

GENERAL BIOLOGY LABORATORY MANUAL



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General Biology Laboratory Manual

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CHAPTER OVERVIEW

Introduction

[0.1: Guidelines for Laboratory Success](#)

[0.2: Lab Safety Rules](#)

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0.1: Guidelines for Laboratory Success

Guidelines for Laboratory Success

1. Review and adhere to the lab safety rules (see next page).
2. Sign the lab safety contract and turn it in to your instructor at the beginning of the first lab meeting.
3. Read the Objectives at the beginning of each laboratory before meeting for class.
4. Understand the hypotheses being tested and how your experiments will test these hypotheses.
5. Be able to describe how your results support or refute your hypotheses.
6. Be able to read and interpret graphs and produce graphs from your own results.
7. Answer all questions within each laboratory exercise.
8. Utilize the “Laboratory Notes” pages at the end of each exercise to make notes as your instructor introduces the day’s activities.

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0.2: Lab Safety Rules

Lab Safety Rules in Student Center Building and Science Wing

- Know the location of the **safety features** of the laboratory, including the safety shower, eyewash, fire extinguisher, spill kits, and the emergency gas & electrical shut-offs.
- Food, drink, gum chewing, and cosmetic application, including lip balm, are not allowed in the laboratories, hallways, or laboratory coaching area.
- **Cell phones**, smart watches, or other similar wrist-wear items are not permitted within the laboratories.
- All personal items not needed for class are to be stored in the **lockers** located in the hallway outside the labs. Students must provide their own lock & overnight storage is not permitted.
- For your safety and the safety of others, **DO NOT** sit on the hallway floors of the Science Hall. There are several seating areas in front of the Science Hall.
- Compliance with the **Laboratory Personal attire policy** and all PPE (personal protective equipment) outlined by the laboratory instructor is essential.
- **Wash** hands before leaving the laboratory, after removing gloves, and after contamination.
- All spills, broken equipment, or exposures must **be reported** to your instructor immediately.
- Lab equipment and supplies must not be removed from the laboratory.
- Know the **disposal guidelines** posted in the lab and utilize them appropriately. If unsure, ask your instructor for guidance.
- Unauthorized experiments or procedures must not be attempted.
- **Leave** your workstation clean and in good order before you leave the lab.
- Compliance with **the instructor syllabus** and rules specific to the area of study is expected and enforced.
- In biohazardous areas (NWTC Green Bay campus - SC360C, SC360F, SC367, SC378, SC379, SC380, SC381. SC382 and NWTC Marinette campus - Gateway Lab SC104 & BT115c), all **laboratory notebooks** must have a wipeable outer covering to disinfect when entering and exiting the lab. All **writing utensils** will be provided and must stay within the laboratory.

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0.3: Lab Safety Contract

? Lab Safety Contract to Submit

I, _____, agree to all lab safety guidelines and understand that failure to adhere to these guidelines may result in grade deductions or dismissal from the course.

Signature: _____

Date: _____

Once you have signed/dated this agreement, please tear out this page and turn it in to your instructor at the beginning of the first lab meeting.

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CHAPTER OVERVIEW

Laboratory 1: The Scientific Method

- [1.1: Lab 1 Introduction](#)
- [1.2: Using the Scientific Method](#)
- [1.3: Applying the Scientific Method](#)
- [1.4: Laboratory 1 Notes Page](#)

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1.1: Lab 1 Introduction

Scientific Method

The scientific method is central to the study of biology. It is a process of acquiring and verifying information through experimentation. The general steps of the scientific method are depicted in the figure below.

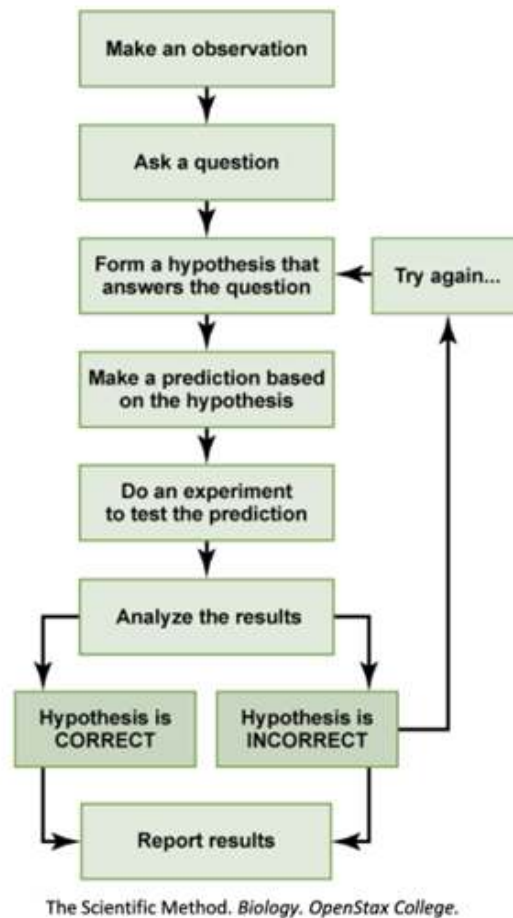


Figure 1.1.1: The Scientific Method. Biology. OpenStax College.

The hypothesis, or suggested explanation for the observation, is the basis for setting up experiments. Good experimental design is essential to the scientific method. A few keys to good experimental design include effective use of controls, reproducibility, a large sample size, and multiple trials. In an experiment, in order to determine that any changes that occur are due to investigator manipulation only, there must be some basis for comparison. A control group is necessary to establish this basis of comparison. In the control group, everything is kept the same as the experimental group except for the independent variable. The experimental group is actually being experimented upon.

For example, in a drug trial, there will be a group that receives the drug (the experimental group) and a group that receives a placebo (the control group). The drug itself is considered the independent variable and any change(s) that occur because of the drug are considered the dependent variable. In order to ensure that it is only the drug causing changes, all other variables must be tightly controlled (such as diet, exercise, smoking, etc.). These are referred to as controlled variables.

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1.2: Using the Scientific Method

Part 1: Using the Scientific Method

Lab Objective and Materials

- In this experiment, you will determine how temperature affects the rate of dissolution of antacid tablets in water.

Materials

- 5 antacid tablets
- 3 beakers (250 mL)
- 1 thermometer
- Tape to label beakers
- Hot plate
- Timer/stopwatch

Procedure:

- Create hot, cold, and room-temperature water.
 - Hot water: Place 200 mL of water in a beaker and heat up to approximately 60°C using a hot plate. Label “Hot”.
 - Cold water: Add ice to approximately 200 mL of water in a separate beaker. Once the water is cold, add 200 mL of cold water to a new beaker. Label “Cold”
 - Room temperature: Place 200 mL of water at room temperature in a beaker. Label “Room Temp”
- Open antacid packages and cut/break each tablet in half.
- Record the temperature (in degrees Celsius/°C) in each cup in Table 1.2.1.
- Drop half of one antacid tablet in each treatment – hot, cold, and room temperature.
- Start the stopwatch and record how long each treatment takes to dissolve the tablet. Record data in Table 1.1 under Run #1.
- Once finished, pour the antacid solution down the drain and rinse the beakers.
- Repeat two additional trials and record data in Table 1.2.1 under Run #2 and Run #3.

Table 1.2.1

Run	Beaker	Temperature (°C)	Time (sec)
Run #1	Hot		
	Room Temp		
	Cold		
Run #2	Hot		
	Room Temp		
	Cold		
Run #3	Hot		
	Room Temp		
	Cold		

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1.3: Applying the Scientific Method

Part 2: Applying the Scientific Method

Lab Objective and Materials

- Investigate the effect of temperature on glow stick brightness. You need to develop a plan for how you will perform your experiment.

Materials

- 3 250-mL beakers
- 3 glow sticks (obtain glow sticks that are all the same color)
- Thermometer
- Timer
- Tap water
- Ice cubes
- Hot plate

Procedure:

- Clearly state the problem or question you are trying to solve.
- State the following:
 - Independent variable:
 - Dependent variable:
 - Hypothesis based on the information you already know:
- Create a detailed step-by-step procedure for your experiment. Refer to what needs to be included for a good experiment.
- You will be measuring brightness based on a scale from 0-10. The uncracked glow stick will be assigned a “0” on the brightness scale. The beaker with room temperature water will be assigned a “5” on the brightness scale.
- Crack the glow stick and shake for about 30 seconds, then submerge the glow stick into the appropriate temperature beaker.
- Record brightness at Time 0 in Table 1.3.1.
- Set a timer for 10 minutes and press “start”.
- Record brightness after 10 minutes have passed in Table 1.3.1.

Table 1.3.1: Glow Stick Brightness

Beaker	Temperature (°C)	Brightness at Time 0	Brightness after 10 minutes
Hot			
Room Temperature			
Cold			

- Show your results in the form of a bar graph, including all necessary information (i.e., x-axis = independent variable, y-axis = dependent variable)
- Did your findings support your initial hypothesis? Explain.

? Follow-Up Questions: Laboratory 1

- What are the steps of the scientific method (in order)?
- What is a hypothesis? What conditions must a good hypothesis satisfy?

3. What is the difference between an independent variable and a dependent variable?
4. What is the purpose of a control in an experiment?
5. When graphing results, does the independent variable go on the x-axis or y-axis?

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1.4: Laboratory 1 Notes Page

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CHAPTER OVERVIEW

Laboratory 2: Organic Molecules

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[2.2: Proteins](#)

[2.3: Carbohydrates](#)

[2.4: Lipids](#)

[2.5: DNA Extraction](#)

[2.6: Laboratory 2 Summary and Notes](#)

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2.1: Laboratory 2 Introduction

Organic Molecules

Students often ask why they have to learn about chemistry in a biology class – isn't that a different science? Technically, yes! Chemistry is considered a physical science. Biology is a life science. Even though it has a different focus, chemistry is closely tied to biology.

Four major biological macromolecules make up the vast majority of living cells, and the way those macromolecules interact is solely due to their chemistry. Cells, tissues, organs, and organisms must ensure a balance in these molecules and many other solutes to maintain their life (this is called homeostasis).

On a larger scale, we consider the importance of chemistry in biology from an ecological standpoint. Scientists are closely following the Arctic ice sheet as it melts, increasing ocean levels worldwide. The fact that ice is less dense than liquid water and, therefore, floats is tied to chemistry. We track carbon dioxide levels and plastic particulates in the ocean too, both of which have an effect on the health of the planet.

Exercise: Biological Macromolecules and Their Indicators

Introduction/Objectives

There are four different biological macromolecules in cells. Each macromolecule has its own indicator test that can signal its presence in a cell or a solution.

Read carefully as the indicator tests are not the same for each macromolecule.

You should obtain your samples in test tubes. Follow the directions for performing each test, then empty and wash your test tubes. Refill them with samples and repeat the processes until you have tested each sample for the presence of each macromolecule.

When recording results in your data tables, use + and – symbols (e.g., +++ = strong positive, + = weak positive, - = negative).

Caution

Wear gloves and goggles while performing this lab!

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2.2: Proteins

Exercise 2A – Proteins

Lab Introduction and Materials

Proteins are made of repeating units of amino acids in complex structures with other functional groups. They are structural elements in the human body and also perform an untold number of chemical functions. They are tested with Biuret Reagent. When protein is mixed with Biuret, the solution turns lavender in color.

Materials:

- 6 test tubes
- Test tube rack
- Disposable pipettes
- 2 mL each of six different sample solutions
- 10-15 drops of Biuret Reagent (for each test tube)

Procedure:

1. Add 2 mL of one sample solution to a test tube.
2. Repeat Step 1 five more times, ensuring each test tube contains a different unknown.
3. Add 10-15 drops of Biuret Reagent to each test tube.
4. Record observations in Table 2.2.1 and answer follow-up questions.

Observations/Follow-Up Questions

Table 2.2.1: Biuret Test Results

Sample Tested	Positive or Negative for Proteins?	Description of Color Change (if any)

1. What does a positive protein test look like?
2. What is the indicator solution for a protein called?
3. What are the individual subunits of a protein molecule called?

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2.3: Carbohydrates

Part 2B - Carbohydrates

Lab Introduction and Materials

- Carbohydrates are made of simple sugars and can form complex substances called polysaccharides (e.g., starch and cellulose) when tightly coiled and chained together. The presence of carbohydrates, specifically reducing sugars, is tested using Benedict's solution. Benedict's solution is blue in color. If a sample changes from blue to red-orange or green after Benedict's solution is added, this is a positive test (and the sample contains a reducing sugar).
- The test for starch is different than the test for reducing sugars because of the difference in structure. Iodine (IKI or Grams) is used to test starch. When added to a substance or solution containing starch, iodine will quickly turn from its normal amber/brown color to dark blue or black. Occasionally, only small amounts of black particulate matter fall out solution. This means the starch was not completely dissolved in the solution – you are seeing clumps of “dye” or indicated starch. This should also be considered a positive result.

Materials for Testing for [Reducing Sugars](#):

- 5 test tubes
- Test tube rack
- Disposable pipettes
- 2 mL each of five different sample solutions
- 10-15 drops of Benedict's solution (for each test tube)
- Hot plate
- Test tube tongs/clamp
- A one-liter beaker containing water (for boiling)

Procedure for Testing for Reducing Sugars:

1. Add 2 mL of one unknown sample solution to a test tube.
2. Repeat #1, ensuring each test tube contains a different unknown.
3. Add 10-15 drops of Benedict's Solution to each test tube.
4. Place the test tubes in a beaker of gently boiling water (this speeds up the process!) and watch for a color change for 2-3 minutes.

Caution

You can place multiple test tubes in the beaker of gently boiling water at one time, but be very careful not to let the water overflow. If your test tubes are bouncing because of the bubbles from the boiling water, your hot plate temperature is too high.

5. Record your results for each test tube in Table 2.3.1.

Observations

Table 2.3.1: Benedict's Solution Test Results

Sample Tested	Positive or Negative for Sugars?	Description of Color Change (if any)

Lab Materials

Materials for Testing for Starches:

- 5 test tubes
- Test tube rack
- Disposable pipettes
- 2 mL each of five different sample solutions
- 5-7 drops of iodine (for each test tube)

Procedure for Testing for Starches:

1. Add 2 mL of one unknown sample solution to a test tube.
2. Repeat #1, ensuring each test tube contains a different unknown.
3. Add 5-7 drops of iodine to each test tube.
4. Record observations in Table 2.3.2 and answer follow-up questions on the next page.

Observations and Follow-Up Questions

Table 2.3.2: Iodine Test Results

Sample Tested	Positive or Negative for Starch?	Description of Color Change (if any)

1. What are monomers of carbohydrates called?
2. What test do you use to test for reducing sugars? What does a positive test look like?
3. What are large, complex carbohydrates called? How do you test for their presence in a solution?

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2.4: Lipids

Part 2C - Lipids

Lab Introduction and Materials

- Lipids (or fats) are tested in a few different ways. When you add lipids to brown paper, a stain will remain when the spot dries. Another test can be completed with a chemical called Sudan IV. When Sudan IV is added to a liquid solution that may contain fat, it will cause the solution to separate into visible fat and non-fat layers.

Materials:

- 6 test tubes
- Test tube rack
- Disposable pipettes
- 2 mL each of six different sample solutions
- 4-5 drops of Sudan IV (for each test tube)

Procedure for Testing for Lipids:

1. Add 2 mL of a single sample solution to a test tube.
2. Repeat #1, ensuring each test tube contains a different unknown.
3. Add 4-5 drops of Sudan IV to each test tube.
4. Record observations in Table 2.4 and answer follow-up questions.

Observations and Follow-Up Questions

Table 2.4.1: Sudan IV Test Results

Sample/Unknown # Tested	Positive or Negative for Lipids?	Description of Color Change (if any)

1. Lipids are also known as _____.
2. What two tests can be used to look for the presence of lipids in a solution?
3. What does a positive test look like (for both tests)?

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2.5: DNA Extraction

Part 2D – DNA Extraction: Finding the Molecule that Does NOT Have an Indicator Test

Lab Introduction and Materials

- Nucleic acids include RNA and DNA, which are made up of building blocks called nucleotides. RNA is a single strand of nucleotides. There are several forms of RNA, which play various roles in protein synthesis. DNA is a double strand of nucleotides held together by hydrogen bonds. DNA segments, called genes, are responsible for directing protein synthesis.
- Since there is no indicator test for nucleic acids. So, for this portion of the lab, we are going to extract DNA from a banana.

Materials:

- Plastic sandwich bag
- Half of a banana (unpeeled)
- 120 mL hot water
- 5 grams salt
- 2.5 mL dish soap
- 250-mL beaker
- Coffee filter
- Rubber band
- Test tube
- Test tube rack
- Chilled 91% ethanol
- Glass stir rod

Procedure for Extracting DNA from a Banana:

1. Pour 120 mL of hot water and 5 grams of salt into a bag of mashed banana. Gently mix for 30-45 seconds.
2. Add 2.5 mL of dish soap into the bag. Mix gently, avoid foam!
3. Place the bottom of a coffee filter into a beaker and secure it over the top with a rubber band.
4. Pour the contents of the bag into the filter and let it sit for several minutes until all liquid is in the beaker (you may throw away the coffee filter and its contents after this).
5. Slowly pour the liquid from the beaker into a test tube, filling it approximately halfway
6. Obtain chilled 91% ethanol. Tilt the tube and slowly pour the ethanol down the side of the tube until there is a column/layer of ethanol 2.5-5 cm tall (you want to keep the ethanol and liquefied banana as separate as possible, so complete this step slowly).
7. Let this layered mixture sit for 8 minutes.

Note

During this wait time, what do you see happening between the ethanol and the banana layer? It looks cloudy and may have tiny bubbles in it. The longer you wait, the more defined this layer becomes. This is the DNA pieces clumping together.

8. Insert a glass stir rod into the test tube and “spool” the DNA.

Follow-Up Questions

1. After performing the DNA extraction, describe the physical appearance and texture of DNA.
2. Draw and label a single nucleotide.
3. Complete the table below to compare/contrast DNA and RNA:

	DNA	RNA
Major Sugar		
Nucleotide Bases		
Single or Double-Stranded		
Function		

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2.6: Laboratory 2 Summary and Notes

? Laboratory 2 Summary Table

Summarize the results of experiments 2A through 2D in the table below (using + and – signs as you did in the data tables earlier).

Table 2.6.1: Summary of Organic Macromolecule Indicator Tests

	Biuret Reagent (Protein)	Benedict's (Sugar)	Solution Iodine (Starch)	Sudan IV (Lipids)
10% Glucose				
1% Starch				
1% Albumin				
Corn Oil				
Lemon Juice				
Unknown				

Laboratory 2 Notes

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CHAPTER OVERVIEW

Laboratory 3: Introduction to Light Microscopy

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[3.2: Total Magnification, Calculating Field of View, and Estimating Specimen Size](#)

[3.3: Examining Prokaryotic Cells](#)

[3.4: Examining Eukaryotic Cells](#)

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[3.6: Laboratory 3 Notes Page](#)

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3.1: Laboratory 3 Introduction

Introduction to Light Microscopy

A cell is the smallest unit of life. Whether comprised of one cell (like Bacteria) or many cells (like humans), we call it an organism. Thus, cells are the building blocks of all organisms.

What does it mean to be microscopic? Cells vary in size. With few exceptions, we cannot see individual cells with the naked eye, so scientists use microscopes (micro = “small”; -scope = “to look at”) to study them. A microscope is an instrument that magnifies an object. We photograph most cells with a microscope – these images are called micrographs.

The microscope you will be using uses visible light and two sets of lenses to produce a magnified image. The total magnification will depend on which objective lens you are using – the highest magnification possible on these microscopes is 1000x – meaning that an object appears 1000x larger than it is.

Lab Objectives

1. Understand proper usage and maintenance of a compound microscope.
2. Determine total magnification, field-of-view, and estimation of the size of a specimen.
3. Observe and differentiate between prokaryotic and eukaryotic cells using a compound microscope.

Microscope Care:

1. The microscope should be turned off when not in use.
2. Always carry a microscope with two hands, one supporting the base and the other holding the arm.
3. Do not touch the glass part of the lenses with your fingers. Use only special lens paper to clean the lenses.
4. When using the microscope, carefully place it on a flat, level bench.
5. Do not drag the microscope across the bench when using it – even small vibrations can cause large movements in the image, especially if being viewed at a high magnification.
6. Proper steps for storing the microscope after use:
 1. Turn the light source down to the dimmest level.
 2. Turn off the power source.
 3. Put the scanning objective lens in place for storage.
 4. Move the stage to its lowest position.
 5. Carefully wrap the power cord before storing the microscope
 6. Open the cupboard door and slide the storage shelf out before placing the microscope down with both hands.

Microscope Use:

Today, you will learn how to properly use a microscope. Don't get frustrated if you cannot find specimens immediately or when switching from low to high power. Like any other skill, this takes time, practice, and patience. I am here to help. Reading and following the information below will guide you through the learning process.

First, let's examine the important parts of the microscope!

1. **Ocular Lens (Eyepiece)** – lenses on the top of the microscope, offer 10x magnification
2. **Nosepiece** – portion of the microscope that holds the objectives over the stage.
3. **Objective lenses** – lenses that are directly above the stage
 1. 4x = scanning objective
 2. 10x = low power objective
 3. 40x = high power objective
 4. 100x = oil immersion lens
4. **Stage Clip** – a metal piece that holds the slide in place

5. **Aperture** – the hole in the middle of the stage that allows light from the illuminator/light source to reach the specimen.
6. **Iris Diaphragm** – regulates the amount of light in the condenser.
7. **Condenser** – focuses the light through the specimen
8. **Illuminator** – light source located below the specimen.
9. **Arm** – located in the back of the microscope, supports the objectives and ocular lenses. Also used for lifting and carrying the microscope.
10. **Mechanical Stage** – The fixed stage is a horizontal platform that holds the specimen.
11. **Coarse Adjustment** – Used to make relatively wide focusing adjustments to the microscope. Also, this should be done keeping in mind that the objective should not hit the slide. In addition, it should be stopped when the object is completely visible through the ocular.
12. **Fine Adjustment** – Used to make relatively small adjustments to the microscope. We use it to bring the specimen in perfect focus once the specimen is visible through the use of the coarse-focus adjustment. Also, focus slowly to avoid contact between the objective and the specimen.
13. **Stage Controls** – These are the controls for moving your slide forward/backward (top knob) and right/left (bottom knob).
14. **Base** – The base supports the microscope and houses the illuminator/light source.
15. **Brightness Adjustment** – This controls the amount of light. Located on the right side
16. **Power Switch** – Turns the microscope on and off.

