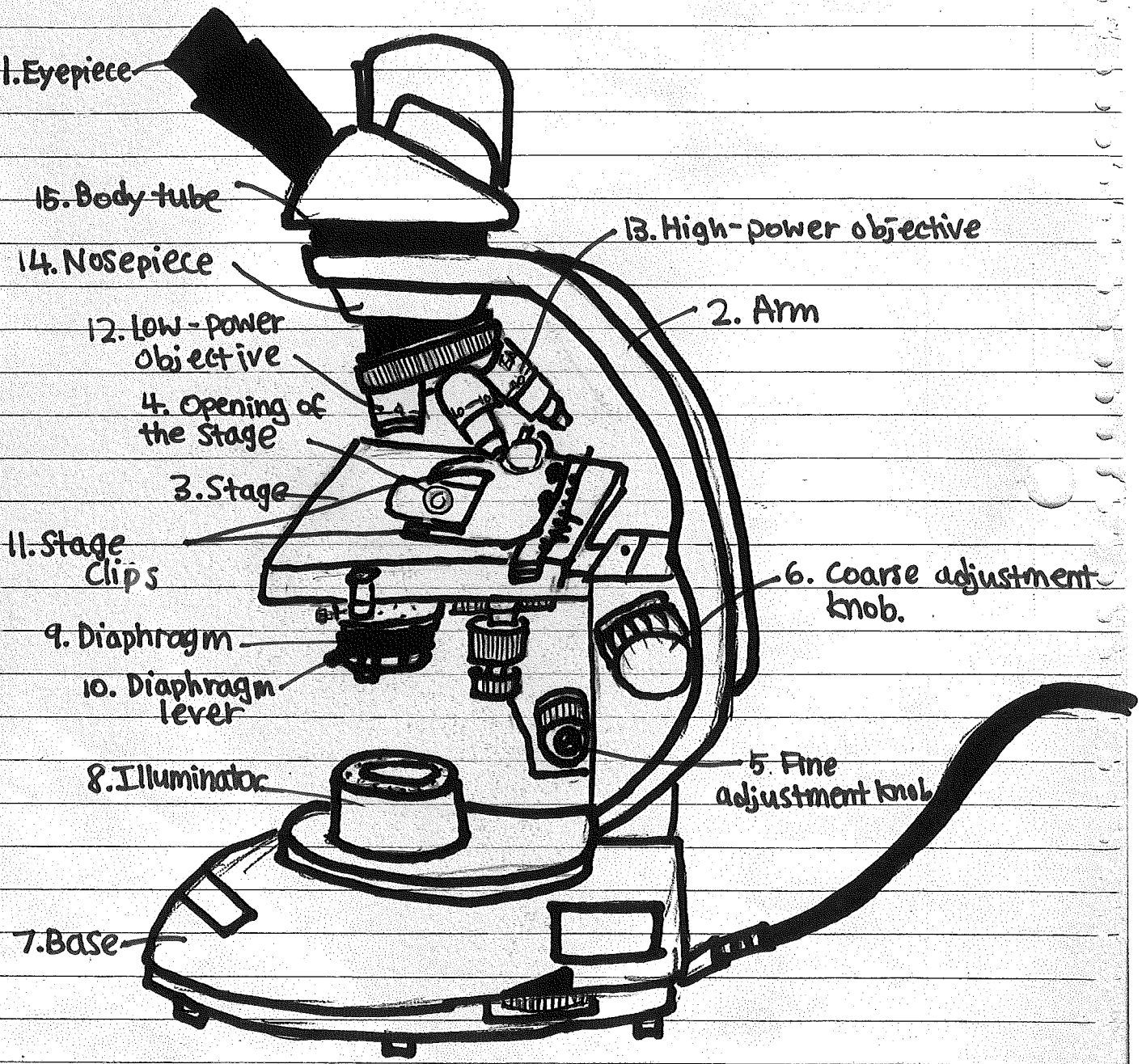
Materials

- Compound Microscope
- Glass slides
- Cover slips
- Eye dropper
- Beaker of water
- The letter “e” cut from newsprint
- Scissors
- Tooth picks
- Iodine
- Plant or algae specimens

Microscope Handling

1. Microscope should be treated with care; put one hand on the arm and the other under the base of the microscope when carrying it.
2. Carry one microscope carefully and properly from the microscope storage area to the working area.
3. Pick up a pair of scissors, newsprint, a slide, and a coverslip.
4. Remove the dust cover of the microscope and set it properly.
5. Examine the microscope and give the function of each of the parts.

Parts of the Microscope

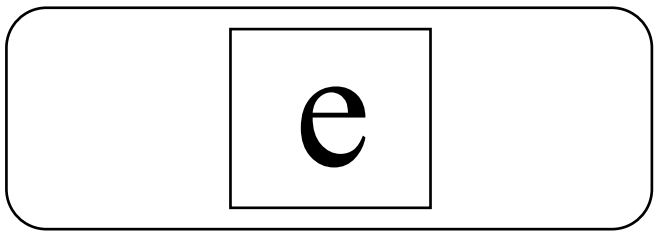


Functions of the Microscope

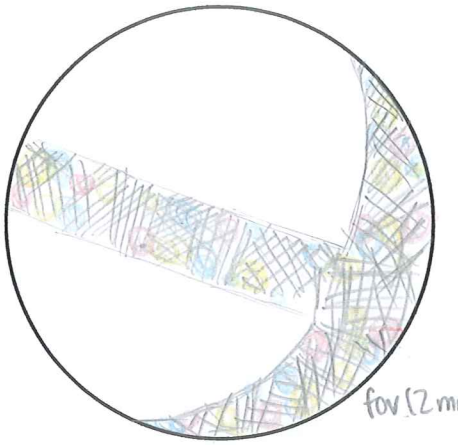
No.	Name	Function
1	Eyepiece	Contains a magnifying lens
2	Arm	Supports the body tube
3	Stage	Supports the slide being observed
4	Opening of the stage	Allows light to pass up to the eyepiece
5	Fine adjustment knob	Moves the body tube slightly to sharpen the image
6	Coarse adjustment knob	Moves the body tube to focus the image
7	Base	Supports the microscope
8	Illuminator	Produces or reflects light up to the eyepiece
9	Diaphragm	Controls the amount of light passing up toward the eyepiece
10	Diaphragm lever	Opens and closes the diaphragm
11	Stage clips	Holds the slide
12	Low-power objective	A shortest objective that magnifies 10X (4X sometimes)
13	High-power objective	A longest objective that magnifies 40X (100X sometimes)
14	Nosepiece	Holds the objectives and can be rotated
15	Body tube	Keeps proper distance between the eyepiece and the objectives.

Preparing a wet mount of the letter "e".

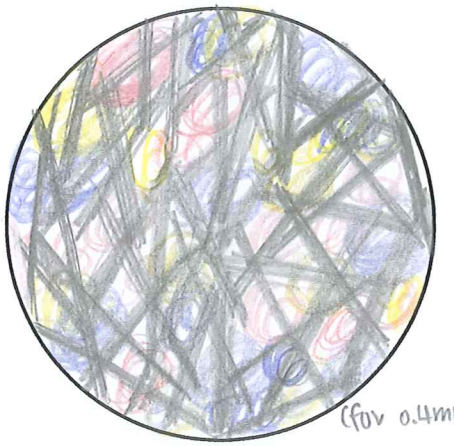
1. Cut out letter "e" from the newspaper.
2. Place it on the glass slide.
3. Cover it with a clean cover slip so it looks like :



4. Place a drop of water on the edge of the cover slip using the eyedropper.
5. Turn on the microscope and place the slide on the stage.



This is an "e" on low power, with the total magnification of 100X. The field of view for this one is 2 mm. When we are working on this, 10X objective was the smallest objective, so we couldn't get the entire "e" and the size of it. However, we got an curved part of an "e".