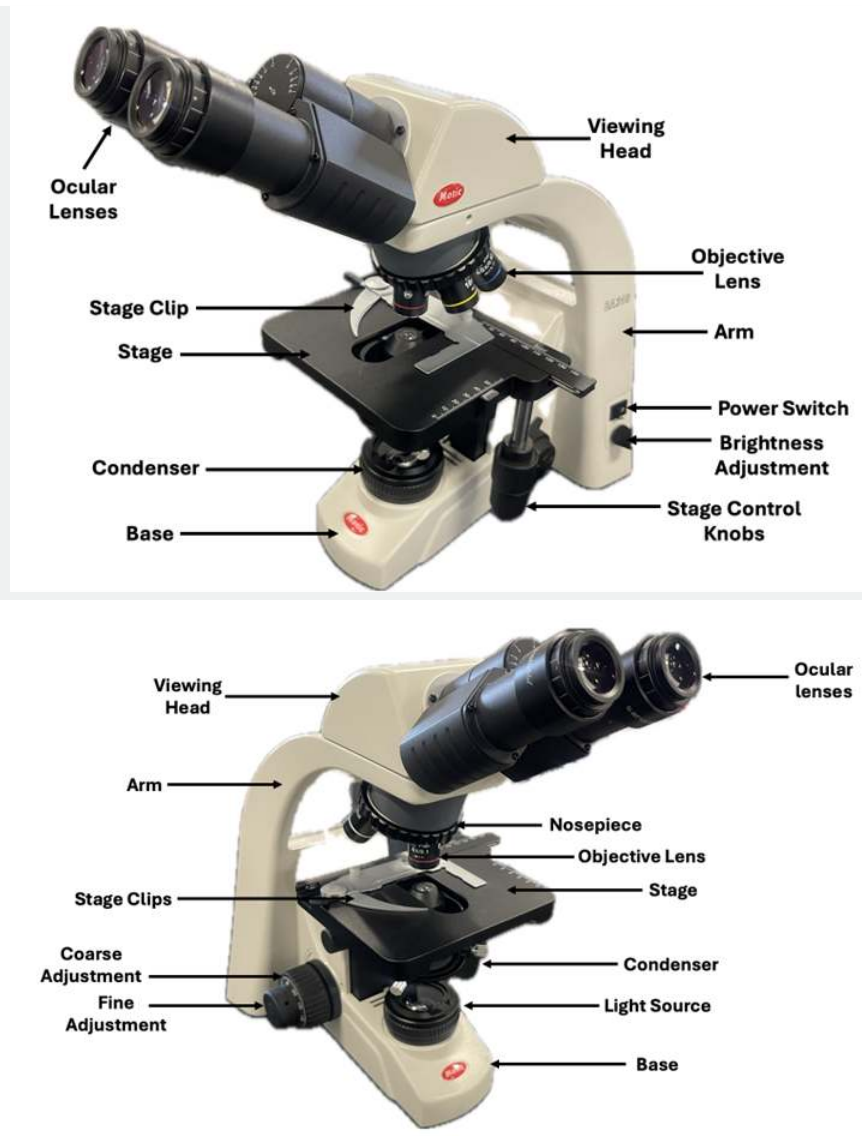


Figure 3.1.1: Important parts of a microscope. (Courtney Mayer, CC BY 4.0)

Appearance of Objects:

1. Objects appear inverted and reversed (i.e., backward and upside-down compared to how the specimen would appear on the slide as seen with the naked eye).
2. Only a thin layer of the specimen is in focus at any level: this is called depth of focus. You will notice that the image changes as you adjust the focus – this is because you can see different layers of the specimen.

Movement of Specimen:

1. Actual movement is opposite to the direction of movement under the microscope.
2. If an organism is moving right, it will appear to be moving left as you look through the ocular lenses.

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3.2: Total Magnification, Calculating Field of View, and Estimating Specimen Size

Total Magnification of Microscopes:

1. The ocular lens/eyepiece of the microscope magnifies the image of the specimen. Oculars are typically 10x lenses, meaning they magnify the image 10 times.
2. The objective lenses also magnify the image of the specimen. The objective lenses are called scanning power (4x), low power (10x), high power (40x), and oil immersion (100x).
3. You can identify the magnification of your microscope lenses by reading the numbers stamped on the objectives and the ocular lenses.

To calculate total magnification of an image under the microscope:

$$\text{Total Magnification} = \text{Ocular lens magnification} \times \text{objective lens magnification}$$

Example: If the ocular lens has a 10x magnification and the objective has a 40x magnification, the total magnification is **10 x 40 = 400xTM**

Changing Objectives:

1. When changing objectives from scanning power to low power to high power, the following changes will occur:
 1. The size of the field of view decreases.
 2. The field of view becomes darker.
 3. The size of the image increases.
 4. The resolution (ability to see detail) increases.
 5. The working distance between the slide and the objective lens decreases.
 6. The depth of focus (thickness of the specimen that is visible) is reduced.
2. When changing from scanning to lower power, the field of view gets smaller. In fact, every time you increase the power of the objective, the field gets smaller.

✓ Steps for Using the Microscope

1. Turn on the microscope using the power switch located on the bottom right-hand side of the base.
2. Ensure your microscope is set at the scanning power (4x) objective.
3. Place the slide on the stage, lining it up with the rectangle and using the stage clip to hold it in place.
4. Look into the eyepiece.



Figure 3.2.1: A slide on the stage of a microscope (Courtney Mayer, CC BY 4.0)

5. Use the coarse adjustment knob to bring the specimen into view. The specimen must be in focus before moving to the next steps.
6. Rotate the nosepiece to the low-power (10x) objective.
7. Refocus using the coarse adjustment knob if necessary.
8. Move the slide using the stage control knobs to center your specimen.
9. Use the fine adjustment knob to get the specimen in perfect focus.
10. Your slide **MUST** be focused on low power before attempting the next step.
11. When moving from the low-power to the high-power objective, you must examine your **SPACING** carefully before moving it into place. Sometimes, your stage will be too high to safely move the high-power lens into place. If you do not check your spacing, you may end up breaking the slide (**NOT GOOD**).
12. Click the nosepiece to the high-power (40x) objective.
13. Do **NOT** use the coarse adjustment knob!
14. **ONLY** use the fine adjustment knob to bring the slide into focus and recenter if needed.
15. Do **NOT** use the oil immersion (100x) objective when viewing anything except bacteria. Below, we will learn how to view bacteria using the oil immersion lens.

Exercise 1: Total Magnification

Directions: Fill in Table 3.2.1 below:

Table 3.2.1: Total Magnification

Name of Lens	Ocular Magnification	Objective Magnification	Total Magnification
Scanning Power			
Low Power			
High Power			
Oil Immersion			

Exercise 2: Calculating Field-of-View (FOV) at 4x, 10x, and 40x

1. The diameter of the field-of-view can be measured using a transparent millimeter ruler on the stage for the scanning and low-power objectives.

2. Start by measuring the FOV in millimeters using the scanning power/4x objective and a clear plastic metric ruler. Next, convert the measurement to micrometers (mm) by multiplying the number of millimeters by 1,000. Record this number in Table 3.2 below.
3. Calculate FOV for the 10x and 40x objectives using the following formula:

$$\text{Unknown FOV} = \text{Known FOV diameter} \times (\text{Total Magnification of Known FOV} \div \text{Total Magnification of Unknown FOV})$$

4. Record FOV diameters for 10x and 40x objectives in Table 3.2.1

? Observations

Table 3.2.1: Field of View

Lens	FOV Diameter (mm)
4x	
10x	
40x	

Exercise 3: Estimating the Size of a Specimen

Remember! All microscope observations begin with the scanning (4x) objective. Three reasons for beginning with the scanning objective are:

1. The lower the power, the easier to focus.
2. The lower the power, the greater the field of view (FOV).
3. Once the object has been found at scanning power with the coarse adjustment knob, there should be no need to use the coarse adjustment knob again. Even after switching to another objective, all further focusing is done with the fine adjustment knob.

For this exercise, you will estimate the size of two specimens using prepared slides. Follow the steps below.

1. Determine the number of cells/specimens that would fit across the diameter of the field of view.
2. Estimate the size of the cell/specimen by dividing the diameter of the field by the number of specimens that would fit across the field.

✓ Example

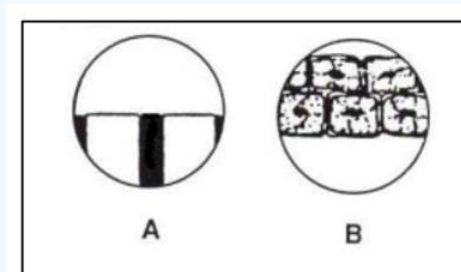
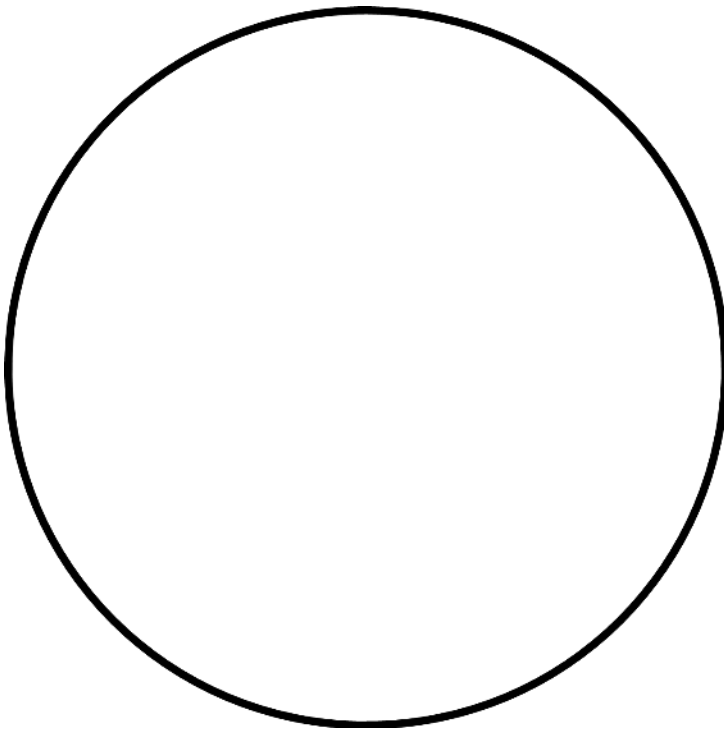


Figure 3.2.2: Example of images from a microscope.

- The field of view in Image A (above) is 2 mm or 2000 μm .
- There are three cells that fit across the 2 mm/2,000- μm field (Image B).
- $2,000 \div 3 \text{ cells} = 666 \mu\text{m}$.
- Each cell is $\sim 660 \mu\text{m}$.

Viewing Specimens/Prepared Slides:

In the circles provided in this chapter, you will make sketches of each specimen/slide.



Genus and Species Name: _____

Objective Lens Used: _____

Estimated Size of Specimen: _____

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3.3: Examining Prokaryotic Cells

Exercise 4: Examining Prokaryotic Cells

By far the most abundant organisms on our planet are the single-celled prokaryotes known as Bacteria and Archaea. You will observe three species of bacteria: a round (coccus) shape, a rod (bacillus) shape, and a spiral (spirochaete) shape. Since bacteria are much smaller than eukaryotic cells, you will need to observe them at 1000x under the oil immersion lens to see them clearly.

How to Use the Oil Immersion Lens:

The most powerful lens of the light microscope is the 100x oil immersion objective. Immersion oil has been formulated so that it has a refractive index identical to that of glass. Thus, there is no refraction of light when it passes from glass to oil and vice versa. Thus, two changes in refractive index can be eliminated by placing a drop of immersion oil on the specimen and immersing the 100x oil immersion objective directly into the drop.

Illustrate the Four-Stage Process of Using the Oil Immersion Lens:

1. Focus very carefully with the 40x objective over the stained specimen on the slide (**once focused, do not alter focus for the next three steps!**).
2. Rotate the nosepiece halfway so that the 40x and 100x objectives straddle the specimen.
3. Apply a small drop of oil directly onto the slide over the specimen.
4. Rotate 100x objective into the immersion oil.

Note

Three important rules for using the oil immersion lens:

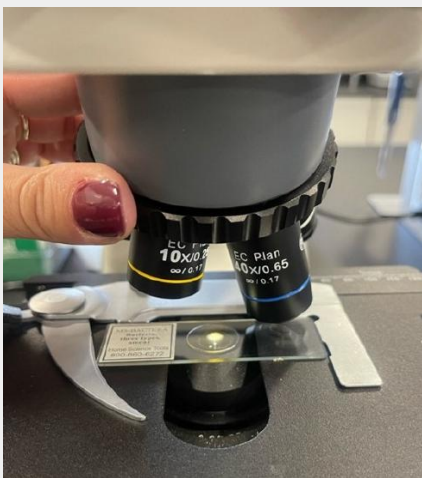
1. Never use an oil immersion lens without oil.
2. Never get oil on any other lens.
3. Clean up all oil when finished.

Protocol



Step 1

Focus at scanning power and then low power on a region of a smeared and stained specimen that is well-spread and stained (not too thin, nor too thick).



Step 2

Rotate the nosepiece to 40x objective, and locate desired portion of the specimen in the center of the field. Refocus very carefully so that the specimen is focused as sharply as possible.

Caution

Do not alter focus for the following steps!



CRITICAL STEP

STEP 3

1. Partially rotate the nosepiece so that the 40x and 100x objectives straddle the specimen.
2. Place a small drop of oil on the slide in the center of the lighted area (take care not to drip oil on the stage).
3. Note the small drop of oil directly over the area of the specimen to be examined.



STEP 4

Rotate the nosepiece so that the 100x oil immersion objective touches the oil and clicks into place (only use fine focus adjustment at this point).

STEP 5

If you are viewing multiple specimens on a single slide, do not alter focusing. Rather, place a drop of oil on the second specimen and use the stage control knobs until the second specimen is in place.

STEP 6

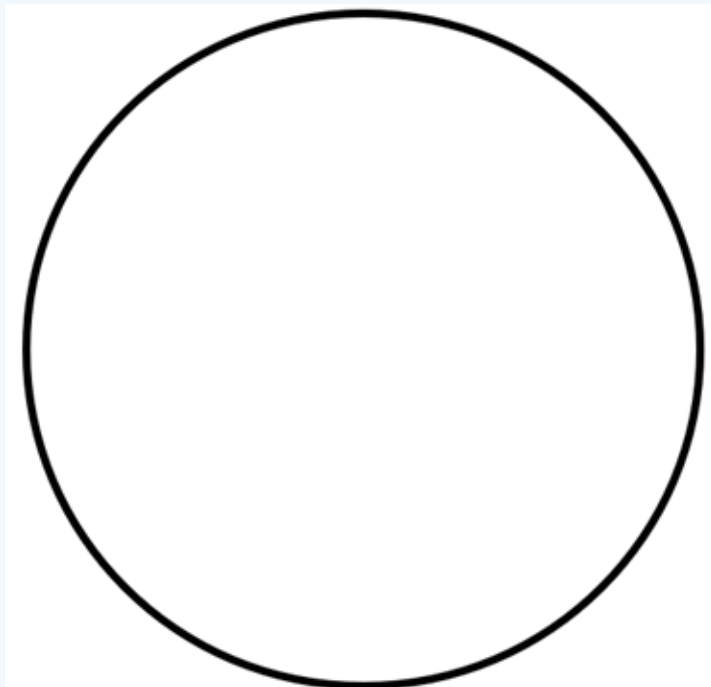
Never go back to the 10x or 40x objectives after you have applied oil to the specimen since oil can ruin the lower power objectives (the 4x objective can be used because it is high enough to be above the oil).

When you have finished, wipe the 100x oil immersion objective carefully with lens paper to remove all oil. Wipe oil from the slide. Clean the stage if there is any oil on it. Recap the immersion oil container securely and store it away.

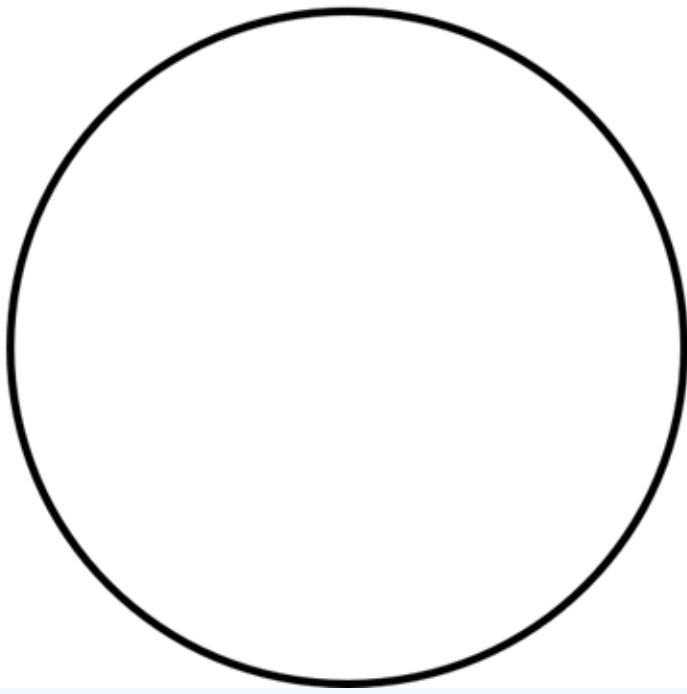
? Observations

Sketch what you see using the oil immersion objective:

Genus and Species Name: _____



Genus and Species Name: _____



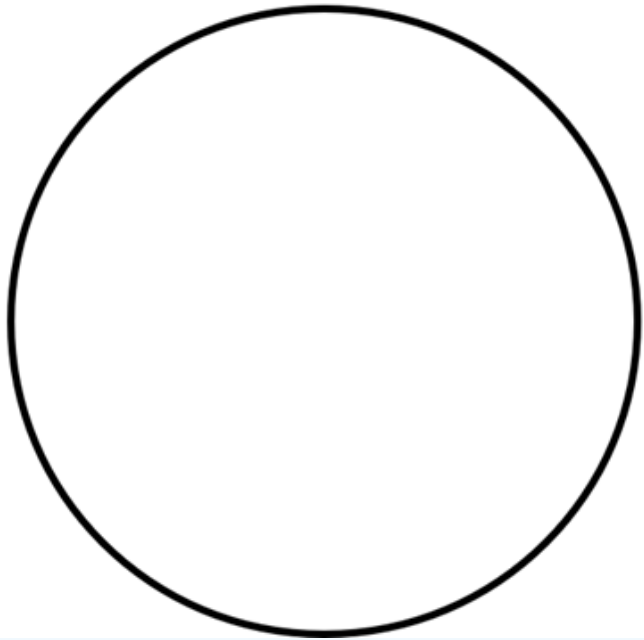
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3.4: Examining Eukaryotic Cells

Exercise 5: Examining Eukaryotic Cells

For the next three specimens, you will be viewing either specimens of your choice or specimens determined by your instructor. Don't forget to record the scientific name and lens you used to make your sketches.

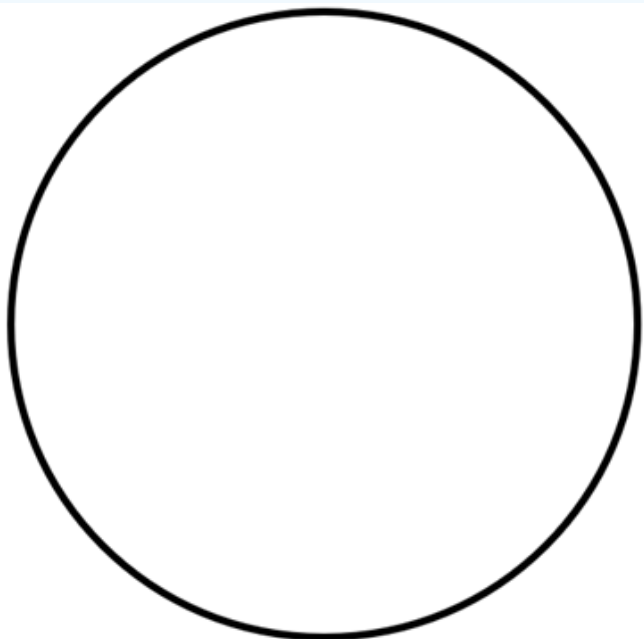
? Observations



Genus and species name: _____

Objective lens used: _____

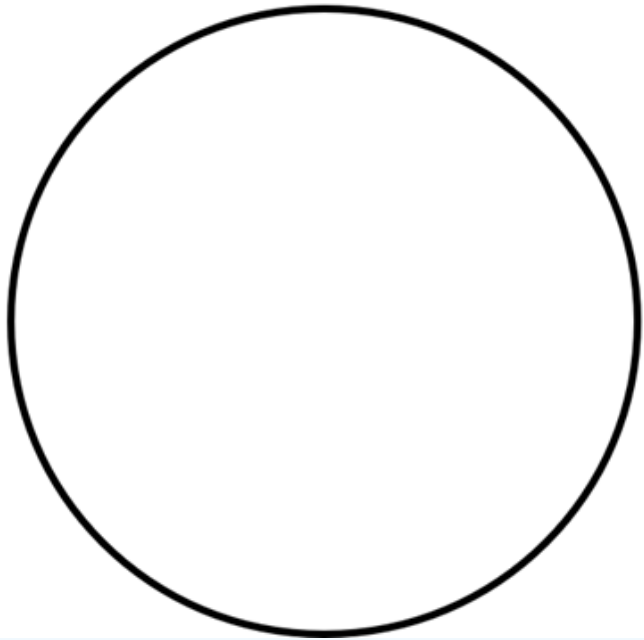
Estimated size of specimen: _____



Genus and species name: _____

Objective lens used: _____

Estimated size of specimen: _____



Genus and species name: _____

Objective lens used: _____

Estimated size of specimen: _____

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3.5: Examining Live Eukaryotic Cells

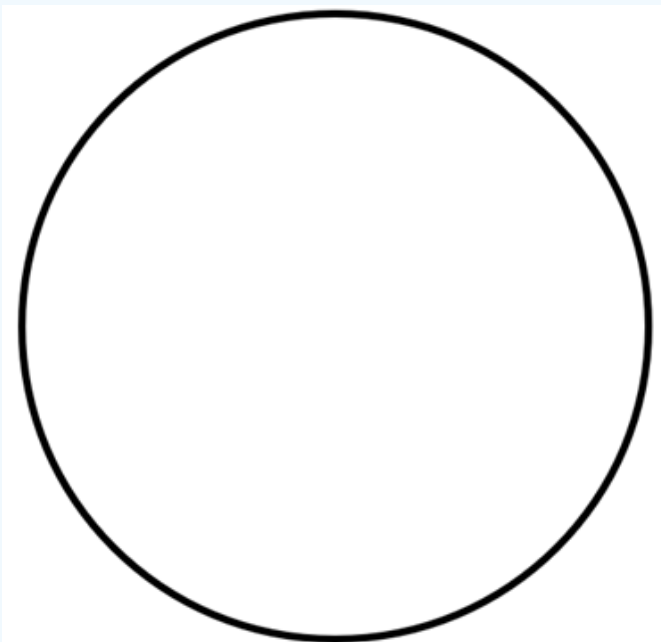
Exercise 6: Examining Live Eukaryotic Cells

For this next exercise, you will examine live organisms. You will need to learn how to prepare a wet mount to complete this. Your instructor will demonstrate first and then you will follow the below instructions to try on your own.