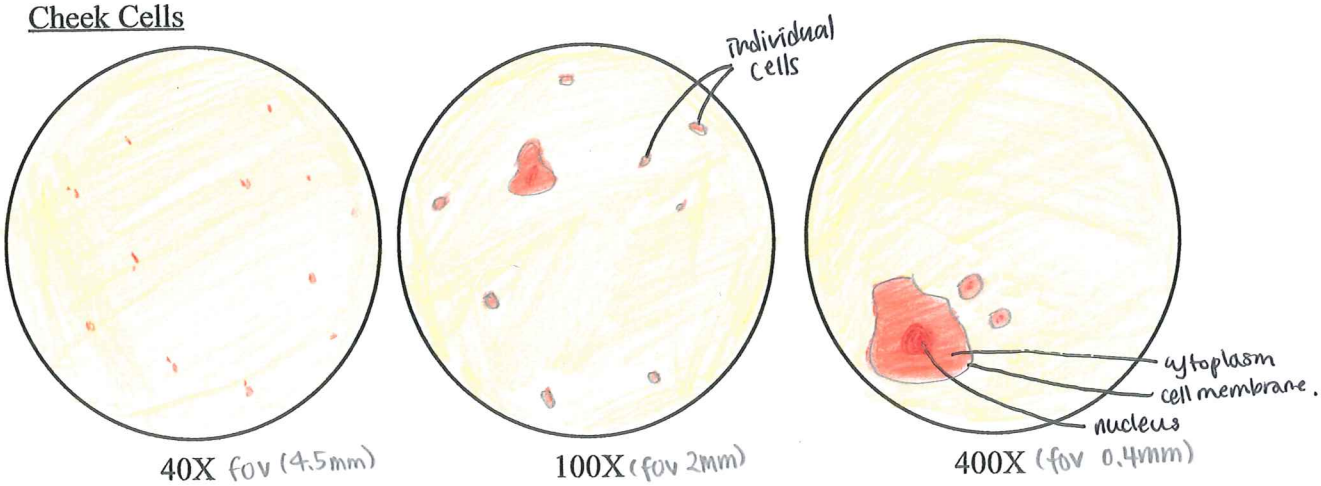
This is an "e" on high power, with the total magnification of 400X. The field of view for this is 0.4 mm. There are black stripes that makes up "e", and there are three colors - red, yellow, and blue - behind the black stripes.

The image seen through the eyepiece and the actual image on the stage are different. The image is reverted 180 degrees. (e→ə) Also, when the slide moves to the upper right area of the stage, the image through the eyepiece moves to the lower left area. Similarly, when the actual slide moves to the lower left side of the stage, the image moves to the upper right area. The intensity of light changes when the diaphragm moves. The image gets darker as the diaphragm moves left and vice versa.

Plant & Animal Cells Close Up!

1. Prepare slides of various plant or algae specimens available in the class using the wet-mount technique described above.
2. Scrape the inside of the cheek using a toothpick.
3. Put the cheek cell on the glass slide and put a drop of water and iodine.
4. Examine under various powers at least and sketch the observations.