Making a wet mount slide:

1. Obtain a clean slide and coverslip.
2. Place the specimen in the center of the slide.
3. Place the edge of the coverslip on one edge of the water drop.
4. Slowly lower the coverslip on top of the specimen at a 45° angle.
5. View the slide on the stage first with the scanning objective. Once you see the image, you can rotate the nosepiece to view the slide with different objectives. You might have to refocus the image for each objective.

? Observations



Genus and species name: _____

Objective lens used: _____

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3.6: Laboratory 3 Notes Page

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CHAPTER OVERVIEW

Laboratory 4: Cellular Biology and Membrane Transport

- [4.1: Laboratory 4 Introduction](#)
- [4.2: Organelle Functions](#)
- [4.3: Cell Models](#)
- [4.4: Cellular Microscopy](#)
- [4.5: Comparing and Contrasting Cell Types](#)
- [4.6: Simple Diffusion](#)
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4.1: Laboratory 4 Introduction

Cellular Biology and Membrane Transport

Introduction:

In this lab, you'll examine various cell types and learn about their internal structures. A prokaryotic cell is the simplest cell. They contain a nucleoid region, a plasma membrane, cytoplasm, ribosomes, and plasmids.

Eukaryotic cells are far more complex. Animal cells contain a nucleus, ribosomes, an endoplasmic reticulum (smooth and rough), Golgi apparatus, lysosomes, mitochondria, a cytoskeleton, centrioles, and a dense cytoplasm. Plant cells have the same organelles as animals, with a few differences. They do not have lysosomes or centrioles. Additional organelles of plant cells include a cell wall, chloroplasts, central vacuoles, and plasmodesmata.

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4.2: Organelle Functions

Part 1: Organelle Functions

Using the resource of your choice, write down the functions of each of these organelles:

Nucleus: _____

Ribosome: _____

Endoplasmic reticulum (rough): _____

Endoplasmic reticulum (smooth): _____

Plasma membrane: _____

Lysosome: _____

Golgi apparatus: _____

Mitochondria: _____

Chloroplast: _____

Centrioles: _____

Central vacuole: _____

Cytoskeleton: _____

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4.3: Cell Models

View each of the models available to you and sketch what you see.

[Part 2A: Bacterial Cell Model](#)

Draw the bacterial model here. It should take up the vast majority of the space. Show a lot of detail, use color, and add labels.

[Part 2B: Plant Cell Model](#)

Draw the plant cell model here. It should take up the vast majority of the space. Show a lot of detail, use color, and add labels.

[Part 2C: Animal Cell Model](#)

Draw the animal cell model here. It should take up the vast majority of the space. Show a lot of detail, use color, and add labels.

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4.4: Cellular Microscopy

Part 3: Cellular Microscopy

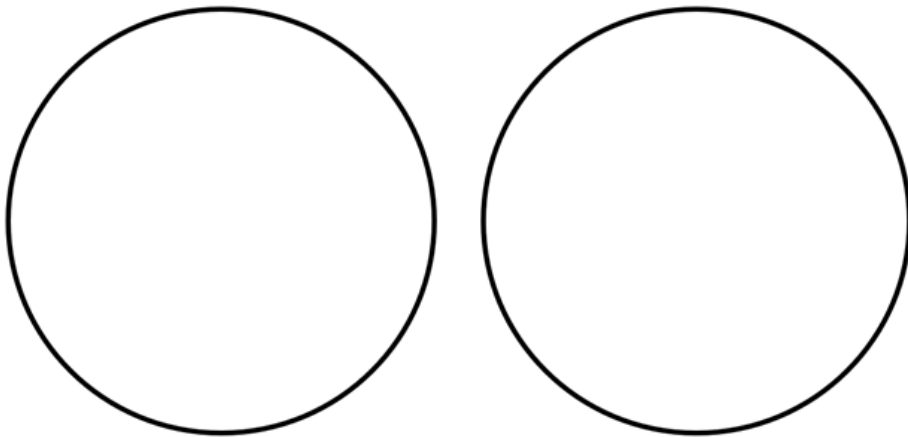
Viewing prepared slides under the microscope:

Directions:

Retrieve a microscope. You will be looking at some generalized cells and reproducing the images you see. You'll notice, especially having drawn the models, that you can't see that much in the prepared slides. **This is completely normal.** We are using a light microscope - the resolution isn't clear enough to see the tiny organelles and they may not have picked up the stains that allow us to see them. Draw a bacterial slide, an animal slide, and a plant slide. Be sure to answer the associated questions with each slide.

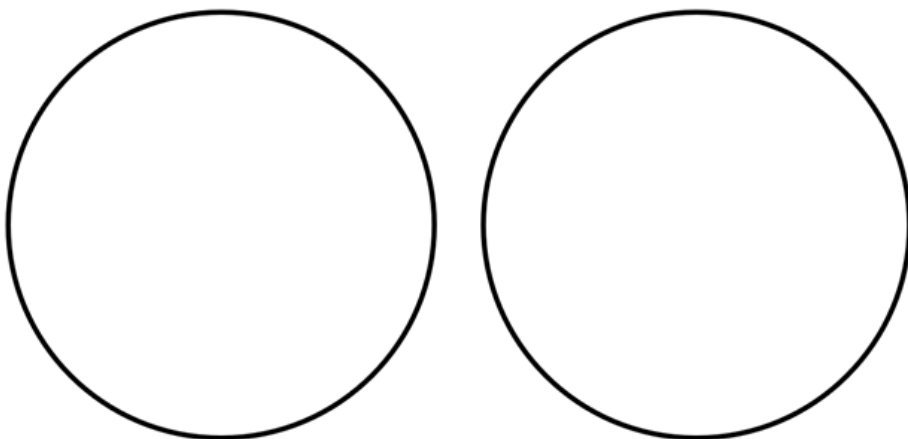
Part 3A:

Obtain a bacterial slide that says it's a mixture of different shapes of bacteria and draw two different magnifications of that slide here. Bacteria are *very* small. At 40x magnification, your slide will most likely appear like a haze of color. Close up, it will appear like a lot of dots. Each dot or strand is an individual bacterial cell. **Label everything that you can** (and note the magnification).



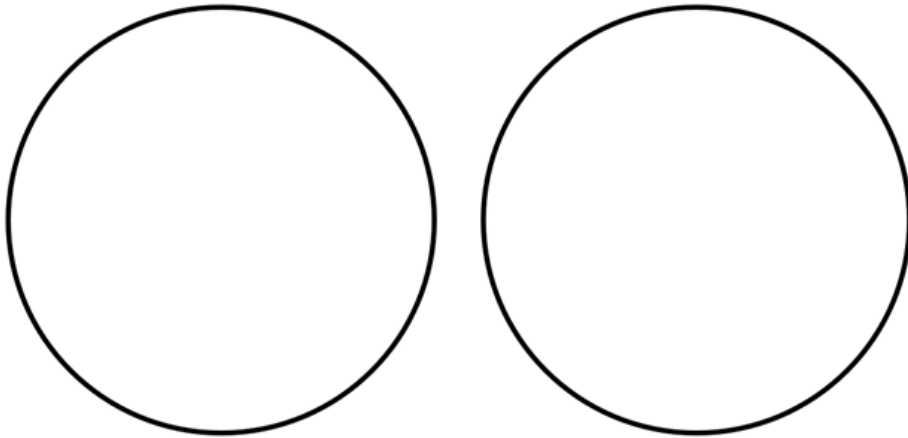
Part 3B:

Obtain a slide that contains typical animal cells and view it at two different magnifications. Your slide is going to contain hundreds, if not thousands, of cells. Look for the defined boundaries of the cell – these are most likely the plasma membranes. The solid, smaller, darkly staining material contained within the cells is likely the nucleus. **Label everything that you can** (and note the magnification).



Part 3C:

Obtain a slide that contains typical plant cells and draw two different magnifications. Like the animal cell slide, it will contain hundreds to thousands of cells. The box-like shapes are the outlines of the cell walls! **Label everything you can** (and note the features that did not exist in the animal cell).



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4.5: Comparing and Contrasting Cell Types

Part 4. Compare and Contrast Cell Types

List the organelles that are found in animal cells only, plant cells only, and those that are found in all three (animals, plants, and bacteria).

Organelles found only in animal cells	Organelles found only in plant cells	Organelles found in animals, plants, and bacteria

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4.6: Simple Diffusion

Part 5: Simple Diffusion

Lab Materials

- Three 250-mL beakers
- Food coloring (dark colors work best)
- Tap water
- Timer
- Hot plate
- Thermometer

Question:

How does temperature affect the diffusion of food coloring?

Hypothesis:

Procedure:

1. Place 200 mL of tap water in a 250-mL beaker and heat with a hot plate until the temperature is approximately 60 degrees Celsius. Label beaker “hot”.
2. In a beaker, make cold water by adding ice to tap water. Place 200 mL of cold water in a 250-mL beaker (this beaker should not have ice in it). Label beaker “cold”.
3. Place 200 mL of room temperature water in a 250-mL beaker. Label beaker “room temp”.
4. Add two drops of food coloring to each beaker, and time how long it takes for the dye in each beaker to diffuse.

Observations

Beaker	Diffusion Time
Hot	
Cold	
Room Temperature	

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4.7: Diffusion Across a Semipermeable Membrane

Part 6: Diffusion Across a Semipermeable Membrane

Because of its structure, the cell membrane is a semipermeable membrane. This means that **SOME** substances can easily diffuse through it (i.e., oxygen and carbon dioxide). Other substances, such as glucose or sodium ions, are unable to pass through the cell membrane unless they are specifically transported via proteins embedded in the membrane itself. Whether or not a substance is able to diffuse through a cell membrane depends on the characteristics of the substance and the characteristics of the membrane. In this exercise, we will make dialysis tubing “cells” and explore the effect of size on a molecule’s ability to diffuse through a “cell membrane”.

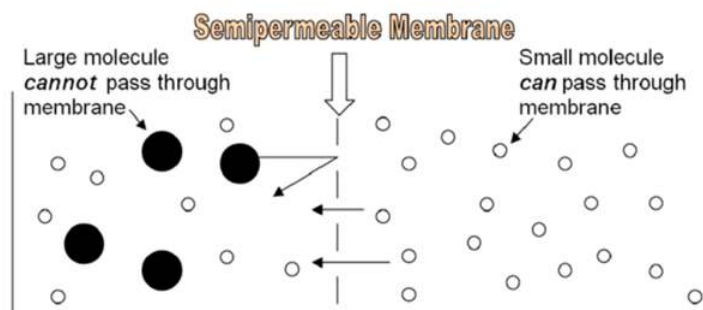


Figure 4.7.1: Illustration of a semipermeable membrane.

The following information might be useful in understanding and interpreting your results in this lab:

- **Phenolphthalein**
 - Atomic formula: $C_{20}H_{14}O_4$
 - Atomic mass: 318.32 g/mol
 - Color of acidic solution: Clear
 - Color of basic solution: Pink
- **Iodine**
 - Atomic formula: I or I_2
 - Atomic mass: 126 g/mol
- **Starch**
 - Atomic formula: $(C_6H_{10}O_5)_n$
 - Atomic mass: HUGE!
 - Color in iodine: Blue-ish
- **Sodium Hydroxide**
 - Atomic formula: NaOH
 - Atomic mass: 40.1 g/mol
 - Acid or base? Base

Lab Materials

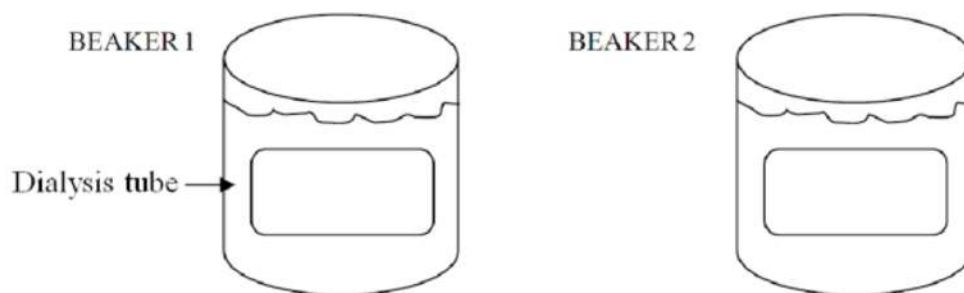
Materials:

- Two pieces of dialysis tubing
- Thread
- Phenolphthalein
- Iodine
- Wax pencil
- Two beakers

- NaOH
- Starch solution
- Pipettor

Procedure:

1. Using a wax pencil, label one beaker “#1” and the other beaker “#2”.
2. Fill beaker #1 with 300 mL of tap water, then add 10 drops of 1 M NaOH. Do not spill the NaOH – it is very caustic!
3. Fill beaker #2 with 300 mL of tap water, then add iodine drops drop by drop until the solution is bright yellow.
4. Now prepare your two dialysis tubing “bags”. Seal one end of each dialysis tube by carefully folding the end “hotdog” style two times and then “hamburger” style one time. Tie the folded portion of the tube securely with string. It is critical that your tubing is tightly sealed to prevent leaks.
5. Add 10 mL of water and three drops of phenolphthalein to one of your dialysis tube bags. Seal the other end of the bag by carefully folding and tying as before.
6. Thoroughly rinse the bag containing phenolphthalein, then place it into the beaker containing NaOH.
7. Add 10 mL of starch solution to the other dialysis tube. Again, seal the bag tightly and rinse as above. Place this bag containing the starch solution into beaker #2.
8. Let diffusion occur between the bags and the solutions in the beakers.
9. After 10 minutes, observe the color changes in the two bags and the external solutions. Draw a picture of each system below:



Record the colors (below) and label contents inside and outside the bags (above):

Colors	Beaker #1 Initial	Beaker #1 Final	Beaker #2 Initial	Beaker #2 Final
Color Inside Bag				
Color Outside Bag (in Beaker)				

? Questions

1. Which substance diffused across the membrane in Beaker #1? How do you know?
2. Which substance diffused across the membrane in Beaker #2? How do you know?
3. Why might some ions and molecules pass through the dialysis bag while others might not?

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4.8: Red Onion Osmosis Lab

Part 7: Red Onion Osmosis Lab

Lab Information and Materials

- Maintaining a controlled internal environment is very important to the health of every cell. The concentration of solutes must be tightly controlled. This is especially important in animal cells that don't have cell walls to help give them structure.
- When the two concentrations (inside and outside the cell) are equal, the system is considered **isotonic**, and no water is gained or lost by the cell.
- When the concentration of solutes in the solution around the cell is higher, it is called **hypertonic**. The system will attempt to equalize – this means that, in a hypertonic solution, the water will leave the cell in an attempt to increase the concentration. Since animal cells lack cell walls, they will collapse in on themselves. When plant cells are placed in a hypertonic solution, the central vacuole loses water, causing the cytoplasm to shrink away from the cell wall (called **plasmolysis**).
- When the concentration of solutes outside of the cell is lower, it is called **hypotonic**. The system attempts to equalize – this means that, in a hypotonic solution, water will enter the cell in an attempt to decrease the concentration. Since animal cells lack cell walls, they can burst/lyse in a hypotonic solution. In plant cells, the central vacuole gains water, causing the cytoplasm to bulge out against the cell wall (called **turgor pressure**).

Materials:

- Microscope
- Slide and slipcover
- Onion
- 15% NaCl solution
- Distilled water
- Pipette
- Paper towel/Kim wipe

Procedure:

1. Remove a small section of tissue from the epidermis (skin) of the onion on the purple side. Once removed, the epidermis is thin and almost transparent.
2. Prepare a wet mount placing the epidermis on a glass slide and adding 1-2 drops of distilled water (hypotonic solution).
3. Place coverslip at a 45-degree angle.
4. Make observations of the onion cells using the 4x, 10x, and 40x objectives.
5. Make a sketch (next page) of the onion cells in a hypotonic solution.
6. Next, take a small piece of paper towel (or a kim wipe) and absorb water from the slide on one side of the coverslip). Then, add 15% NaCl (hypertonic) solution with a pipette to the other side.
7. Wait 2-5 minutes.
8. Make observations of the onion cells using the 4x, 10x, and 40x objectives.
9. Make a sketch of onion cells in a hypertonic solution.

Onion Cells in Hypotonic Solution (Distilled Water) at Scanning, Low-Power, and High-Power:

Question: Why do the cells appear this way in a hypotonic solution?

Onion Cells in Hypertonic Solution (15% NaCl solution) at Scanning, Low-Power, and High-Power:

Question: Why do the cells appear this way in a hypertonic solution?

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4.9: Laboratory 4 Notes Page

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CHAPTER OVERVIEW

Laboratory 5: Heart Rate of Daphnia Upon Exposure to Ethanol and Caffeine

[5.1: Laboratory 5 Introduction](#)

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5.1: Laboratory 5 Introduction

Laboratory 5: Heart Rate of *Daphnia* Upon Exposure to Ethanol and Caffeine

Introduction:

Daphnia magna, commonly known as water fleas, are freshwater arthropods with a clear external skeleton (carapace) and jointed appendages. The heart rate (heartbeats per minute) of *Daphnia* has previously been shown to be sensitive to various chemical exposures. In this experiment, you will examine the heart rate of *Daphnia* upon ethanol and caffeine exposure. In humans, caffeine is a well-known stimulant that increases heart rate, while ethanol is a depressant that decreases heart rate.



Figure 5.1.1: *Daphnia magna*.

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5.2: Daphnia Experiment

Lab Objectives and Materials

- The purpose of this study is to determine if the heart rate in *D. magna* behaves similarly to the heart rate of humans when exposed to caffeine and ethanol.
- Please work in a group of four for this experiment. Two members of the group should measure the effects of caffeine on heart rate, while the other two members should measure the effects of ethanol on heart rate. Share your data with your group members.

Materials:

- *Daphnia* in culture liquid
- Transfer pipettes
- Depression slides
- Compound microscope or Stereoscope
- Small container for “used” *Daphnia*
- 1%, 2% and 4% ethanol in a dropper bottle
- 0.5%, 1% and 1.5% caffeine in a dropper bottle

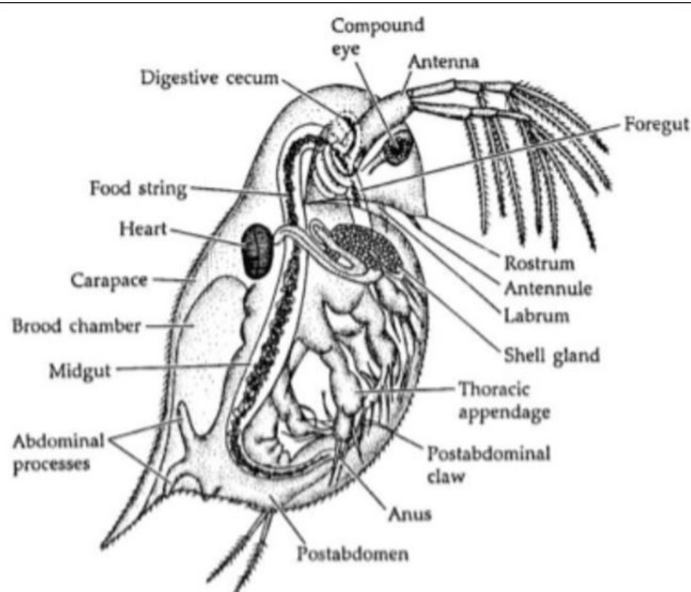


Figure 5.2.1: Labeled parts of *Daphnia*.

Experimental Procedure:

1. Using a clean pipette, carefully transfer a *Daphnia* onto a depression slide.
2. Place the slide under the microscope and focus on the *Daphnia* so that you can see the beating heart.
3. Have your lab partner time 20 seconds for you as you count the number of heartbeats during that 20-second interval. You want to make your measurements quickly, so that the *Daphnia* does not become stressed in the small volume of water and under the heat of the microscope.
4. Record the number of heartbeats in the data table on the next page. Multiply the number by 3 to get the number of beats per minute (bpm).
5. Take at least three separate heart rate measurements for each individual *Daphnia* and calculate the average of the three measurements.
6. When you have finished recording the heart rate in water (the CONTROL solution), remove as much water as you can from the depression slide using a transfer pipette. Then add ONE DROP of the 1% ethanol solution to the slide. Turn the light OFF and

wait 30 seconds.

7. Turn the light back on and count the number of heartbeats for 20 seconds again, repeating at least 3 times. Multiply each count by 3 to get the heart rate per minute. Record in the data table.
8. Place the *Daphnia* into the “used” container, clear off the depression slide, then repeat steps # 1-9 with a new *Daphnia* two more times.
9. Repeat the above steps # 1-9 using the caffeine solutions.
10. You will use three different *Daphnia* for each of the three ethanol solutions and you will use three different *Daphnia* for each of the three caffeine solutions. In other words, a total of 9 *Daphnia* will be used to look at the effects of ethanol on heart rate in *Daphnia*, and a total of 9 *Daphnia* will be used to look at the effects of caffeine on heart rate in *Daphnia*.

? Results and Data Collection

Table 5.2.1: Effect of 1% Ethanol on Heartrate

Daphnia	Water		1% Ethanol	
<i>Daphnia 1</i>	20 seconds	Beats/min. (20 sec x 3)	20 seconds	Beats/min. (20 sec x 3)
Count 1				
Count 2				
Count 3				
<i>Daphnia 2</i>				
Count 1				
Count 2				
Count 3				
<i>Daphnia 3</i>				
Count 1				
Count 2				
Count 3				
Mean	XXXXXXXX		XXXXXXXX	

Table 5.2.2: Effect of 2% Ethanol on Heartrate

Daphnia	Water		2% Ethanol	
Daphnia 1	20 seconds	Beats/min. (20 sec x 3)	20 seconds	Beats/min. (20 sec x 3)
Count 1				
Count 2				
Count 3				
Daphnia 2				
Count 1				
Count 2				
Count 3				
Daphnia 3				
Count 1				
Count 2				
Count 3				
Mean	XXXXXXXX		XXXXXXXX	

Table 5.2.3: Effect of 4% Ethanol on Heartrate

Daphnia	Water		4% Ethanol	
Daphnia 1	20 seconds	Beats/min. (20 sec x 3)	20 seconds	Beats/min. (20 sec x 3)
Count 1				
Count 2				
Count 3				
Daphnia 2				
Count 1				
Count 2				
Count 3				
Daphnia 3				
Count 1				
Count 2				
Count 3				
Mean	XXXXXXXX		XXXXXXXX	

Table 5.2.4: Effect of 0.5% Caffeine on Heartrate

Daphnia	Water		0.5% Caffeine	
Daphnia 1	20 seconds	Beats/min. (20 sec x 3)	20 seconds	Beats/min. (20 sec x 3)
Count 1				
Count 2				
Count 3				
Daphnia 2				
Count 1				
Count 2				
Count 3				
Daphnia 3				
Count 1				
Count 2				
Count 3				
Mean	XXXXXXXX		XXXXXXXX	

Table 5.2.5: Effect of 1% Caffeine on Heartrate

Daphnia	Water		1% Caffeine	
Daphnia 1	20 seconds	Beats/min. (20 sec x 3)	20 seconds	Beats/min. (20 sec x 3)
Count 1				
Count 2				
Count 3				
Daphnia 2				
Count 1				
Count 2				
Count 3				
Daphnia 3				
Count 1				
Count 2				
Count 3				
Mean	XXXXXXXX		XXXXXXXX	

Table 5.2.6: Effect of 1.5% Caffeine on Heartrate

Daphnia	Water		1.5% Caffeine	
Daphnia 1	20 seconds	Beats/min. (20 sec x 3)	20 seconds	Beats/min. (20 sec x 3)
Count 1				
Count 2				
Count 3				
Daphnia 2				
Count 1				
Count 2				
Count 3				
Daphnia 3				
Count 1				
Count 2				
Count 3				
Mean	XXXXXXXX		XXXXXXXX	

Data Analysis

Graph these data in your lab manual (below, Figure 1). Note that the dependent variable should be on the Y-axis and the independent variable on the X-axis. Include a legend or key and an informative caption below the graph, including the sample size for each treatment.

Figure 1. Average Heart Rate of Daphnia

Interpretation

Based on your data, what are the effects of ethanol and caffeine on heart rate in *Daphnia*? Explain your conclusions. What do you think would happen if you left *Daphnia* in each solution over a period of 30 minutes or several hours?

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CHAPTER OVERVIEW

Laboratory 6: Mitosis and Cytokinesis

- [6.1: Laboratory 6 Introduction](#)
- [6.2: Mitosis of Onion Root Rip](#)
- [6.3: Mitosis of Whitefish Blastula](#)
- [6.4: Simulating Mitosis Using Beads](#)
- [6.5: Laboratory 6 Notes](#)

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6.1: Laboratory 6 Introduction

Laboratory 6: Mitosis and Cytokinesis Introduction

All cells come from preexisting cells, and eukaryotic cells must undergo mitosis in order to form new cells. The replication of a cell is part of the overall cell cycle (Figure 6.1.1), which is composed of interphase and M phase (mitotic phase). The M phase, which consists of mitosis and cytokinesis, is the portion of the cell cycle where the cell divides, reproducing itself. Mitosis is the division of the nucleus and its contents.

In mitosis, DNA that has been copied in the S phase of interphase is separated into two individual copies. Each copy will end up in its own cell at the end of M phase.

Mitosis has several steps: prophase, prometaphase, metaphase, anaphase, and telophase (Figure 6.1.2). The spindle fibers, which are formed by the cell as mitosis progresses, are used to attach to chromosomes, align them down the middle of the cell, and pull chromosomes apart into their identical individual chromatids which will end up in separate cells. As mitosis is nearing its end and the cell is in telophase, the cytoplasm also divides so that both new cells will have their own fluid, organelles, etc. This division of the cytoplasm is called cytokinesis. Mitosis and cytokinesis can be viewed under a microscope.

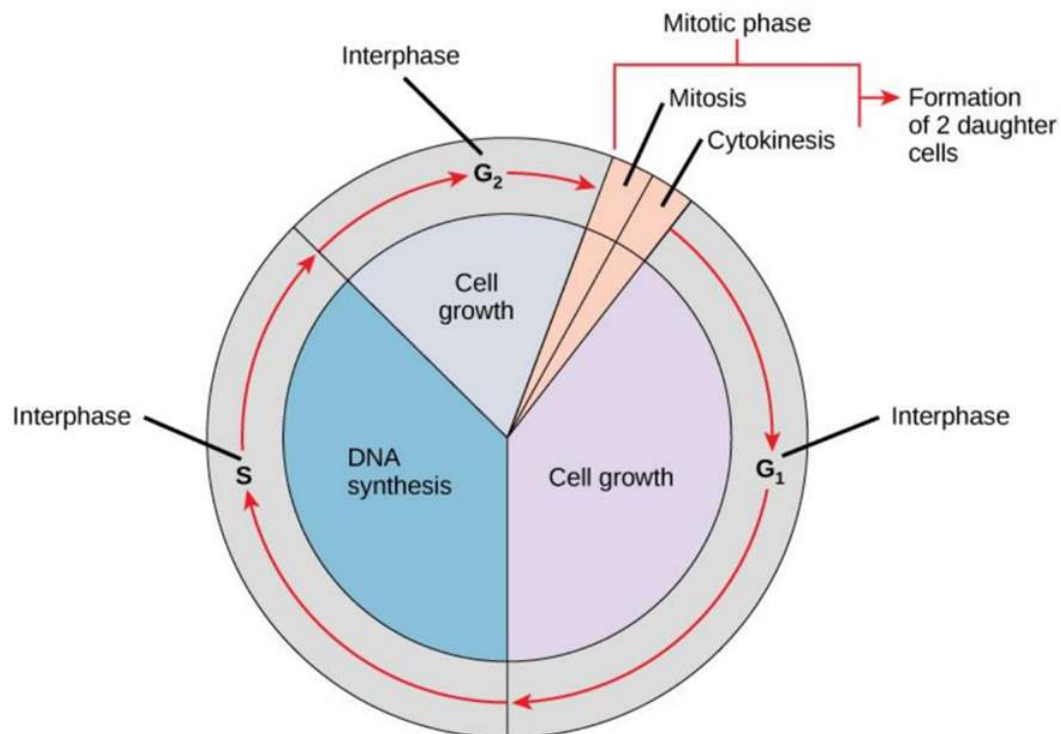


Figure 1- The cell cycle. *Biology. OpenStax College.*

Figure 6.1.1: The cell cycle. *Biology. OpenStax College.*

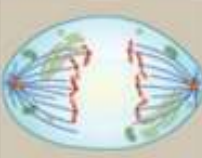
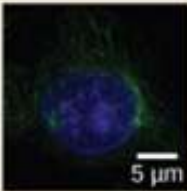
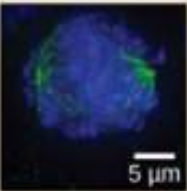
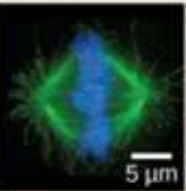
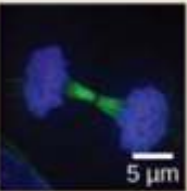
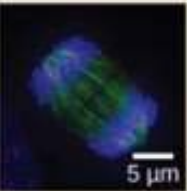
Prophase	Prometaphase	Metaphase	Anaphase	Telophase	Cytokinesis
					
- Chromosomes condense and become visible - Spindle fibers emerge from the centrosomes - Nuclear envelope breaks down - Nucleolus disappears	- Chromosomes continue to condense - Kinetochores appear at the centromeres - Mitotic spindle microtubules attach to kinetochores - Centrosomes move toward opposite poles	- Mitotic spindle is fully developed, centrosomes are at opposite poles of the cell - Chromosomes are lined up at the metaphase plate - Each sister chromatid is attached to a spindle fiber originating from opposite poles	- Cohesin proteins binding the sister chromatids together break down - Sister chromatids (now called chromosomes) are pulled toward opposite poles - Non-kinetochore spindle fibers lengthen, elongating the cell	- Chromosomes arrive at opposite poles and begin to decondense - Nuclear envelope material surrounds each set of chromosomes - The mitotic spindle breaks down	- Animal cells: a cleavage furrow separates the daughter cells - Plant cells: a cell plate separates the daughter cells
					

Figure 2- Stages of M phase. *Biology. OpenStax College.*

Figure 6.1.2: Stages of M phase. Biology. OpenStax College.

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6.2: Mitosis of Onion Root Rip

Exercise 1: Mitosis of Onion Root Rip

(Adapted from Cell Biology Laboratory Manual Online Dr. William H. Heidcamp, Biology Department, Gustavus Adolphus College, St. Peter, MN 56082)

Lab Materials

Materials

- Prepared slide of onion (*Allium*) root tip
- Microscope

Procedure

1. Obtain a slide of *Allium* root tip for observation of the stages of mitosis in a plant cell.
2. Examine the slide under a microscope.
3. Draw and label all stages of mitosis below. Figure 6.2.1 can be used to help with this.

Interphase Prophase Metaphase

Anaphase Telophase and Cytokinesis

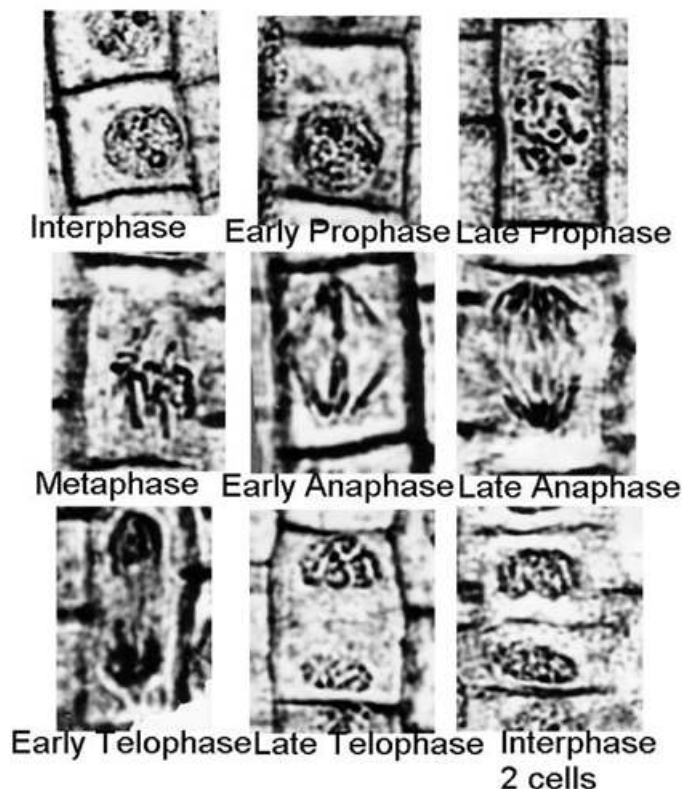


Figure 3- Onion root tip mitosis. Dr. William H. Heidcamp, Biology Department, Gustavus Adolphus College, St. Peter, MN 56082 -- cellab@gac.edu).

Figure 6.2.1: Onion root tip mitosis. Dr. William H. Heidcamp, Biology Department, Gustavus Adolphus College.

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6.3: Mitosis of Whitefish Blastula

Exercise 2: Mitosis of Whitefish Blastula

(Adapted from Cell Biology Laboratory Manual Online Dr. William H. Heidcamp, Biology Department, Gustavus Adolphus College, St. Peter, MN 56082)

Lab Materials

Materials

- Prepared slide of whitefish blastula
- Microscope

Procedure

1. Obtain a slide of a whitefish blastula for observation of the stages of mitosis in an animal cell. Since early embryogenesis involves rapid cellular division, the whitefish blastula has long served as a model of mitotic division in animals. It also has the advantage of demonstrating clear spindle formation in the cytoplasm.
2. Examine the slide under a microscope.
3. Draw and label all stages of mitosis below.

Interphase Prophase Metaphase

Anaphase Telophase and Cytokinesis

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6.4: Simulating Mitosis Using Beads

Exercise 3: Simulating Mitosis Using Beads

Lab Materials

Materials:

- Baggy full of beads and strings

Procedure:

1. Organize beads into “chromosomes” as shown in Figure 6.4.1.
2. Simulate the steps of interphase (specifically S phase) and then M phase using the beads. Hint: The chromosomes in Figure 6.4.1 have not been through the S phase yet, so you will eventually need more beads than are shown in Figure 6.4.1. The strings in the bag are used to simulate spindle fibers.

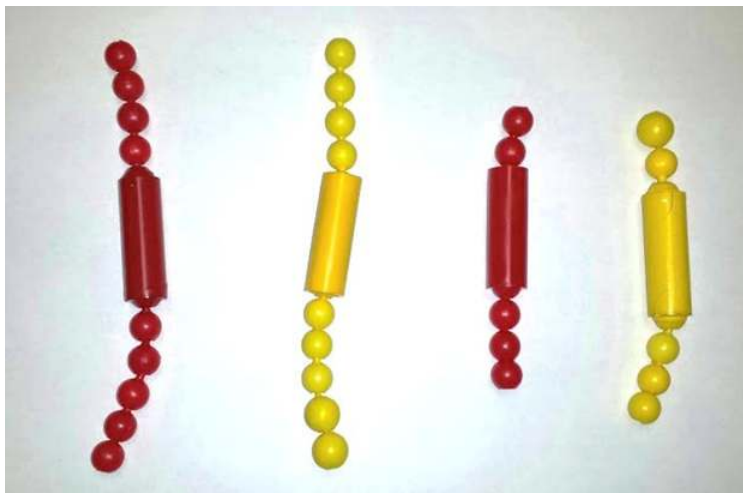


Figure 6.4.1: How to set up bead chromosomes to simulate mitosis.

? Questions

1. Most nerve cells in the adult human central nervous system, as well as heart muscle cells, do not divide. In contrast, cells lining the inside of the small intestine divide frequently. Discuss this difference in terms of why damage to the nervous system and heart muscle cells (think stroke or heart attack) is so dangerous. What do you think might happen to tissues such as the intestinal lining if a disorder blocks mitotic cell division in all cells of the body?
2. How do mitosis and cytokinesis differ?

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6.5: Laboratory 6 Notes

Laboratory 6 Notes

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CHAPTER OVERVIEW

Laboratory 7: Meiosis and Heredity

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[7.2: Modeling the Stages of Meiosis](#)

[7.3: Heredity](#)

[7.4: Human Genetics Problems](#)

[7.5: Laboratory 7 Notes Page](#)

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7.1: Laboratory 7 Introduction

Part 1: (Meiosis) Introduction

Before meiosis, the cell makes a copy of the DNA in each chromosome. Then, during meiosis, there are two cell divisions, Meiosis I and Meiosis II. These two cell divisions produce four haploid daughter cells. Meiosis I is different from mitosis because each pair of homologous chromosomes lines up next to each other and then the two homologous chromosomes separate. Meiosis I produces daughter cells with half as many chromosomes as the parent cell, so the daughter cells are haploid. Each daughter cell has a different chromosome from the original pair of homologous chromosomes.

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7.2: Modeling the Stages of Meiosis

Activity: Modeling the Stages of Meiosis

Lab Materials

Materials

- Chromosome modeling kits
 - Magnetic centromeres (x8)
 - Several red and yellow beads

Procedure:

A diploid cell with two homologous pairs of chromosomes (as in the previous modeling exercise) will be modeled as it moves through meiosis. First, you will model meiosis I. Then, you will model meiosis II as described below.

Modeling Meiosis I:

1. Build two pairs of replicated homologous chromosomes (i.e., chromosomes that have gone through interphase) using the pop beads. Build one large pair of homologous chromosomes (one red, one yellow) and one smaller pair of homologous chromosomes (one red, one yellow).
2. **Prophase I** – First, model synapsis by matching up your pairs of homologous chromosomes to form two tetrads. Next, model crossing-over, which can occur several times along the length of the chromosomes. Use red and yellow beads to represent exchanged segments of chromatids on the inner non-sister chromatids of the tetrad pairs. Place a yellow bead on an inner red (maternal) chromatid to represent DNA exchanged from the paternal chromatid. Place a red bead on an inner yellow (paternal) chromatid to represent DNA from the maternal chromatid. Make a minimum of 1 crossover for each pair of homologous chromosomes.
3. **Metaphase I** – Line up the pairs of homologues in the center of your imaginary cell (pairs align independently).
4. **Anaphase I** – Separate the pairs of homologous chromosomes by moving them toward opposite poles of the cell.
5. **Telophase I and Cytokinesis I** – Completely separate your pairs of homologous chromosomes to represent them moving into two different “cells”. These two cells will enter Meiosis II. Note that each daughter cell has half the number of chromosomes as the parental cell. Thus, the cells have been reduced from **diploid (2n)** to **haploid (n)**. “n” refers to the **number of *pairs* of chromosomes that are characteristic of a species**. Humans have an “n” of 23, so a diploid (2n) human cell has 46 chromosomes, and a haploid (n/1n) human cell has 23 chromosomes.

Modeling Meiosis II

6. There is no DNA replication before the second cell division stage of meiosis. The stages of Meiosis II proceed very much like mitosis. The two cells created in Meiosis I will enter into **Prophase II**. The chromosomes in each cell contain two sister chromatids, which are condensed and distributed throughout the nucleus.
7. **Metaphase II** - Line up the individual chromosomes on the equator (middle) of the cell. Sister chromatids remain attached at the centromere during Metaphase II.
8. **Anaphase II** – Pull the magnets (centromeres) apart that attach the sister chromatids to each other to separate them, then move each chromatid toward opposite poles. After separation at the centromere, the chromatids are now called chromosomes.
9. **Telophase II** begins when chromosomes reach opposite poles of the cell. Nuclear division happens in telophase. Nuclear envelopes and nucleoli reappear. Condensed chromosomes begin to decondense and uncoil. The formation of separate nuclear envelopes divides the nuclei and marks the end of telophase. Cytokinesis II divides the cytoplasm of each cell into two. A total of four cells are present at the end of Cytokinesis II.

? Follow-Up Questions

1. How many chromosomes are in the original parent cell? _____
2. How many chromosomes are in each daughter cell? _____
3. Are the chromosomes in the daughter cells identical to the chromosomes in the original parent cell? _____
4. What is the meaning of the term “diploid”? What abbreviation do we use to represent diploid? Name two diploid cells in humans.
5. What is the meaning of the term “haploid”? What abbreviation do we use to represent haploid? Name two haploid cells in humans.
6. In what stage of the cell cycle does DNA replication occur?
7. What specific feature of cytokinesis can you use to distinguish between plant and animal cell division?

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7.3: Heredity

Part 2: (Heredity) Introduction

In diploid organisms, each body cell (or 'somatic cell') contains two copies of the genome. So, each somatic cell contains two copies of each chromosome and two copies of each gene. The exceptions to this rule are the **sex chromosomes** that determine sex in a given species. For example, in the XY system that is found in most mammals - including human beings - males have one X chromosome and one Y chromosome (XY), and females have two X chromosomes (XX). The paired chromosomes that are not involved in sex determination are called **autosomes**. Human somatic cells have 46 chromosomes: 22 pairs of autosomes and one pair of sex chromosomes (X and Y).

The different forms of a gene that are found at a specific point (or locus) along a given chromosome are known as **alleles**. Diploid organisms have two alleles for each autosomal gene--one inherited from the mother and one inherited from the father.

Mendelian Inheritance Patterns:

Within a population, there may be a number of alleles for a given gene. Individuals with two copies of the same allele are referred to as homozygous for that allele; individuals with copies of different alleles are known as **heterozygous** for that allele. The inheritance patterns observed will depend on whether the allele is found on an autosomal chromosome or a sex chromosome and on whether the allele is **dominant** or **recessive**.

If the phenotype associated with a given version of a gene is observed when an individual has only one copy, the allele is said to be autosomal dominant. The phenotype will be observed whether the individual has one copy of the allele (is heterozygous) or has two copies of the allele (is homozygous). If the phenotype associated with a given version of a gene is observed only when an individual has two copies, the allele is said to be autosomal recessive. The phenotype will be observed only when the individual is homozygous for the allele concerned. An individual with only one copy of the allele will not show the phenotype but will be able to pass the allele on to subsequent generations. As a result, an individual heterozygous for an autosomal recessive allele is known as a **carrier**.

Scientists use a grid-like tool (Punnett Square) to make predictions about various genetic problems. The Punnett Square shows only the **probability** (the chance of something occurring) of what might occur and not the actual results. For example, if one wants to flip a coin 100 times since there are two sides to the coin, one would expect 50 heads and 50 tails. If they flip the coin 100 times, they may actually get 60 heads and 40 tails. The Punnett Square only shows the chances of what might occur each time the event is undertaken.

* It doesn't matter how often you flipped a coin or how many times it's already shown heads; the probability is ALWAYS 50% of heads/tails. For example, if I've tossed a coin 4 times and all four times it came up heads. What is the probability that my next toss will be heads?

Simulating a One-Trait Cross:

Procedure:

1. Each group will pick up two paper bags filled with 15 black (B) beads and 15 white (b) beads. This represents two heterozygous parents ($Rr \times Rr$).
2. At the same time, each student will reach into their bag and pull out one of the beads. The only possibilities that can be made from this selection are: BB (homozygous black), Bb (heterozygous black), or bb (homozygous white). Mark the resulting genotype and phenotype in the data table.
3. Return the beads back into the bag and conduct the same process 14 more times (15 total trials).

Data Table:

Trial	Offspring Genotype	Offspring Phenotype
1		
2		
3		
4		
5		
6		
7		
8		
9		
10		
11		
12		
13		
14		
15		

? Follow-Up Questions

1. What is the dominant trait? How do you know it is dominant?
2. What is the recessive trait?
3. What are the genotypes of the parents?
4. What are the phenotypes of the parents?
5. Fill in the Punnett Square below using the parents given in the procedure:

6. What is the genotypic ratio?
7. What is the phenotypic ratio?

Cystic Fibrosis (CF)

1. In humans, cystic fibrosis is caused by a recessive allele on chromosome 7. A person must inherit two of these alleles to have the disease. What are the chances of a child having the disease if both of the parents are heterozygous?

2. If a person who is heterozygous for cystic fibrosis is married to someone who has the disease, what are the chances that their child will have CF?

Fainting Gene in Goats

1. In goats, a recessive gene causes the goats to “faint” when they are startled. A farmer has a goat, Gertrude, that is a carrier for the fainting gene (Gg). Gertrude is mated with a goat that is homozygous dominant. How many of their offspring will be fainters?

2. Test crosses are performed to determine the genotype of an organism. A farmer buys a goat (Gary) whose owner claims has sired fainting goats, though Gary has never fainted himself. What type of goat (genotype/phenotype) should the farmer mate with Gary to determine if Gary is a carrier of the “fainting” gene?

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7.4: Human Genetics Problems

Definition: Dominant versus Recessive Traits

Dominant = trait that will be expressed in the individual's phenotype when present

Recessive = trait that will be masked in the presence of a dominant allele (will only show up in an individual's phenotype if two recessive alleles are inherited)

Abundance of Dominant vs. Recessive Traits

Directions: In the table below, note which phenotype you possess (dominant or recessive) for each of the traits listed. Gather data from the rest of your class, then calculate the percentages of individuals for each trait.