Cheek Cells

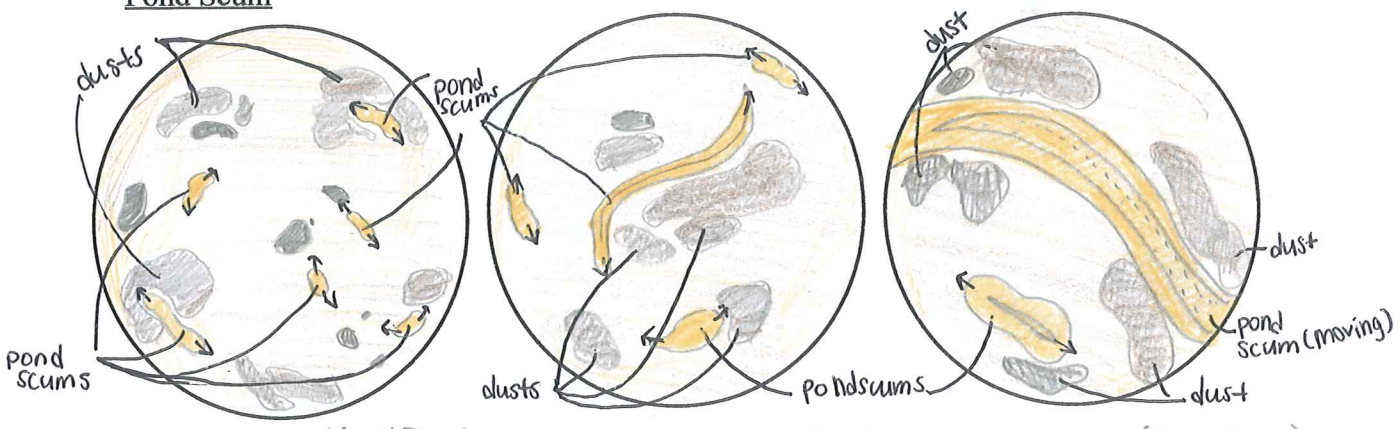


40X fov (4.5mm) 100X (fov 2mm) 400X (fov 0.4mm)

These are the observations of cheek cells. The left one is the lowest power, 40X, and the field of view is 4.5 mm. The middle one, with total magnification of 100X, has 2 mm for the field of view. The right one is oh the highest power, 400X, and the field of view is 0.4 mm. Thus, the biggest cell shown in the 400X graph indicates that its diameter is around 0.1 mm. The background of the specimen is yellow because it was stained with iodine.

5. Take several samples of "pond scum" and make a slide. Again, examine and sketch the observations. Also, examine other prepared slides available in class.

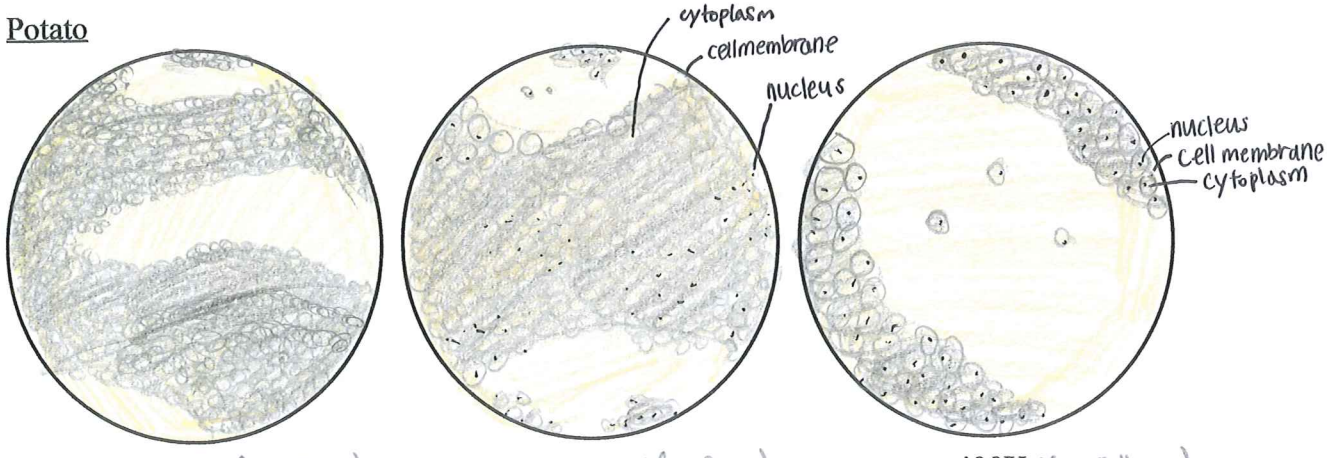
Pond Scum



40X (fov 4.5mm) 100X (fov 2mm) 400X (fov 0.4mm)

These are the observations of the pond scum. Unlike others, there were living organisms in this slide. The gray and black dots in the drawings are probably dust or tiny rocks, and the brownish yellow drawings are the pond scums. They were moving front and back, like a worm. When the magnification was 40X, it seemed that the whole cluster was moving, but when 100X, a long pond scum appeared and it kept moving. From this, the length of the scum would be around 1.3 mm.