Trait	Number of Individuals	Percentage of Individuals
Hairline: Widow's Peak (WW or Ww) Straight (w)	Widow's Peak = _____ Straight Hairline = _____	% with dominant trait = _____ % with recessive trait = _____
Earlobes: Unattached (E) Attached (e)	Unattached = _____ Attached = _____	% with dominant trait = _____ % with recessive trait = _____
Tongue Rolling: Can roll tongue (R) Cannot roll tongue (r)	Can roll = _____ Cannot roll = _____	% with dominant trait = _____ % with recessive trait = _____
Freckles: Have freckles (F) No freckles (f)	Freckles = _____ No freckles = _____	% with dominant trait = _____ % with recessive trait = _____
Dimples: Present (D) Absent (d)	Present = _____ Absent = _____	% with dominant trait = _____ % with recessive trait = _____
Chin: Cleft chin (C) No cleft (c)	Cleft chin = _____ No cleft = _____	% with dominant trait = _____ % with recessive trait = _____
Pinky Finger: Bent (P) Straight (p)	Bent pinky = _____ Straight pinky = _____	% with dominant trait = _____ % with recessive trait = _____
Hitchhiker Thumb: Straight thumb (T) 90-degree bend in thumb (t)	Straight thumb = _____ Hitchhiker thumb = _____	% with dominant trait = _____ % with recessive trait = _____
Interlocking of Fingers: Left over right (L) Right over left (l)	Left over right = _____ Right over left = _____	% with dominant trait = _____ % with recessive trait = _____

Follow-Up Question

Based on your data, are dominant traits always the most common?

Definition: Genotype vs. Phenotype

Genotype = genetic makeup of an individual

Phenotype = individual's physical appearance

Genetic counselors are trained to detect inheritance patterns of genetic diseases based on information they obtain from the family. Imagine that you are a genetic counselor and you must solve the following cases based on the information provided. Use the following steps to solve each problem:

1. Write down the genotype and phenotype of the parents (ex. DD → dimples).
2. Use a Punnett square to cross the potential gametes of the parents.
3. Determine the probability based on the Punnett square.

? Practice Questions

Problem 1: Widow's Peak versus Straight Hairline

A widow's peak (V-shaped hairline) is a dominant trait (W), while a straight hairline is recessive (w). A heterozygous man (Ww) and a woman with a straight hairline (ww) have children.

Questions:

1. What are the possible genotypes of their children?
2. What percentage of their children will have a widow's peak?
3. What percentage will have a straight hairline?

Problem 2: Freckles

Freckles (F) are dominant over no freckles (f). Two parents who are both heterozygous for freckles (Ff) have a child.

Questions:

1. Use a Punnett square to show all possible genotypes.
2. What are the chances their child will have freckles?
3. What are the chances their child will not have freckles?
4. What is the genotypic ratio of the offspring?

Problem 3: Tongue Rolling

The ability to roll your tongue is a dominant trait (R), and the inability to roll is recessive (r).

A woman who **can roll her tongue** has a child with a man who **cannot** roll his tongue. Their child **cannot** roll their tongue either.

Questions:

1. What is the genotype of the man?
2. What must be the genotype of the woman?
3. What are the chances their next child can roll their tongue?

Problem 4: Attached Earlobes

Attached earlobes (e) are recessive to free-hanging/unattached earlobes (E).

A woman with **free-hanging/unattached earlobes** has a father with **attached earlobes**. She marries a man who also has **attached earlobes**.

Questions:

1. What is the woman's genotype?
2. What is the man's genotype?
3. What are the possible genotypes and phenotypes of their children?
4. What is the probability that a child will have attached earlobes?

Problem 5: Dimples

Having dimples (D) is dominant over not having dimples (d). Two parents both **have dimples**, and their child **does not**.

Questions:

1. What must the genotypes of the parents be?
2. What is the probability that their next child will have dimples?

3. Can either parent be homozygous dominant?

Problem 6: Hitchhiker's Thumb

A straight thumb (T) is dominant over a hitchhiker's thumb (t).

Two individuals with **straight thumbs** have a child with a **hitchhiker's thumb**.

Questions:

1. What are the genotypes of the parents?
2. Show a Punnett square for this cross.
3. What is the probability that they have another child with a hitchhiker's thumb?

Problem 7: Widow's Peak – Part 2

Revisit the widow's peak trait (W = dominant, w = recessive).

If two heterozygous parents have four children, what is the probability of:

1. All four having a widow's peak? Note: See box at the bottom of the page if you're not sure how to calculate probabilities.
2. Two having widow's peaks and two with straight hairlines?
3. At least one child with a straight hairline?

Problem 8: Tongue Rolling – Part 2

A heterozygous man who can roll his tongue and a woman who cannot roll her tongue have children.

Questions:

1. What are the possible genotypes and phenotypes of their children?
2. If they have four children, what is the expected ratio of tongue rollers to non-rollers?
3. Could any of their children be homozygous dominant? Why or why not?

Note: Calculating Probabilities

To calculate the probability of **all independent events** occurring together, **multiply** the individual probabilities of each event.

Example:

Two parents are both **carriers** for a recessive trait (genotype Aa). The chance that one child **inherits the recessive trait (aa)** is:

- **P(child is aa) = 1/4**

What is the probability that **two children in a row** will inherit the recessive trait?

- **P(child 1 is aa) = 1/4**
- **P(child 2 is aa) = 1/4**

These are independent events (one child doesn't affect the next), so:

$P(\text{both children have trait}) = 1/4 \times 1/4 = 1/16$

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CHAPTER OVERVIEW

Laboratory 8: Genetics of Sickle Cell Anemia

[8.1: Laboratory 8 Introduction](#)

[8.2: Mendelian Genetics and Probability](#)

[8.3: Laboratory 8 Notes Page](#)

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8.1: Laboratory 8 Introduction

Laboratory 8: Genetics of Sickle Cell Anemia Introduction

Hemoglobin is a protein found in red blood cells (RBCs) that transports oxygen throughout the body. The hemoglobin protein consists of four polypeptide chains: two alpha chains and two beta chains. Sickle cell disease (also called sickle cell anemia) is caused by a genetic mutation in the DNA sequence that codes for the beta chain of the hemoglobin protein. The mutation causes an amino acid substitution, replacing glutamic acid with valine. Due to this change in amino acid sequence, the hemoglobin tends to precipitate (or clump together) within the RBC after releasing its oxygen. This clumping causes the RBC to assume an abnormal “sickled” shape. Individuals who are homozygous for the normal hemoglobin allele (*HBA*) receive a normal hemoglobin allele from each parent and are designated **AA**. People who are homozygous for normal hemoglobin do not have any sickled RBCs.

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8.2: Mendelian Genetics and Probability

Procedure:

1. Watch the short film “The Making of the Fittest: Natural Selection in Humans” (a resource provided by HHMI BIOINTERACTIVE). While watching, pay close attention to the genetics of sickle cell trait and the connection to malaria infection.
2. Answer the following questions regarding genetics, probability, and pedigrees.

? Mendelian Genetics and Probability Questions

1. If two people with sickle cell trait have children, what is the chance that a child will have normal RBCs in both high- and low-oxygen environments? What is the chance that a child will have sickle cell disease? Write the possible genotypes in the Punnett square.

1. What is the chance that a child will have sickle cell trait?
 2. What are the chances that these parents will have three children who are homozygous for normal RBCs? Show your work.
 3. What are the chances that these parents will have three children with sickle cell trait? Show your work.
 4. What are the chances that all three of their children will have sickle cell disease? Show your work.
 5. What are the chances that these parents will have two children with sickle cell trait and one with sickle cell disease? Show your work.
2. An individual who has the sickle cell trait has children with a person who does not carry the HbS (sickle cell) allele.
 1. What are the genotypes of the parents? _____
 2. In the Punnett square, show all the possible genotypes of their children.

3. What are the chances that any one of this couple’s children will have sickle cell disease?
 4. If this couple lives in the lowlands of East Africa, what are the chances that one of their children would be resistant to malaria if exposed to the malaria parasite?
3. A woman with sickle cell disease has children with a man who has sickle cell trait. Answer the following questions:
 1. What are the genotypes of the parents? _____

2. What is the genetic makeup of the gametes the mother can produce? _____
3. What is the genetic makeup of the gametes the father can produce? _____
4. In the Punnett square, show all the possible genotypes of their children. Then, summarize the genotype and phenotype ratios of the offspring.

5. What are the chances that any one of this couple's children will have sickle cell disease?
6. If this couple moved to an area where malaria was endemic, which of their children would be more likely to survive if infected with the parasite? Explain your answer.

? Blood Type Genetics Questions

In humans, ABO blood type results from multiple alleles: I^A , I^B , and i^O . A few simple rules of blood type genetics are that:

4. Two parents who are heterozygous for Type A blood and have sickle cell trait have children. Answer the following questions:
 1. What is the genotype of the parents? _____
 2. What are the genetic makeups of all the possible gametes they can produce:
 3. Complete the two-trait Punnett square to determine the frequency of the different phenotypes in the offspring. Note: Consider blood type and normal vs. mutant hemoglobin in the various phenotypes).

5. Now try a different way of solving a two-trait cross. Because of Mendel's second law of heredity (law of independent assortment), you can work with the blood type gene and the hemoglobin gene separately. Set up two one-trait crosses with the following parents: the mother is heterozygous for Type B blood and has sickle cell trait and the father has Type AB blood and also has the sickle cell trait.

1. What are the chances that a child of this couple will have Type B blood and sickle cell trait?
2. What are the chances that a child will have Type AB blood and will not have sickle cell disease?
3. What are the chances that a child will have Type B blood and sickle cell disease?
4. What are the chances that a child will have Type B blood and normal hemoglobin?

? Pedigrees Questions

6. The following pedigree traces sickle cell disease through three generations of a family. Use the pedigree to answer the following questions:

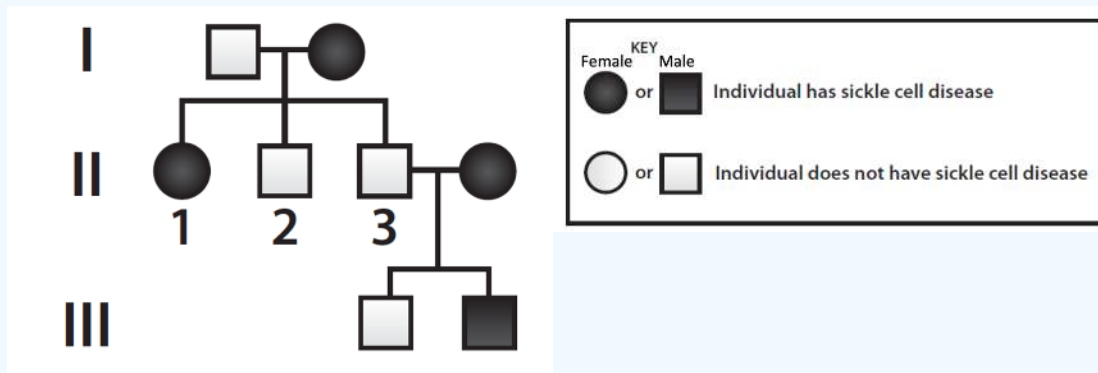


Figure 8.2.1: Pedigree chart and legend.

1. What is the genotype of the father in the first generation? _____
2. What is the genotype of the daughter in the second generation? _____
3. What is the genotype of individual 3 in the second generation? How do you know?
4. If the couple in the second generation has another child, what are the chances the child will have the following:
 1. Sickle cell disease _____
 2. Sickle cell trait _____
 3. Normal hemoglobin _____
5. If the entire family moves to an area where malaria is endemic, four of the five males in the pedigree will have a genetic advantage over the other individuals in the family. Explain why this is.
7. The following pedigree traces sickle cell disease through four generations of a family living in New York City. Use the pedigree to answer the following questions.

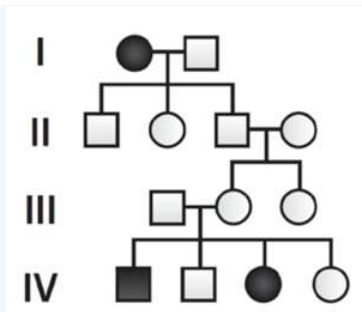


Figure 8.2.2: Pedigree chart.

1. What is the genotype of the mother in the first generation? _____
2. What are the possible genotypes of the father in the first generation? _____
3. What can you say about the genotype of all the children of the couple in the first generation? Explain your answer.
4. Regarding your answer to 6c, based on where the family resides, why would this genotype be considered a disadvantage?
5. What are the genotypes of the parents in the third generation? Explain how you know.
6. What is the possible genotype (or genotypes) of the mother in the second generation?
7. If the couple in the third generation has another child, what are the child's chances of the following?
 1. Having sickle cell disease _____
 2. Having sickle cell trait _____
 3. Being homozygous for normal RBCs _____
8. The following pedigree traces sickle cell disease through four generations of a family living in the highlands of eastern Africa. Use the pedigree to answer the following questions:

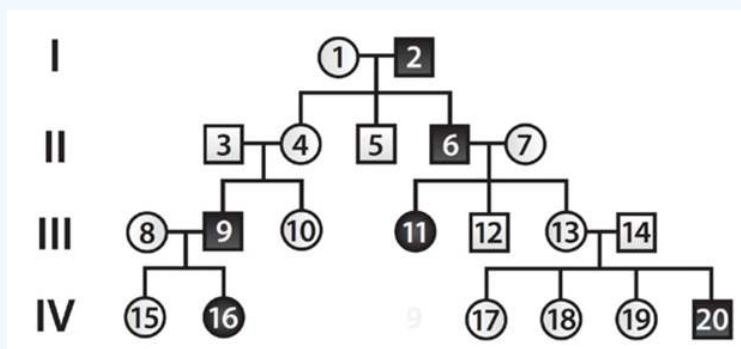


Figure 8.2.3: Sickle cell pedigree chart.

1. What are the genotypes of the following individuals?
 1. Individual 1: _____
 2. Individual 2: _____
 3. Individual 7: _____
 4. Individual 10: _____
 5. Individual 13: _____
 6. Individual 17: _____
2. If individuals 13 and 14 have another child, what are the chances that the child will have sickle cell disease?
3. If the same couple has three more children, what are the chances that all three children will have sickle cell disease? Show your work.
4. Based on where the family lives, does the sickle cell trait off a genetic advantage? Explain.

5. If individuals 8 and 9 have four more children, what are the chances that two of the children will be homozygous for normal RBCs? Explain why.
9. Imagine that you are a genetic counselor, and a couple planning to start a family comes to you for information. Jerome was married before, and he and his first wife have a daughter with sickle cell disease. The brother of his current wife, Michaela, died of complications from sickle cell disease, but neither of her parents had the disease.
 1. Draw a pedigree representing this family. Be sure to clearly label Jerome and Michaela.
 2. What is the probability that Jerome and Michaela will have a baby with sickle cell disease? Note that neither Jerome nor Michaela has sickle cell disease. Explain/show your work.
10. Natasha and Demarcus are planning on having children. Each has a sister with sickle cell disease. Neither Natasha nor Demarcus nor any of their parents have the disease, and none of them have been tested to see if they carry the sickle cell trait.
 1. Draw a pedigree representing this family. Be sure to clearly label Natasha and Demarcus.
 2. Based on this incomplete information, calculate the probability that, if this couple, has a child, the child will have sickle cell disease. Show your work.

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CHAPTER OVERVIEW

Laboratory 9: Biology of DNA

[9.1: Laboratory 9 Introduction](#)

[9.2: Gene Instructions for Making Proteins](#)

[9.3: Sickle Cell Anemia](#)

[9.4: Introduction to Gel Electrophoresis](#)

[9.5: Laboratory 9 Notes Page](#)

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9.1: Laboratory 9 Introduction

Laboratory 9: Biology of DNA Introduction

A gene is a segment of DNA that provides the instructions for making a protein. Proteins have many different functions that influence our characteristics. The chart below shows an example:

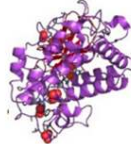
Gene in DNA	→	Protein	→	Characteristic
	→		→	
One version of a gene provides instructions to make normal protein enzyme.	→	Normal enzyme makes the pigment molecule in skin and hair.	→	Normal skin and hair color
Another version of the gene provides instructions to make defective protein enzyme.	→	Defective enzyme does not make this pigment molecule.	→	Albinism (very pale skin and hair)

Figure 9.1.1: Chart of genes, proteins, and characteristics.

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9.2: Gene Instructions for Making Proteins

Part 1. How Does a Gene Provide the Instructions for Making a Protein?

Genes Use a Two-Step Process:

The first step is the **transcription** of the gene.

Transcription produces **messenger RNA (mRNA)**.

The second step is the **translation** of the mRNA molecule.

Translation produces **proteins**.

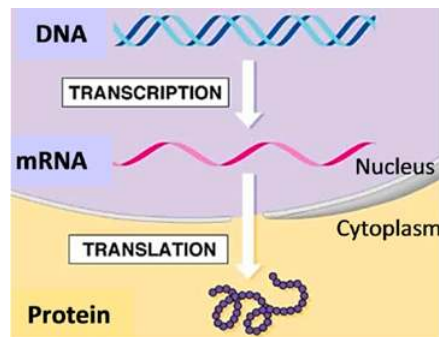


Figure 9.2.1: Two-step process for producing proteins.

? Questions

1. DNA and RNA are polymers of nucleotides.

1. The four nucleotides in **DNA** are:

2. The four nucleotides in **RNA** are:

2. During transcription, mRNA is produced from DNA. Given the following DNA sequence, provide the corresponding mRNA sequence:

DNA = TAC CCC GTC CAA ATA GGT TTA CAT

mRNA =

3. During translation, a ribosome translates the information on mRNA codon by codon. tRNA bearing the appropriate anticodon adds the corresponding amino acid. Copy the mRNA codons from #3 and provide the correct tRNA anticodons:

mRNA =

tRNA =

4. Which step takes place in the nucleus: transcription or translation? Explain why this process must occur there.

5. What is the name of the enzyme involved in transcription? What is its role?

6. DNA replication (occurs during the S phase of interphase in the cell cycle) and transcription (part of protein synthesis) are processes that can be easily confused with one another. In the table below, DNA replication is described in the first column. Fill in the blanks in the second column to summarize the differences between DNA replication and transcription.

DNA Replication	Transcription
All chromosomes/DNA is replicated.	_____ is transcribed.
DNA is made. DNA is double-stranded.	mRNA is made. mRNA is _____-stranded.
DNA polymerase is the enzyme that carries out DNA replication.	_____ polymerase is the enzyme that carries out transcription.
Thymine is used in DNA. A pairs with T in DNA.	Thymine is replaced by _____ in RNA. A pairs with _____ in mRNA.

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9.3: Sickle Cell Anemia

Part 2. Understanding How the Gene for Sickle Cell Hemoglobin Results in Sickle Cell Anemia

Introduction: Different versions of the same gene are called **alleles**. Alleles share the same general sequence of nucleotides, but they differ by at least one nucleotide.

? Questions

1. Fill in the blanks below to show how different alleles can result in different characteristics/phenotypes.

In DNA, different alleles of a gene have different sequences of _____ → leads to different sequences of _____ in mRNA → produces a different sequence of _____ in a protein → protein differs structurally and functionally → leads to different characteristics/phenotype.

2. Complete the table below to illustrate how the nucleotide sequence in a gene determines the sequence of amino acids in a protein. To begin, use the base-pairing rules to write the nucleotides of mRNA. Draw a rectangle around each codon in the mRNA molecule. Then, write the nucleotides for the tRNA anticodons. Finally, write the corresponding amino acid for each codon.

Nucleotide sequence at the beginning of the hemoglobin gene in DNA.	CACGTA	
Transcription of beginning of hemoglobin gene produces nucleotide sequence in mRNA.		
Anticodons in the tRNA molecules will bring the correct amino acids during translation.		
Amino acid that goes with each codon (use table below to help you with this).		

mRNA Codon	GUG	GAG	CUG	CCU	CAU	ACU
Corresponding Amino Acid	Val (Valine)	Glu (Glutamic Acid)	Leu (Leucine)	Pro (Proline)	His (Histidine)	Thr (Threonine)

Next, you will analyze how different alleles of the hemoglobin gene can result in sickle cell anemia.

3. Using the table on the next page, compare the partial nucleotide sequences of the “Normal Hemoglobin Gene” and the “Sickle Cell Hemoglobin Gene”. Highlight any difference(s) you see.
4. Complete the table by filling in the mRNA codons for both the normal and sickle cell hemoglobin genes along with the corresponding amino acids (use the same table you used for #9 to find the amino acids).

Normal vs. Sick Cell Hemoglobin

Normal Gene	Hemoglobin	CACGTAGACTGAGGACTC					
Transcription produces:		Codon 1	Codon 2	Codon 3	Codon 4	Codon 5	Codon 6
Normal mRNA	Hemoglobin						
Translation produces:		Amino Acid 1	Amino Acid 2	Amino Acid 3	Amino Acid 4	Amino Acid 5	Amino Acid 6
Normal Protein	Hemoglobin						
Sickle Cell Gene	Hemoglobin	CACGTAGACTGAGGACAC					
Transcription produces:		Codon 1	Codon 2	Codon 3	Codon 4	Codon 5	Codon 6
Sickle Cell mRNA	Hemoglobin						
Translation produces:		Amino Acid 1	Amino Acid 2	Amino Acid 3	Amino Acid 4	Amino Acid 5	Amino Acid 6
Sickle Cell Protein:	Hemoglobin						

5. Compare the amino acid sequences of the normal versus sickle cell hemoglobin genes. What difference(s) do you observe?

Sickle cell hemoglobin and normal hemoglobin differ in only a single amino acid out of more than 100 amino acids in the complete hemoglobin protein. This difference in a single amino acid results in the different properties of sickle cell hemoglobin compared to normal hemoglobin.

Hemoglobin is carried inside red blood cells. Normal hemoglobin dissolves in the watery cytosol of red blood cells. Sickle cell hemoglobin is less soluble in the cytosol because:

- Valine (Val) is much less water-soluble than glutamic acid (Glu).
- Amino acid 6 is a crucial location on the outer surface of the hemoglobin protein.

The chart below shows how the lower solubility of sickle cell hemoglobin results in the symptoms of sickle cell anemia:

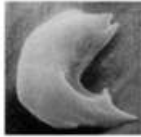
Genes in DNA	→	Protein	→	Characteristics
2 copies of the allele that codes for normal hemoglobin (SS)	→	Normal hemoglobin dissolves in the cytosol of red blood cells. 	→	Disk-shaped red blood cells can squeeze through the smallest blood vessels → normal health 
2 copies of the allele that codes for sickle cell hemoglobin (ss)	→	Sickle cell hemoglobin can clump in long rods in red blood cells. 	→	If sickle cell hemoglobin clumps in long rods → sickle-shaped red blood cells → clogged small blood vessels + fragile red blood cells → pain, damage to body organs + anemia = sickle cell anemia 

Figure 9.3.1: Sickle Cell hemoglobin.

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9.4: Introduction to Gel Electrophoresis

Introduction to Gel Electrophoresis:

One of the commonly used techniques to compare genomes is DNA gel electrophoresis. This technique is used to separate DNA fragments of different lengths resulting from a restriction enzyme digestion (using an enzyme to cut the DNA, essentially a DNA scissor). It takes advantage of the fact that a DNA molecule, or fragment, has an overall negative charge, and is thus attracted to a positive electric field. In addition to a digested DNA sample, DNA gel electrophoresis requires a gel electrophoresis chamber, which consists of a water-tight apparatus with a positive electrode on one end and a negative electrode on the other, containing an agarose gel.