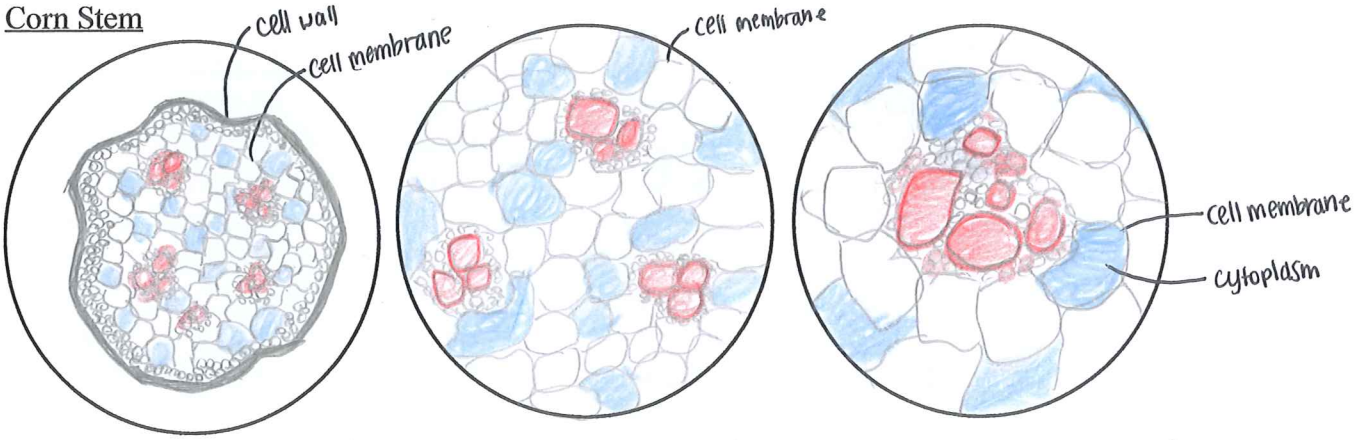
Potato



40X (fov 4.5mm) 100X (fov 2mm) 400X (fov 0.4mm)

These are the observations of the potato. Because I put too much iodine, it was hard to observe the specimen. It rather seemed like a black cluster, not a group of cells. However, as I magnified, I could see cells more clearly. From the drawings of 400X view, one potato cell would have a diameter of about 0.03 mm. However, there might be some errors since my drawings are not perfect, and there was too much iodine.