The agarose gel, which is typically the shape of a thin slab, is a semi-solid gelatin matrix that has tiny holes that DNA fragments can move through. The gel is placed into the chamber with the wells closest to the negative electrode. The chamber is then partially filled with a clear electrophoresis buffer, which contains ions that allow it to carry electrical current. The gel has several empty spaces on one end called loading wells into which the DNA samples are loaded. Once the DNA samples are loaded into the wells, the chamber is then connected to a power supply and voltage is applied.

Because of DNA's negative charge, fragments will migrate through the gel toward the positive end of the chamber. The smaller the DNA fragment, the faster it will migrate (run) through the gel matrix, resulting in the separation of the fragments based on size. Once electrophoresis is complete, the smallest DNA fragment in each well will be closest to the positive electrode, and the largest DNA fragment will be closest to the loading well (the negative electrode). DNA fragments of the same size will collect at the same location in their lane, creating a band that can be seen. Any given band within a gel may have millions of DNA fragments of equal length.

Lab Materials

Materials:

- Micropipette
- Pipette tips
- Gel electrophoresis chamber
- Agarose gel
- Electrophoresis buffer
- DNA samples

Procedure:

1. Take the agar gel that has been set and solidified.
2. Remove the cover of the chamber by sliding it off while holding the chamber firmly.
3. Place the gel in the center of the electrophoresis chamber, oriented such that the wells are closest to the negative (black) electrode of the chamber.
4. If necessary, add more electrophoresis buffer to the chamber so that the top of the gel is covered by approximately 1cm of buffer.
5. Carefully load 10 μ l of each sample into the wells of the gel.

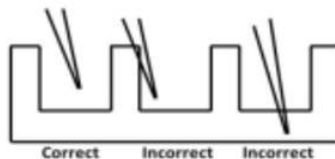


Figure 9.4.1: Correct way to load sample.

6. Place the cover on the gel electrophoresis chamber and connect the electrode terminals to the power source.

1. Make sure that the negative (black) and positive (red) color-coded indicators on the cover and chamber are properly oriented.
2. Likewise, make sure that the electrodes are plugged into the correct electrode terminals on the power source. Before proceeding to the next step, ask the instructor to inspect your setup.
7. Plug in and turn on the power source and set it to 75V. You should be able to see bubbles forming at each end of the chamber.
8. Run your gel for approximately 60 minutes.
 1. Caution: Do NOT allow your gel to run too long, or else the fragments will literally run out of the end of the gel!
 2. Carefully monitor the progress of your samples every 5 minutes.
 3. Typically, the gel is done and should be turned off when the blue 'fast dye' has run approximately 2/3rds down the gel.
 4. When you think the gel has finished running, let your instructor know.
9. After electrophoresis is complete, turn off and unplug the power source, then carefully remove the cover from the chamber.
10. Using the figure below, draw a picture of your results and answer the questions below.
11. Follow your instructor's instructions on how to clean and reset your gel electrophoresis experiment.

Results:

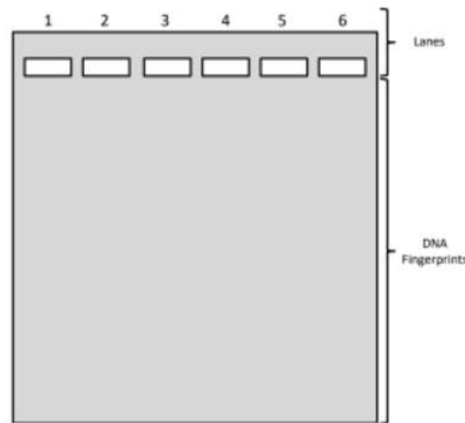


Figure 9.4.2: Blank results for gel electrophoresis.

? Follow-Up Questions

1. What would happen if you positioned the agarose gel backward within the chamber, with the sample wells on the positive electrode end of the electrophoresis chamber?
2. Describe why larger (longer) fragments move through an agarose gel more slowly than smaller (shorter) fragments?
3. Below are the results of a gel electrophoresis run used to determine the paternity of a child. The first lane contains the child's sample. The second lane contains the mother's sample. Which lane (3, 4, or 5) belongs to the child's father? How can you tell?

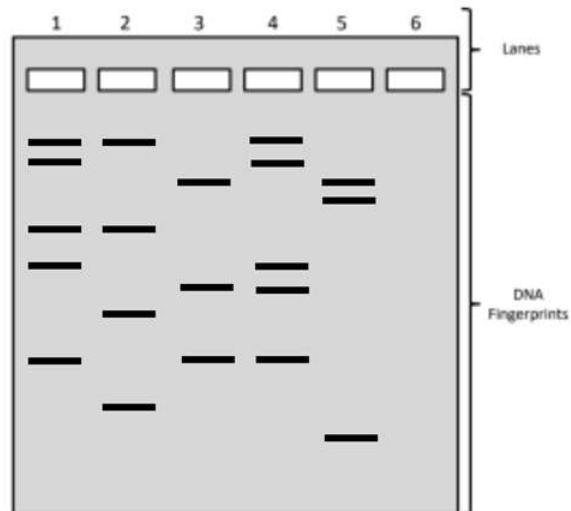


Figure 9.4.3: Results of a gel electrophoresis

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CHAPTER OVERVIEW

Laboratory 10: Evolution and Natural Selection

[10.1: Introduction to Laboratory 10](#)

[10.2: Best Beaks](#)

[10.3: Laboratory 10 Notes Page](#)

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10.1: Introduction to Laboratory 10

Laboratory 10: Evolution: Natural Selection Introduction

While in the Galapagos Islands, Darwin noticed that the beaks of various finches were of different lengths, shapes, and sizes. The differences he observed led him to several of his most important ideas about the mechanisms and processes that are responsible for descent with modification, including natural selection.

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10.2: Best Beaks

Lab Materials

Materials:

- 4 wooden craft sticks
- 2 rubber bands
- 10 pennies
- Timer
- Ruler
- 25 large grapes
- 25 raisins
- 1 plate
- 2 bowls

Sialia Mexicana (the Western Bluebird) is a small passerine bird of the family Turdidae. It has a thin, straight beak and feeds primarily on worms, berries, and insects, but it is also known to be a voracious consumer of grapes and raisins. In this exercise, you will simulate the functional consequences of variation in beak shape (short, stout beaks compared to long, thin beaks) of the Western Bluebird and how these might be reflected in subsequent generations. For the purposes of this activity, let's assume:

- There is variation in beak length among individuals in a population.
- Variation in beak length is heritable.
- Individuals with one type of beak length survive or reproduce better than others.

Procedure:

1. Make your short, stout beak (Figure 10.2.1) by placing three pennies between two of the wooden craft sticks about one inch (2.54 cm) from one end.
2. Secure the wooden craft sticks together with one of the rubber bands.



Figure 10.2.1: The three-penny short, stout beak.

3. Now, make your long, thin beak (Figure 10.2.2) by placing seven pennies between the remaining two wooden craft sticks, this time approximately 2.5 inches (6.35 cm) from one end, and secure with a rubber band.

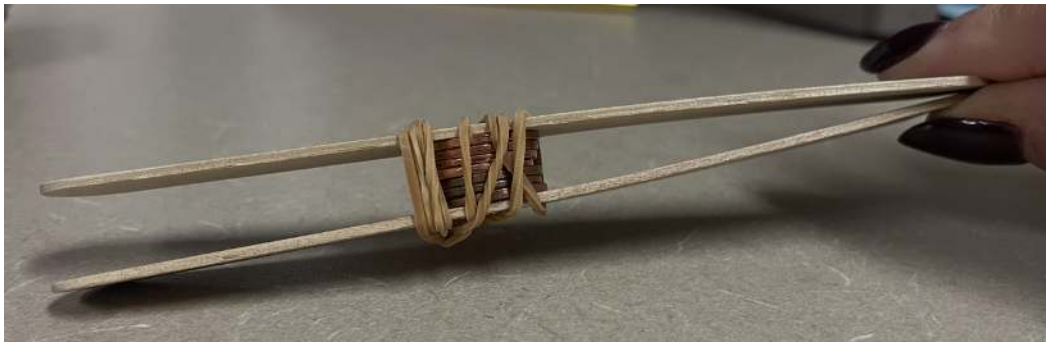


Figure 10.2.2: The seven-penny long, thin beak.

4. Spread the raisins evenly over the surface of the plate and set your timer for 30 seconds.
5. For 30 seconds, attempt to pick up as many raisins as possible with your small beak and place them in the bowl.
6. Record the number of raisins picked with the small beak in Table 10.2.1.
7. Return the raisins to the plate.
8. Complete a second trial, attempting to pick up as many raisins as possible with the small beak in 30 seconds. Record your results in Table 10.2.1.
9. Now, attempt to pick up as many raisins as possible with the large beak and place them in the bowl in 30 seconds.
10. Record the number of raisins picked up with the large beak in Table 10.2.1.
11. Clear the plate.
12. Spread the grapes out evenly on the plate, and again, set your timer for 30 seconds.
13. Using your small beak, attempt to pick up as many of the grapes as possible in 30 seconds and place them in the bowl.
14. Record the number of grapes picked up with the small beak in Table 10.2.2.
15. Now, using the large beak, attempt to place as many of the grapes as possible in the bowl in 30 seconds.
16. Record the number of grapes picked up with the large beak in Table 10.2.2.
17. Return the grapes to the plate.

Table 10.2.1: Number of Raisins Acquired by Beak Size.

Beak Length	Trial Number	Trial Time	Number of Raisins
Short, stout	1	30 sec.	
Short, stout	2	30 sec.	
Long, thin	1	30 sec.	
Long, thin	2	30 sec.	

Table 10.2.2: Number of Grapes Acquired by Beak Size.

Beak Length	Trial Number	Trial Time	Number of Grapes
Short, stout	1	30 sec.	
Short, stout	2	30 sec.	
Long, thin	1	30 sec.	
Long, thin	2	30 sec.	

? Follow-Up Questions

1. Assuming that the grapes represent the most common food source available to the Western Bluebird to be eaten in a normal year, which beak is better suited for eating/retrieving the grapes?
2. Now, imagine that a severe drought results in there being numerous raisins available, but very few grapes. Which beak is better suited for eating/retrieving raisins?
3. Following a drought year, how would you predict the frequency/abundance of long-beaked Western Bluebirds to change?
4. Does the predicted change in the abundance of birds with short, stout beaks reflect stabilizing, disruptive, or directional selection?
5. If grapes are the most common food source for most of the years, and drought occurs infrequently, how could there be variation in beak length?

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CHAPTER OVERVIEW

Laboratory 11: Microevolution and Macroevolution

[11.1: Introduction to Laboratory 11](#)

[11.2: Microevolution](#)

[11.3: Macroevolution](#)

[11.4: Laboratory 11 Notes Page](#)

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11.1: Introduction to Laboratory 11

Microevolution Introduction

Nearly every population of organisms is genetically diverse, meaning that no two individuals are genetically identical and possess the exact same alleles (unless they are identical twins). The alleles an individual has describes their genotype. Genotype determines one's phenotype (physical appearance). Natural selection only acts on phenotype – individuals who are better adapted to their environment are more likely to survive and reproduce, thus causing the frequency of their alleles in the population to increase. Conversely, those whose traits are selected against are less likely to survive and reproduce, therefore causing their alleles in the population to decrease. A change in allele frequency in a population is known as microevolution.

Every individual inherits two copies of each allele (one from each parent), and alleles can act differently. Some alleles are dominant (and, therefore, expressed) while others are recessive (i.e., masked in the presence of a dominant allele). In order for an allele to have an effect on an individual's phenotype, it must be expressed. If the phenotype generated by a certain allele faces selection pressure (whether in favor of or against their phenotype), there is potential for changes in allele frequencies in the population. The outcome is ultimately determined by how various phenotypes interact with their environment.

In this exercise, we will look at our class as the “population”. A deadly, incurable virus has stricken our population and it is highly transmissible from person to person through the exchange of body fluids. The question is... will you survive?

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11.2: Microevolution

Procedure:

1. Each person should obtain a pipette and a test tube from the test tube rack in the front of the room.
2. Randomly pick two beads from the container in the front of the room – this will represent your genotype.
3. Wander around the lab and “exchange fluids” with four of your classmates (ew!). To “exchange fluids”, place about 10 drops of fluid from your test tube into another person’s and have them place about 10 drops of fluid into your test tube.
4. Once you have completed #3, your instructor will add a small amount of cabbage extract to your tube. The color of your solution will indicate whether you have been infected or not (your instructor will provide a color scale for reference).

? Data/Follow-Up Questions

Are you still alive??? If you’re not infected, you’ll be fine. But if you ARE infected, genetic diversity plays a role in whether or not you’ll survive the viral plague. Your instructor will tell you which genotype is resistant to the virus. People with that genotype will survive and be the founders of a new population.

Record the class data in the table below and calculate the frequency of the “A” and “a” alleles before and after viral transmission.

Timeline	# students with AA genotype	# students with Aa genotype	# students with aa genotype	Frequency of A allele	Frequency of a allele
Before Virus					
After Virus					

To Calculate Allele Frequencies:

Frequency of A allele:

$$\frac{[(2 \times \# \text{ of AA}) + (\# \text{ of Aa})]}{(2 \times \text{total } \# \text{ of surviving students})}$$

(2 x total # of surviving students)

Frequency of a allele:

$$\frac{[(2 \times \# \text{ of aa}) + (\# \text{ of Aa})]}{(2 \times \text{total } \# \text{ of surviving students})}$$

2 x total # of surviving students)

1. In this simulation, which allele was selected for, and which allele was selected against?
2. What changes in allele frequencies occurred in response to the virus?

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11.3: Macroevolution

Macroevolution Exercise Introduction

Speciation is the formation of a new species. In order for a new species to come into existence, mutations must occur in a population that introduces genetic diversity. The phenotypes that arise from these mutations are then subject to natural selection. The varied adaptations/traits of individuals in the population will make some more likely to survive and reproduce than others.

Allopatric speciation occurs when populations of the same species are separated by a geographic barrier. Over time, as these separate sub-populations face different selection pressures within their respective environments, they become genetically distinct, resulting in the formation of a new species!

In this exercise, you will imagine that your workstation is a remote island. On this island, lives the species, *Triplodonticus megalcephalii*. Due to continental drift, each island has become isolated from all the others, and since *Triplodonticus megalcephalii* are poor swimmers, they're unable to travel to different islands. On each island, the population of *T. megalcephalii* will be exposed to different random mutations and varying selection pressures.

Procedure:

On your bench, you will find a deck of cards. The blue card shows a picture of *Triplodonticus megalcephalii* with its starting population size on the island. The yellow cards represent mutations that arise and become fixed in all members of the population. The green cards represent selection pressures and their influence on populations with various traits. As you follow the directions on the upcoming pages, your population will mutate, reproduce, and respond to various selection pressures.

? Exercise

Generation 1:

In the space below, draw *Triplodonticus megalcephalii* and write down your initial population size.

Population Size of Generation 1: _____

Generation 2:

Shuffle the mutation cards and place them face down. Then, pick one mutation card at random.

What is the mutation that occurred? _____

Given the new mutation that has just arisen in the population, redraw your creature, keeping the original body plan but adding the new mutation (do not modify the creature otherwise).

Shuffle the selection pressure cards and place them face down. Pick one card at random and write down the selection pressure here: _____

Using the information on the card, calculate the new population size of Generation 2: _____

Was the selection pressure positive, negative, or neutral? _____

Generation 3:

Pick another mutation card

What is the mutation that occurred? _____

Redraw your creature below with the new mutation that occurred:

Pick another selection card and write down the selection pressure here: _____

Using the information on the card, calculate the new population size of Generation 3: _____

Was the selection pressure positive, negative, or neutral? _____

Generation 4:

Pick another mutation card

What is the mutation that occurred? _____

Redraw your creature below with the new mutation that occurred:

Pick another selection card and write down the selection pressure here: _____

Using the information on the card, calculate the new population size of Generation 4: _____

Was the selection pressure positive, negative, or neutral? _____

Generation 5:

Pick another mutation card

What is the mutation that occurred? _____

Redraw your creature below with the new mutation that occurred:

Pick another selection card and write down the selection pressure here: _____

Using the information on the card, calculate the new population size of Generation 5: _____

Was the selection pressure positive, negative, or neutral? _____

Finally, make up a scientific name for your creature. Will you keep the original name, or do you think a new name would be appropriate?

Creature's name: _____

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CHAPTER OVERVIEW

Laboratory 12: Plant Diversity

[12.1: Introduction to Laboratory 12](#)

[12.2: Seedless Plants](#)

[12.3: Seed Plants](#)

[12.4: Laboratory 12 Notes Page](#)

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12.1: Introduction to Laboratory 12

Introduction

Plants (kingdom Plantae) are **autotrophs**; they make their own organic nutrients. The term “organic” refers to compounds that contain carbon. Organic nutrients, such as sugars, are made by photosynthesis.

Plants are adapted to living on land. For example, the above-ground parts of most plants are covered by a waxy layer called a cuticle to prevent water loss. **Aquatic plants are secondarily adapted to living in water.**

Some evidence that suggests that plants evolved from the green algae is:

- They both use chlorophyll a, chlorophyll b, and carotenoid pigments during photosynthesis.
- The primary food reserve of both is starch.
- They both have cellulose cell walls.

Genetic and morphological evidence indicates that plants evolved from a group of green algae called charophyceans. Many charophyceans inhabit shallow freshwater environments. Natural selection may have favored individuals capable of surviving occasional drying in these environments and this gave rise to land plants.

These traits occur in plants but not charophyceans. Some evolved independently in other algae.

- Apical meristems
- Alternation of generations
- Spores with protective walls
- Spores produced in sporangia
- Gametes are produced in multicellular structures called gametangia; Antheridia produce sperm; Archegonia produce eggs
- Multicellular dependent embryos
- Many have a cuticle that waterproofs and offers some protection

Alternation of Generations

The basic alternation of generations life cycle is illustrated below.

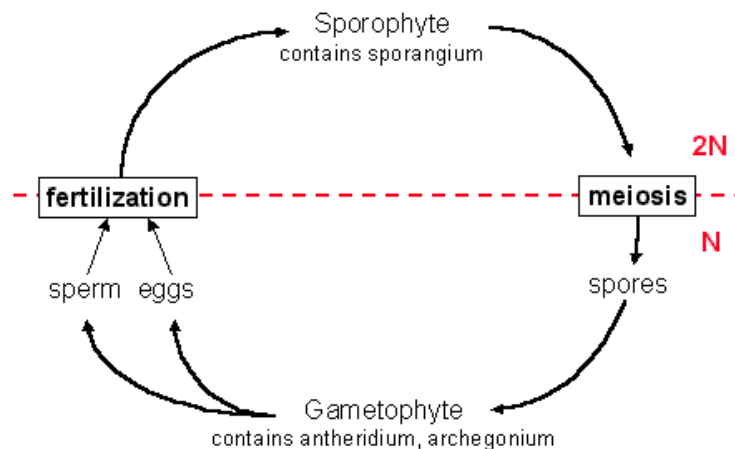


Figure 12.1.1: Spores produce gametophytes.

The diploid plant that produces spores is called a **sporophyte**. The haploid plant that produces gametes is called a **gametophyte**.

Some protists also have an alternation of generations life cycle, but the structures that produce gametes in protists are usually single cells. Plants produce gametes in multicellular structures that have an outer protective layer. Sperm are produced in structures called **antheridia** (sing. antheridium), eggs are produced in **archegonia** (sing. archegonium). As in protists and fungi, spores of plants are produced in **sporangia** (sing. sporangium).

A **dependent sporophyte** is a sporophyte that is small and grows attached to the gametophyte. It obtains nutrients from the gametophyte. An **independent sporophyte** grows separately from the gametophyte. Similarly, a **dependent gametophyte** is small

and grows attached to the sporophyte, while an **independent gametophyte** grows separately from the sporophyte.

The evolutionary trend in plants has been from plants with a dominant gametophyte and reduced, dependent sporophyte (ex., Mosses) to plants with a dominant, independent sporophyte and a reduced, dependent gametophyte (ex., Seed plants).

Classification

Evolutionary relationships among the plants are shown below.

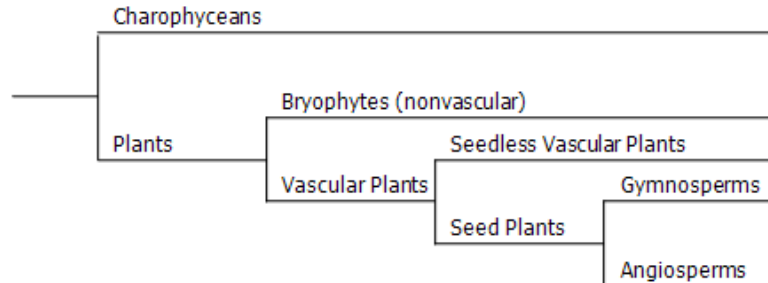


Figure 12.1.2: An evolutionary tree starting with two branches: Charophyceans and plants.

We will study the following phyla of plants.

Characteristics	Classification
Bryophytes (no vascular tissue)	Liverworts (Phylum Hepatophyta) Mosses (Phylum Bryophyta) Hornworts (Phylum Anthocerophyta)
Seedless vascular plants	Club mosses, Spike Mosses, Quillworts (Phylum Lycopphyta)Horsetails, Whisk Ferns, Ferns (Phylum Pterophyta)
Gymnosperms (vascular, naked seeds)	Conifers (Phylum Coniferophyta) Cycads (Phylum Cycadophyta) Ginkgos (Phylum Ginkgophyta) Gnetophytes (Phylum Gnetophyta)
Angiosperms (vascular, protected seeds)	Flowering Plants (Phylum Anthophyta)Monocots Eudicots

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12.2: Seedless Plants

Seedless Plant Lab

Introduction

Three major groups of land plants, Bryophytes, Lycophytes, and Ferns, are called seedless plants because they do not produce seeds (an adaptation that developed in later groups). A commonality of seedless plants is that they produce flagellated sperm, which require the presence of water to successfully fertilize an egg. For this reason, seedless plants are not as well-adapted to terrestrial life as more recently evolved groups.

Bryophytes, a gametophyte-dominant group of plants, includes the mosses, liverworts, and hornworts. These plants are seedless and lack vascular tissue as well as true leaves, stems, and roots.

Lycophytes, which includes club mosses, spike mosses, and horsetails, are a sporophyte-dominant group. Lycophytes are the first group of land plants to possess vascular tissue and true plant organs (roots, stems, and small leaves called microphylls).

Ferns are the third major group of land plants to evolve. They are sporophyte-dominant and possess vascular tissue. A notable trait that evolved in the ferns is megaphylls, large leaves that enhance the plant's ability to absorb solar energy.

In this portion of the lab, you will be observing examples of the three groups of seedless plants (bryophytes, lycophytes, and ferns). As you observe the specimens provided and describe the life cycles for each, think about the similarities and differences you see in the three groups.

Lab Objectives

At the conclusion of the lab, the student should be able to:

- Explain what is meant by “alteration of generations”
- Explain the difference between the sporophyte and gametophyte generation in plants. State which generation is haploid and which is diploid
- Name the process that makes spores and state if spores are haploid or diploid
- Name the process that creates sperm and egg from spores and state if sperm and egg are haploid or diploid
- Name the phyla discussed in the lab and give an example of a plant from each
- Identify and know the function of the archegonium and the antheridium
- Identify the fern structures discussed
- Understand the seedless plant life cycle

Bryophytes

Phylum: Bryophyta (Mosses)

1. Observe different kinds of moss on display and note the body form of the gametophyte.



Figure 12.2.3: Moss growing on a rock.

2. Obtain live sporulating moss (if available) and identify the sporophyte and gametophyte generations.

1. Is the green “leaf-like” tissue the gametophyte or sporophyte?
2. Is the stalk that emerges from the green “leaf-like” tissue the gametophyte or sporophyte?

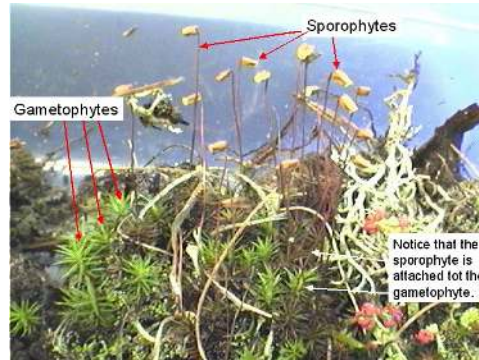


Figure \(\backslash\)
PageIndex

{4}\): Moss gametophytes and sporophytes

4. Draw the life cycle of a typical bryophyte such as moss. Your drawing should contain the following terms:
 1. 2N (Diploid)
 2. N (Haploid)
 3. Sporophyte
 4. Sporangium
 5. Meiosis
 6. Spores
 7. Gametophyte
 8. Antheridium
 9. Sperm
 10. Archegonium
 11. Egg
 12. Fertilization
4. Is the archegonium male or female?
5. What cell is produced in the archegonium?
6. Is this cell haploid or diploid?
7. Is the antheridium male or female?
8. What cell is produced in the antheridium?
9. Is this cell haploid or diploid?
10. Observe a slide showing the antheridial head of a moss. Begin using the scanning (4X) objective and then switch to the low-power objective (10X).
11. What is produced in this structure (the antheridium)?
12. Show where the antheridium occurs on the live moss plant. Indicate where this structure occurs in the life cycle diagram that you prepared (above).

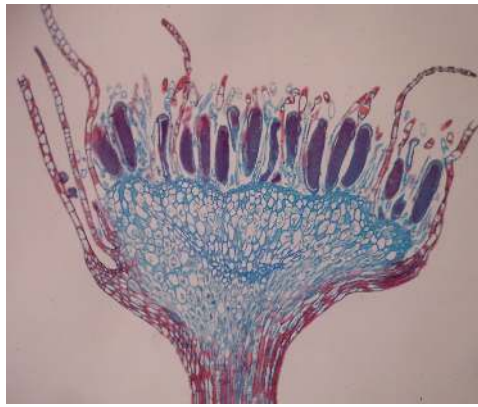


Figure 12.2.5: Mnium (a moss) antheridial head

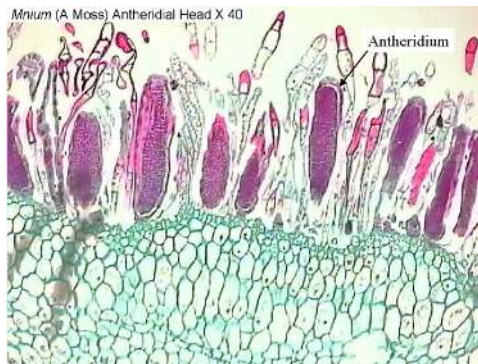


Figure 12.2.6: Mnium (a moss) antheridial head x40

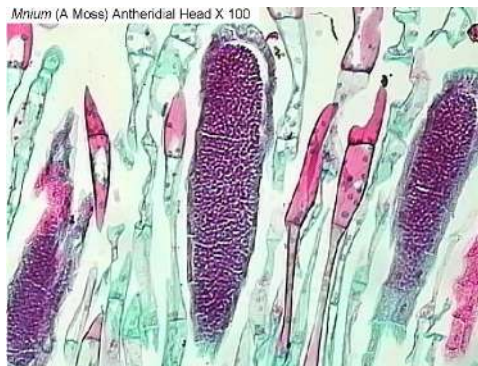


Figure 12.2.7: Mnium (a moss) antheridial head x100

13. Observe a slide showing the archegonial head of a moss. Begin using the scanning (4X) objective and then switch to the low-power objective (10X).
14. What is produced in the archegonium?
15. Show where the archegonium occurs on the live moss plant. Indicate where this structure occurs in the life cycle diagram that you prepared (above).

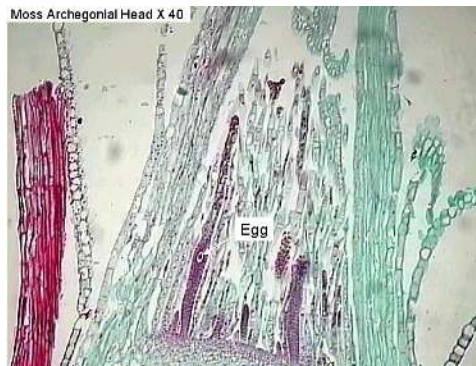


Figure 12.2.8: Moss archegonial head x 40



Figure 12.2.9: Moss archegonial head x 100

16. After the egg is fertilized, it grows and produces a sporophyte. A capsule containing a sporangium is found at the tip of the mature sporophyte. Refer back to Figure 2 on the previous page to view a sporophyte.
17. What is produced within the sporangium?
18. Be sure that you can identify the sporophyte and the sporangium on the live moss plant. Indicate where these structures occur in the life cycle diagram that you prepared (above).
19. How are moss spores dispersed to new locations?

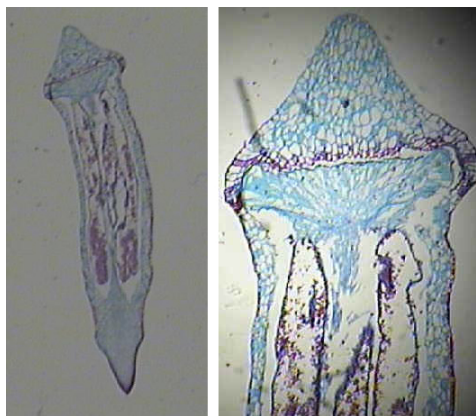


Figure 12.2.10: Moss capsule containing spores

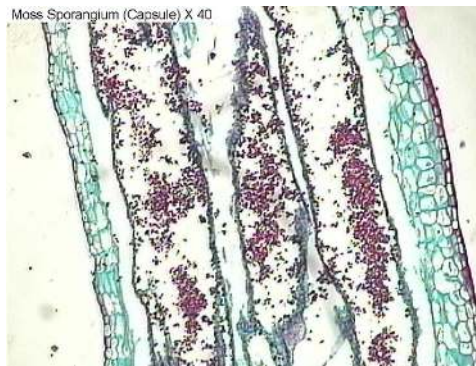


Figure 12.2.11: Moss capsule x40

Phylum: Lycophyta

Members of this phylum have horizontal stems, upright stems, and small, spike-shaped leaves called **microphylls**.

Club Mosses

Observe a specimen of live club mosses such as *Lycopodium*. Find **rhizomes**. Identify **microphylls**. Do the specimens have any **strobili**? (Be sure to look up these words if you do not understand them.)



Figure 12.2.19: Club moss (*lycopodium*)

Spike Mosses

Observe a specimen of a spike moss such as *Selaginella*. Note the structure of the **microphylls**.



Figure 12.2.20: Spike moss (*selaginella*)

Horsetails

Observe a live horsetail if available. Find the a **strobilus**. What reproductive structures are contained within the strobilus?



Figure 12.2.17: Horsetails



Figure 12.2.18: Horsetail showing strobilus

1. Observe the lycophyte slides provided to you. Begin using the scanning (4X) objective and then switch to the low-power objective (10X).

Note

The life cycles of lycophytes are very similar to that of bryophytes.

Phylum: Pterophyta (Ferns)



Figure 12.2.13: Fern gametophyte

1. Draw the life cycle of a fern. Your drawing should contain the following terms:
 1. 2N (Diploid)
 2. N (Haploid)
 3. Sporophyte
 4. Sorus
 5. Sporangium
 6. Meiosis
 7. Spores
 8. Gametophyte
 9. Antheridium
 10. Sperm

11. Archegonium
12. Egg
13. Fertilization

2. Observe sori on the underside of a fern leaf. Are sporangia visible? Indicate where this structure occurs in the life cycle diagram that you prepared (above).



Figure 12.2.14: Fern showing sori on underside of leaf



Figure 12.2.15: Fern sori x40

3. Is the frond sporophyte or gametophyte?
4. What cell is produced in the sori?
5. Is this cell diploid or haploid?
6. View a slide of a fern gametophyte showing antheridia. What reproductive cells are produced by gametophytes? Indicate where the gametophyte occurs in the life cycle diagram that you prepared.

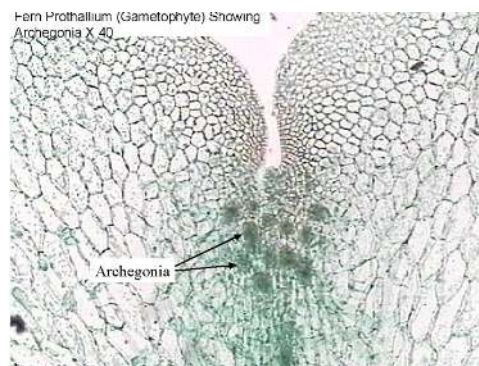


Figure 12.2.16: Fern prothallium (gametophyte)

7. View the display that includes the fern prothallus.
8. What shape is the prothallus?
9. Is the prothallus part of the sporophyte or gametophyte generation?

10. What reproductive structures are present on the prothallus?
11. What cells are made in the archegonium?
12. What cells are made in the antheridium?

? Follow-Up Questions

1. Is gametophyte tissue haploid or diploid?
2. Is sporophyte tissue haploid or diploid?
3. Is the moss life cycle gametophyte or sporophyte dominant?
4. Is the lycophyte life cycle gametophyte or sporophyte dominant?
5. Is the fern life cycle gametophyte or sporophyte dominant?
6. In the life cycle of plants, the process of meiosis produces what type of cell?
7. Does the gametophyte or sporophyte generation produce spores?
8. Which type of cell division is used to produce gametes in plants? How is this different than animals/humans?
9. State one reason why bryophytes, lycophytes, and ferns are considered primitive plants.
10. What is meant by the idea of “alteration of generations?”

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12.3: Seed Plants

Lab Objectives

- Define the terms (meanings of the names) gymnosperm and angiosperm
- State what type of cells create eggs and what type of cells create sperm in gymnosperms and angiosperms
- Describe the general characteristics of seed plants.
- Understand the basic gymnosperm and angiosperm life cycle
- Recognize the difference between a male and female pine cone
- Explain the seed plant alteration of generations including sporophyte and gametophyte structures
- Identify and know the function of the microscopes and the megaspores
- Identify the flower parts and which structures and male and female
- Explain and recognize the difference between a monocot and a eudicot flower
- Distinguish between the different types of fruits and give an example of each type

[Download a PDF of the lab to print.](#)

Procedure

Gymnosperms

1. Name the four subgroups within gymnosperms.
2. We will focus on conifers. Observe the conifer leaf samples available.
3. How does the needle-like leaf benefit the conifer?
4. Reproduction in Pines: Draw a simple pine life cycle in the space below. Be sure to include the terms egg, embryo, fertilization, megaspore, microscope, gametophyte, sporophyte, meiosis, mitosis, and pollen. [Use this website to get you started.](#)
5. Observe the pine cones on display. Are pine cones haploid or diploid?
6. Are male or female pine cones larger?
7. View the slide of the pollen (male) pine cone cross-section.
 - A. Can you find the microspores on the slide?
 - B. Are microspores haploid or diploid?
 - C. What process do microscopes undergo to form pollen grains?
 - D. Locate a pollen grain on the slide and draw one below.
8. View the slide of the seed/ovulate (female) pine cone cross-section.
 - A. Can you find the megaspores on the slide?
 - B. Are megaspores haploid or diploid?
 - C. What process do megaspores undergo to form the egg or ovule?
 - D. Can you find the egg on the slide?
 - E. Is the tissue surrounding the egg haploid or diploid?
 - F. Once the egg is fertilized, what structure will form?
9. View the pine seeds on display
 - A. Are the seeds haploid or diploid?
 - B. How will the seeds be dispersed through the environment?
 - C. What cell division process will the seeds undergo to create a new pine tree

Angiosperms

1. Draw a simple angiosperm life cycle in the space below. Be sure to include the terms egg, embryo, fertilization, megaspore, microspore, gametophyte, sporophyte, meiosis, mitosis, and pollen. Use this website to get you started: <http://www.sumanasinc.com/webcontent/animations/content/angiosperm.html>

2. Use the flower model to identify the different structures.

A. Collectively, the male flower parts are called the _____.

B. Collectively, the female flower parts are called the _____.

C. Collectively, the petals of the flower are called the _____.

D. Collectively, the sepals of the flower are called the _____.

3. Use the table below to describe the function of each flower part and if it is male, female, or neither.

Flower structure	Function	Male/Female/Neither
Anther		
Filament		
Stigma		
Style		
Ovary		
Petal		
Sepals		

4. Dissect a live flower. Start with the outside (sepals, petals) and work your way in. Identify each structure as you dissect the flower.

A. How many petals does the flower have?

B. Is the flower a monocot or a eudicot? What characteristic did you use to determine this?

C. Once you dissect the flower, dispose of the flower parts. You do not have to cut into the ovary or the anther.

5. View the slide of the lily mature female gametophyte (if one is available).

A. Can you locate the egg?

B. Is the egg haploid or diploid?

C. What type of cell underwent mitosis to create the egg?

D. Is the ovary (tissue surrounding the egg) haploid or diploid?

6. View the slide of the lily anther cross-section.

A. Can you locate the pollen grains?

B. Is the pollen haploid or diploid?

C. What cell type underwent mitosis to create the pollen?

D. Is the anther (tissue surrounding the pollen) haploid or diploid?

7. Observe the live bean seed (if one is available, otherwise skip).

A. Is the seed haploid or diploid?

B. How many cotyledons does the bean seed have?

C. Is the bean a monocot or a eudicot?

8. Observe the live corn seed (if one is available, otherwise skip).

A. How many cotyledons does the corn seed have?

B. Is the corn a monocot or a eudicot?

9. Fruits. There will be several fruit examples available in the lab. Use the table below to characterize the fruits you view.

Name of fruit	Simple, Aggregate, or Multiple	Dry or fleshy

Monocots and Eudicots

1. Observe the monocot root slide and model. Draw what you see under the microscope. Label the vascular tissue, ground tissue, and dermal tissue.

2. Observe the eudicot root slide and model. Draw what you see under the microscope below. Label the vascular tissue, ground tissue, and dermal tissue.

3. Observe the monocot stem slide and model. Draw what you see under the microscope below. Label the vascular tissue, ground tissue, and dermal tissue.

4. Observe the eudicot stem slide and model. Draw what you see under the microscope below. Label the vascular tissue, ground tissue, and dermal tissue.

? Review Questions

Answer the review questions below.

1. What does 'gymnosperm' mean?
2. What group of gymnosperm plants is the largest?
3. What type of spore is used for male reproduction in seed plants?
4. Through mitosis, the male spore develops into what structure?
5. What type of spore is used for female reproduction in seed plants?
6. Through mitosis, the female spore develops into what structure?
7. What does 'angiosperm' mean?
8. What structures of the flower are female?
9. What structures of the flower are male?
10. What is the function of the petals of the flower?
11. The process of _____ occurs in the flower anther to create haploid _____ followed by mitosis to create _____.
12. The process of _____ occurs in the flower ovary to create the haploid _____ followed by mitosis to create the _____ and the $n+n$ _____.
13. State one difference between monocots and eudicots.
14. What part of the flower develops into a fruit?
15. How is a simple fruit different from a complex fruit?
16. Give an example of a fleshy fruit
17. Give an example of a dry fruit.

18. If a plant's leaves have parallel veins, is it a monocot or a eudicot?
19. You are observing a cross-section of a plant root under the microscope and notice that the xylem elements are star-shaped. Is this root from a monocot or a eudicot?

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CHAPTER OVERVIEW

Laboratory 13: Invertebrates

[13.1: Introduction to Laboratory 13](#)

[13.2: Mollusk Dissection](#)

[13.3: Arthropod Dissection](#)

[13.4: Laboratory 13 Notes Page](#)

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13.1: Introduction to Laboratory 13

Arthropods are the most diverse group of animals on Earth, characterized by their segmented bodies, jointed appendages, and exoskeletons. In this lab, we will explore and compare the anatomy of two major arthropod groups—**crustaceans** (e.g., crayfish) and **insects** (e.g., grasshoppers)—to better understand their shared traits, specialized adaptations, and evolutionary relationships.

Despite differences in habitat and form, crustaceans and insects share a common ancestry. Both groups evolved from a segmented ancestor and display homologous features that have been modified through millions of years of evolution to suit different lifestyles, whether it's swimming in aquatic environments or flying through the air.

By performing careful dissections and examining both external and internal structures, students will:

- Identify key features of the **arthropod body plan**, such as segmentation, tagmata (body regions), and jointed appendages
- Compare **specialized adaptations** for movement, respiration, feeding, and reproduction
- Explore **homologous structures** and functional differences across terrestrial and aquatic environments

This lab will provide hands-on insight into how a single evolutionary blueprint can give rise to an incredible variety of body forms and behaviors across the arthropod phylum.

Questions to consider while completing the lab:

- How do the body segments and appendages of crustaceans and insects compare in structure and function?
- What do similarities in anatomy tell us about their evolutionary history?
- How have crustaceans and insects adapted to their respective environments through anatomical modifications?

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13.2: Mollusk Dissection

Materials needed for this exercise:

- Preserved bivalve specimen
- Dissection tray and pads
- Dissection tools: scalpel, scissors, forceps, probe, pins
- Gloves, lab coat, safety goggles
- Disinfectant and cleanup materials
- Lab notebook for diagrams and notes

Caution

- Always wear gloves and goggles.
- Handle the scalpel and scissors carefully.
- Be respectful—this is a real animal that was once alive.
- Dispose of waste properly and clean/dry tools thoroughly afterward.

Phylum Mollusca Overview:

The phylum Mollusca contains four major groups - bivalves (e.g., clams), chitons, gastropods (e.g., snails), and cephalopods (e.g., squid, octopus, cuttlefish). All mollusks have a **visceral mass**, a **mantle**, and a **foot**. The visceral mass contains the digestive, excretory, and reproductive organs. The mantle is a covering. It may secrete a shell. The foot is muscular and is used for locomotion, attachment, and/or food capture.

Bivalves (Class Bivalvia)

The mantle and foot can be seen in the Figure 13.2.1. The visceral mass is underneath the gill.

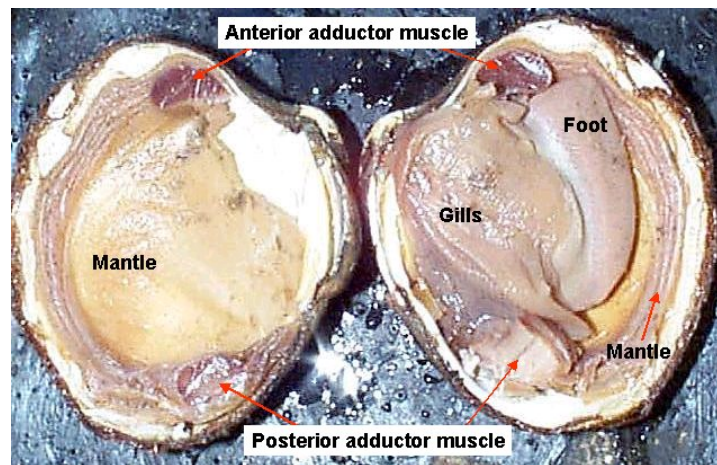


Figure 13.2.1: Anterior Adductor Muscle

There may be a **radula**, a structure that resembles a tongue but contains hard plates and is often used for scraping food. The coelom is reduced and limited to the region near the heart.

Most mollusks have an **open circulatory system**, but cephalopods (squids, octopuses) have a **closed circulatory system**. The blood pigment of mollusks is hemocyanin, not hemoglobin. The heart of a clam can be seen in the photograph below. Bivalves have three pairs of ganglia but do not have a brain.

Most mollusks have separate sexes, but most snails (gastropods) are hermaphrodites. Some marine mollusks have a ciliated larval form called a **trochophore**.

Chitons (Class Polyplacophora)

Chitons have a dorsal shell composed of 8 plates. A ventral foot is used for locomotion and for attachment to rocks. It pulls itself close to rocks for protection. Observe the chiton on display.



Figure 13.2.2: Left: chiton, dorsal surface. Right: ventral surface

Snails (Class Gastropoda)

Gastropods have an elongated, flattened foot and usually a head and shell, although nudibranchs (sea slugs) and terrestrial slugs lack a shell.

Most are marine, but there are also numerous freshwater and terrestrial species. Herbivorous gastropods use a radula to scrape food from surfaces. Carnivores may use a radula to bore a hole through surfaces such as bivalve (clam) shells. Some gastropods, such as the slug (below), do not have a shell.

The larvae undergo **torsion** during development. It is a twisting that positions the visceral mass so that the anus is above the head. It is due to one side of the visceral mass growing faster than the other. The advantage (or function) of torsion is uncertain, but it may be to balance the animal, or it may be to allow the head to be withdrawn into the shell first when predators approach.

Some gastropod species are terrestrial and have lungs for gas exchange, while others are aquatic and use gills.

The space near the entrance to the shell that is bound by the mantle is the **mantle cavity**. Aquatic gastropods have gills located in the mantle cavity. The mantle of terrestrial gastropods functions as a lung.

Observe the snails and slugs on display.



Figure 13.2.3: A slug. Slugs do not have shells.

Squid, Octopus, Nautilus, and Relatives (Class: Cephalopoda)

Cephalopods are predators and live in a marine environment. A closed circulatory system allows them to move rapidly in pursuit of prey. They move by jet propulsion; water in the mantle cavity is squirted rapidly through a siphon. The foot has evolved into tentacles around the head. Cephalopods have a powerful beak-like structure to tear apart prey. The sense organs of cephalopods are well-developed.