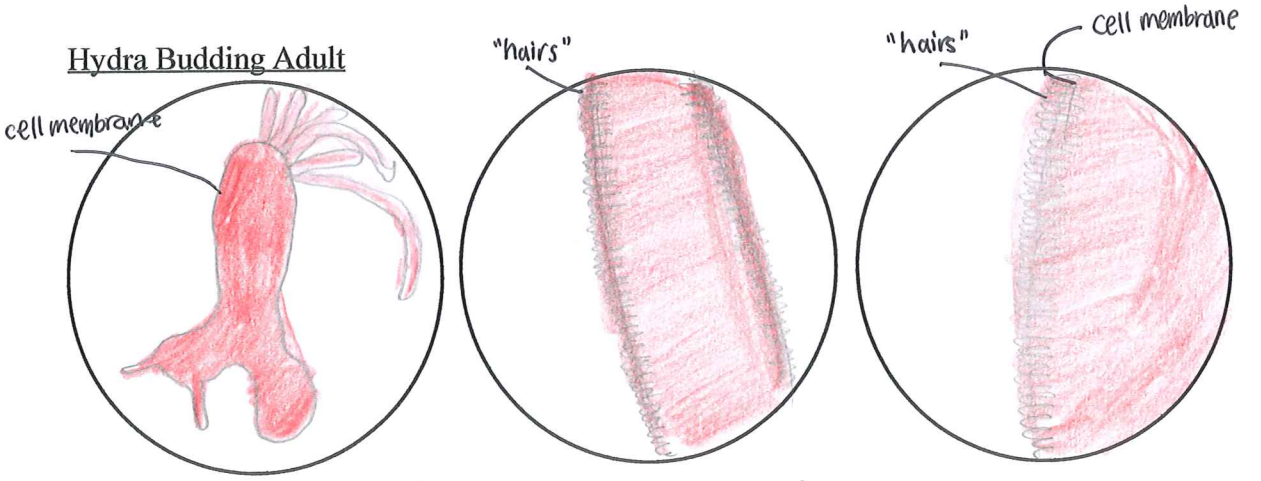
Corn Stem



40X (fov 4.5mm) 100X (fov 2mm) 400X (fov 0.4mm)

These are the observations of corn stem. This was not made by me and my partner; it was one of the prepared slides in the class. In the 40X view, I could see the entire cell, and thus I can assume that the cell's diameter is around 3.6 mm. There is a thick cell wall, and there are tiny cells around that wall. There are blue, white and red cells. When it is magnified, the red cell turns out to be a group of about three ore more red cells. Also, they are surrounded by tiny cells.

Hydra Budding Adult



40X (fov 4.5mm) 100X (fov 2mm) 400X (fov 0.4mm)

These are observations of hydra budding adult. It was also one of the prepared slides. As seen in the 40X view, the specimen seems to be almost 4.5 mm long. It has two heads and several tails at each ends. When it is magnified, its edge is made up of "hairs". The entire specimen is red in color. I couldn't get clear image in the 400X view, but I could still recognize the image/specimen.

Determining Total Magnification

There is a rule for determining total magnification of a compound microscope. To find out the total magnification, multiply the number on the eyepiece magnification and the number on the objective magnification. For instance, the total magnification of a low power objective:

Eyepiece magnification	(X)	Objective magnification	=	Total Magnification
10	(X)	4	=	40X

And for a high power objective:

Eyepiece magnification	(X)	Objective magnification	=	Total Magnification
10	(X)	100	=	1000X

Conclusion (includes answers to the conclusion questions)

When handling a light microscope properly, there are two procedures that should be taken carefully. First, we should start observing an object from the low-power objective. To get the image focused when using the low-power objective, we should use coarse adjustment knob to adjust it first, not fine adjustment knob. Fine adjustment knob should be used later, such as when looking at high-power objectives. Otherwise, the slide and the objective can be damaged. Also, we should always treat the microscope with great care. To be specific, the microscope should be carried with two hands; one holding the arm, and another holding the base. Also, when putting down the microscope, it should be placed gently. When not using the microscope, it should be covered with the dust cover and kept.

The compound light microscope is the most common microscope that are used in biology classes. The light microscope is also called compound microscope because it contains two lenses. Unlike a simple microscope that uses one lens, a compound microscope uses more than one lens.

As shown in the section of observing an "e", the images observed under the light microscope are reversed and inverted. This is because when the light passes the lens of the eyepiece, the image gets inverted since those lenses are usually convex lenses. The image is erected again on the retina.

The specimen must be centered in the field of view on low power before going to high power because if the specimen is observed on high power from the beginning, it gets very hard to find the specimen. When low-power objective is used from the beginning, we are can start the observation with the entire image of the specimen. Then, it gets much easier to observe the specimen when magnified.

Let's suppose that there is a microscope that has a 20X ocular (eyepiece) and two objectives of 10X and 43X respectively. Then the low-power magnification of this microscope would be:

(Eyepiece magnification) x (Objective magnification) = (Total Magnification)

(20) x (10) = (**200X**)

Also, the high-power magnification of this microscope would be:

(Eyepiece magnification) x (Objective magnification) = (Total Magnification)

(20) x (43) = (**860X**)

There are three steps to make a proper wet mount of the letter "e". First, place the letter "e" from the newspaper on the clean glass slide. Then, put a drop of water on the specimen using a pipette. Finally, place a clean coverslip and remove any excess water at the edge. Make sure when putting a coverslip, the lower one edge of the coverslip so that it touches the side of the drop of water at about a 45 degrees angle. Also, lower the coverslip slowly using a needle or probe.

When going from low to high power using the compound microscope, the field of view and the amount of available light changes. The field of view narrows down and gets smaller as the objectives uses higher power. Also, the amount of available light decreases when going from low to high-power objectives.

Therefore, the user may have to combat the problems incurred with the microscope when the power changes from low to high. Since the field of view gets smaller, the user may move the stage around and observe the specimen. He may also adjust diaphragm so that there is appropriate amount of light that would not hurt his eye.

In addition, the procedure for using the microscope differs slightly under high-power as opposed to low-power. When using a low-power objective, a coarse adjustment knob should be used to focus the image, unless it is very easy to damage both the slide and the objective. On the other hand, when using a high-power objective, use a fine adjustment knob to focus the image sharply, since the specimen is generally focused when using a low-power objective previously.

Other than the compound light microscope, there is also a microscope called stereomicroscope. Unlike the compound light microscope which gives two-dimensional views, the stereomicroscope gives three-dimensional image. Therefore, stereomicroscopes are often called dissecting microscopes because they offer the depth of field which is necessary to control the objects while observing them.

In addition to light microscopes, there is another type of microscopes - electron microscopes. Electron microscopes use beams of electrons, not light, to produce images. Electron microscopes can present more clear images of even smaller objects compared to the light microscopes. Electron microscopes are classified into two main types: transmission electron microscopes (TEMs) and scanning electron microscopes (SEMs). TEMs shine a beam of electrons through a thin specimen, and thus they reveal very details inside the cell. SEMs scan a narrow beam of electrons back and forth across the surface of a specimen. Therefore, they offer three-dimensional images of the surface of the specimen in a very realistic and dramatic way.

Timeline for the Various Discoveries of Early Microscopy

1590 Zaccharias Janssen and son Hans Janssen, two Dutch eye glass makers, created the forerunner of the compound microscope and the telescope.

1665 Robert Hooke, English physicist, looked at a silver of cork through a microscope lens and discovered cells.

1674 Anton van Leeuwnehoek built a simple microscope with one lens. He used it in observing blood, yeast, insects and other small objects.

18 C Microscopes improved as the technical innovations took place. Scientists found out that combining two lenses reduces "chromatic effect", the disturbing halos resulted from differences in refraction of light.

1903 Richard Zsigmondy invented the ultramicroscope. It enabled scientists to study objects below the wavelength of light.

1931 Ernst Ruska began to build the electron microscope. This was a TEM.

1932 Frits Zernike created the phase-contrast microscope. This enabled scientists to study colorless or transparent objects.

1936 Erwin Wilhelm Muller invented the field emission microscope.

1951 Erwin Wilhelm Muller invented the field ion microscope, which is the first to see atoms.

1981 Gerd Bining and Heinrich Rohrer developed the scanning tunneling microscope (STM).

1986 Gerd Bining, Quate, and Gerber created the atomic force microscope (AFM).

In this lab activity, I observed not only the external features and functions of the microscope, but also the specimens magnified through the microscope. I also made a specimen myself, and drew my observations carefully. Although some fundamental procedures were things that I already learned from the middle school, it was a good opportunity to remind the steps that I should be careful when carrying out, such as placing a coverslip, carrying a microscope, and procedures of observing specimen safely.

I remember myself having hard time getting clear image of a specimen. I improved my skills of focusing images through this activity. Now I can use a coarse adjustment knob and a fine adjustment knob without any problems.

However, I think I barely controlled the intensity of light using diaphragm. Thus, if I have any chance to use a microscope again next time, I will try to get used to using diaphragm and controlling the amount of light. Also, although I kept reminding myself that I should be careful when treating the microscope and slides, I think I was not still careful when placing the specimen on the stage and changing the objectives from low to high. So, I will also try to improve this next time.

Also, I dropped to much iodine when staining the specimen, especially when making a potato slide. Because the specimen was stained too much, it was hard to distinguish the cells. So next time, I should drop less but enough amount of iodine so that I can observe the specimen more clearly.