Mollusks are the simplest animals with eyes. Some mollusks have **lenses** and, therefore, are capable of forming clear images. The camera-type eyes of some cephalopods (squid, octopus) are capable of focusing and forming clear images. Cephalopods are fast-moving predators, and well-developed camera-type eyes help them catch prey.

Well-developed brains (especially in octopuses) give them a high learning capacity. Cephalopods can hide from enemies by using camouflage and/or releasing a dark-colored fluid from ink sacs.

The shell of a nautilus encloses the animal. A squid's shell is small and internal. Octopuses do not have shells. Examine representative cephalopods on display.



Figure 13.2.4: A Squid

Bivalve Dissection:

Bivalves have two shells (valves) held closed by powerful muscles. The shell is produced by the underlying mantle; it grows along the outer margins. They use their foot for burrowing. Mussels use their foot for the production of threads for attachment.

The gills are large because they are used for filter-feeding as well as respiration. Food is trapped by mucus on the gills and moved by cilia. Water enters and exits through siphons.

Obtain a preserved clam for dissection and place it on a dissecting tray. Remove one of the **valves** (shells) by inserting a scalpel and cutting the adductor muscles on each side of the hinge. See the diagram below for the location of the **adductor muscles**.

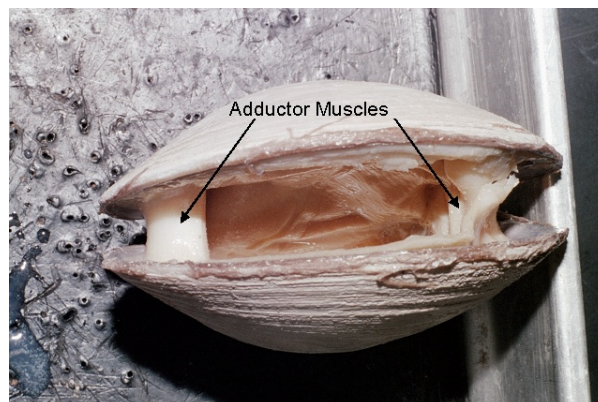


Figure 13.2.5: Adductor muscles of a clam.



Figure 13.2.6: The anterior and posterior adductor muscles are being cut so that the valves can be pulled apart.

The **mantle** is a membrane that surrounds the internal structures and is characteristic of all mollusks. The portion of the mantle from the exposed surface may have remained attached to the valve that was removed or it may be covering the internal structures. Find the mantle and if necessary, remove it to expose the internal structures of the clam.

Identify the foot, visceral mass, gills, and labial palps. The gills are large because they are used for filter-feeding as well as respiration. Food is trapped by mucus on the gills and moved by cilia to the mouth. Cilia on the labial palps also direct food and mucus to the mouth.

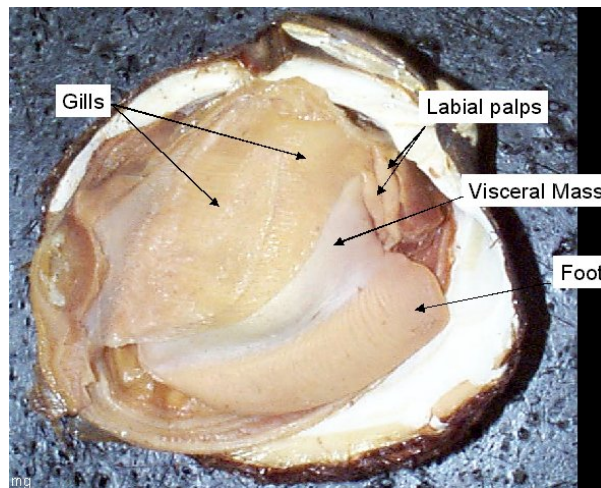


Figure 13.2.7: Interior of mollusk.

The heart can be found in the area dorsal to and slightly posterior to the visceral mass. The coelom of bivalves is reduced and limited to the area surrounding the heart. Because the coelom is reduced, it is difficult to see the digestive organs. Cut through the visceral mass and identify the intestine.

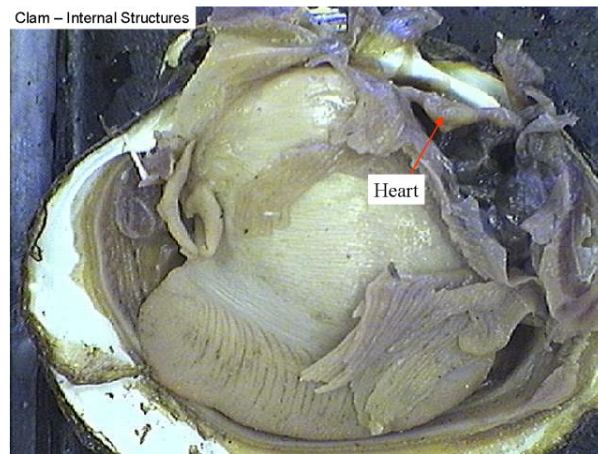


Figure 13.2.8: Clam internal structures.

In the photograph below, the foot and visceral mass have been lifted so that the mouth can be seen. Notice the labial palps on either side of the mouth.

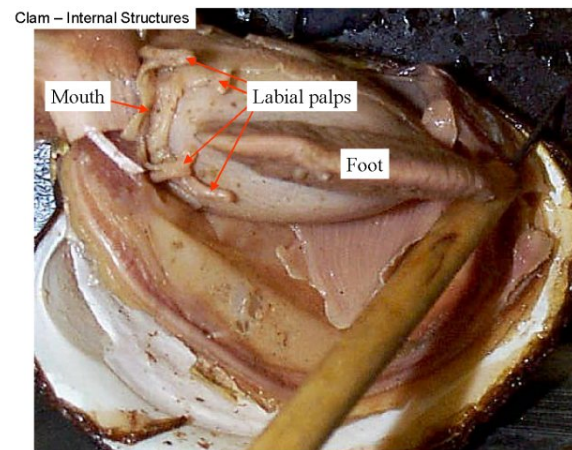


Figure 13.2.9: Clam internal structures.

Cephalopod Observation:

Observe the dissected squid that is on display.

Identify the following external structures:

- Mantle
- Fins
- Tentacles (longer, x2) and arms (shorter, x8)
- Siphon
- Eyes

Identify the following internal structures:

- Gills
- Hearts (one systemic heart, two branchial hearts)
- Cecum (digestive organ)
- Gonad (reproductive organs)
- Ink sac
- Pen (internal "shell")

Follow-Up Questions

1. What type of cephalopod is on display?
2. How is the "foot" of this animal modified compared to that of the bivalve?
3. This animal is also referred to as a decapod. Why? (Hint: "deca" = ten)
4. Does this animal exhibit cephalization?
5. What organ(s) does this animal use for respiration?
6. What is the siphon used for?
7. How many hearts does a squid have? What are they called, and what are their functions?
8. Since this animal does not have a hard exoskeleton, how does it protect itself?
9. Complete the table below to compare/contrast the two mollusks you observed.

Question	Bivalve	Cephalopod
Does the animal exhibit cephalization?		
What sort of respiratory organ does the animal use?		
What structure(s) does the animal use for movement?		
How does the animal acquire food?		
How does the animal protect itself?		

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13.3: Arthropod Dissection

Materials needed for this exercise:

- Preserved crayfish and grasshopper specimens
- Dissection tray and pads
- Dissection tools: scalpel, scissors, forceps, probe, pins
- Gloves, lab coat, safety goggles
- Disinfectant and cleanup materials
- Lab notebook for diagrams and notes

Caution

- Always wear gloves and goggles when working with specimens.
- Handle the scalpel and scissors carefully.
- Be respectful—this is a real animal that was once alive.
- Dispose of waste properly and clean/dry tools thoroughly afterward.

Introduction to Arthropods:

Three clades of arthropods are listed below. Note: Our main focus will be crustaceans and arthropods for this exercise, but there may be other arthropods on display for you to observe.

- Chelicerates
 - Arachnids—(Spiders, Scorpions, Harvestmen, Ticks and Mites)
 - Horseshoe Crabs
- Myriapods
 - Millipedes
 - Centipedes
- Pancrustaceans
 - Crustaceans
 - Decapods
 - Isopods
 - Krill
 - Copepods
 - Barnacle
 - Insects

Arthropods have an exoskeleton that cannot enlarge as the animal grows. As a result, it must be shed from time to time to allow growth (called ecdysis).

Life Cycle

The young of some arthropods look like the adults. The change from young to adult that these species undergo is called **incomplete metamorphosis**.

In many species, the **egg** hatches to produce a **larva** (pl. larvae) that does not look like the **adult**. At some point in the maturation process, the larva will produce a **pupa** (pl. pupae). In this phase, its tissues will become reorganized into the adult form. This type of development is called **complete metamorphosis**.

Observe the displays for incomplete and complete metamorphosis.

1. List an example of an arthropod that undergoes incomplete metamorphosis.
2. List an example of an arthropod that undergoes complete metamorphosis.

Chelicerates

The first pair of appendages of the members of this phylum are pincer-like or fang-like mouthparts called chelicerae.

Arachnids (Spiders, Scorpions, Harvestmen, Ticks, Mites)



Figure 13.3.1: Left: Scorpion. Middle: Tick X 40. Right: Mite X 40.

Horseshoe Crabs

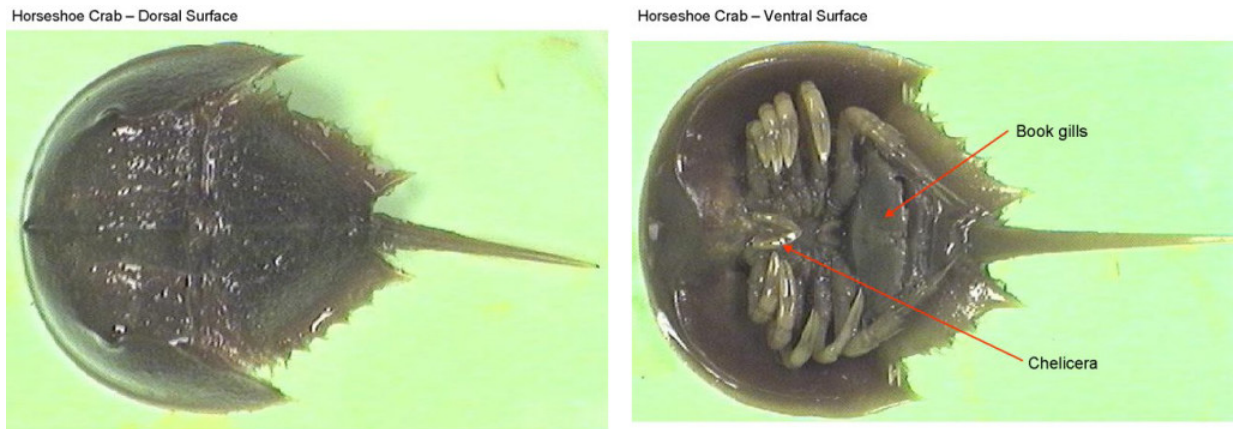


Figure 13.3.2: Left: Horseshoe crab dorsal surface. Right: Horseshoe crab ventral surface

Crustaceans—Lobsters and Relatives

Decapods



Figure 13.3.3: Crab

Crayfish Dissection

Obtain a crayfish and identify the structures shown in the photograph below. You should familiarize yourself with the names of the external structures shown in the photograph below.

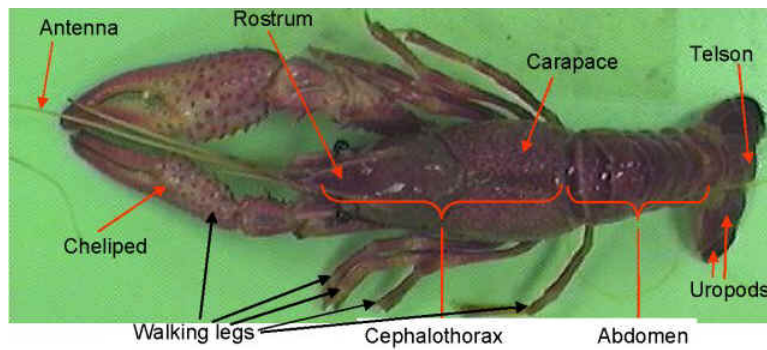


Figure 13.3.4: External structures of a crayfish.

The photographs below show the ventral surface of male and female crayfish. Identify the sex of your crayfish. Find somebody in class who has the opposite sex and view the ventral surface of their crayfish. Notice that crayfish have five pairs of **swimmerets**. In males, the anterior two pairs are large and less flexible than those of females. They are used to transfer sperm to the female.

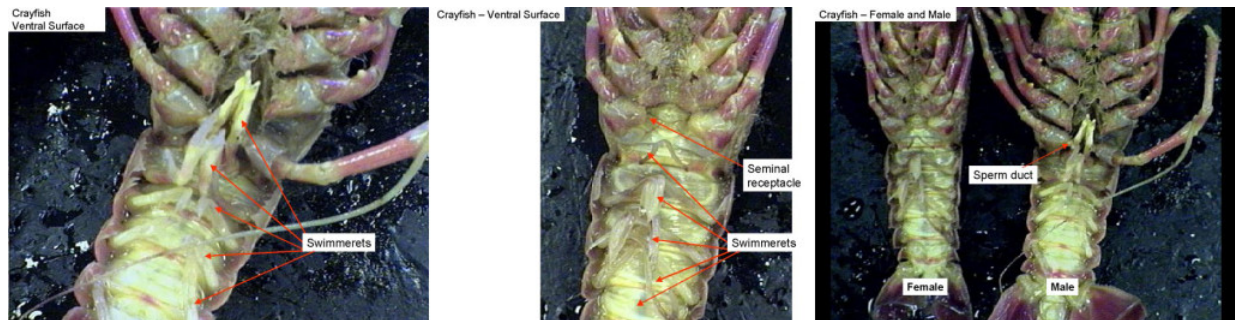


Figure 13.3.5: Ventral surface of male and female crayfish

Find the **seminal receptacles** located between the third and fourth walking legs. These structures function to receive sperm from the male during mating.

Observe the sperm duct in males, located at the base of the fifth walking leg.

Begin your dissection by cutting along the midline of the carapace from the posterior edge to an area just behind the two eyes (see the photographs below). Next, cut laterally just behind the eye until you reach the ventral edge. Carefully remove the piece of carapace that you have cut to expose the gills underneath. As you peel this piece of carapace away, it may be necessary to reach underneath with the point of the scissors or needle to brush away and detach any tissue from the interior of the animal that is attached to the carapace.



Figure 13.3.6: Cutting the midline.

Your crayfish should look like the first photograph below. Next, remove the carapace on the other side.

Using a needle, carefully separate the first row of **gills** and notice that there is another row underneath. Remove one of the legs and observe how the gills are attached to the walking legs.

The first two photographs below show the gills on the left side of a crayfish. The third photograph is a crayfish with the entire carapace removed. The first row of gills on each side has been moved aside to expose a row underneath it.

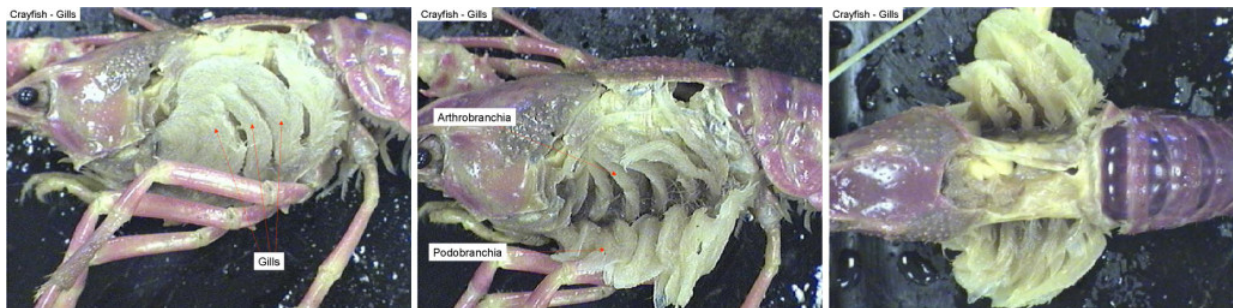


Figure 13.3.7: Dissected crayfish

Crayfish – Leg Showing Gill Attachment

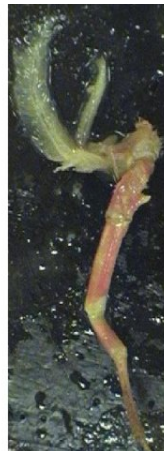


Figure 13.3.8: Crayfish leg showing gill attachment

Brush all of the gills to the side and then carefully cut the membrane (epidermis) that covers the internal organs.

The **heart** is a small diamond-shaped structure located below where the posterior edge of the carapace was. It may be difficult to see the blood vessels attached to the heart. Openings (ostia) in the side of the heart should be visible. These allow blood to enter the circulatory system.

Large white **digestive glands** can be seen on each side of the **stomach**. They produce digestive enzymes.

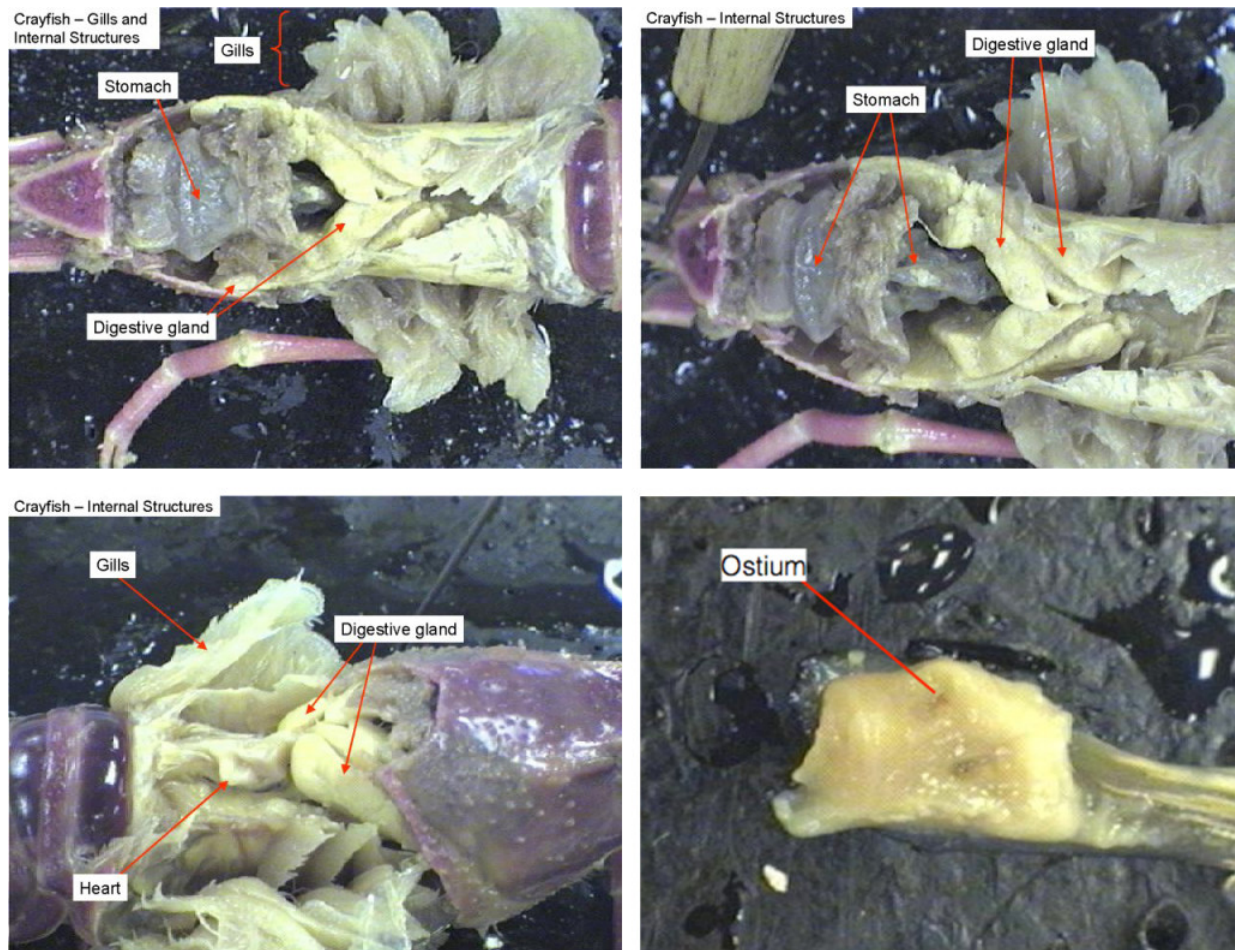


Figure 13.3.9: Crayfish internal structures

Remove the digestive glands and then carefully remove the stomach. Notice how the stomach is attached to the mouth. Cut open the stomach and observe the tooth-like structures at the anterior end for grinding food. This is called the **gastric mill**.

The **green glands** are positioned ventrally near the anterior end of the body cavity. They are spherical and will appear to be embedded within the surface. Their function is excretion. Find the openings for the green glands on the outer surface near the base of each antenna.

Find the **brain** near the anterior end and the two ventral nerve cords attached to the brain.

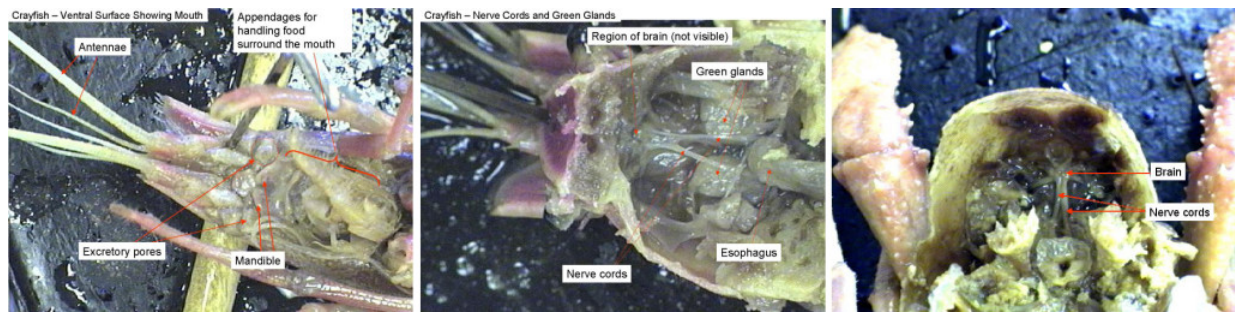


Figure 13.3.10: Left: Crayfish ventral surface. Middle: Crayfish viewed from above. The stomach has been removed, revealing the nerve cords and green glands underneath. Right: Crayfish viewed from above, looking into the head region. The stomach has been removed. The brain can be seen.

Copepods and Krill



Figure 13.3.11: Left: A copepod (Cyclops, male). Right: Krill (Gammarus).

Barnacles



Figure 13.3.12: Left: Balanus—a barnacle. Right: A stalked barnacle

Isopods

Isopod



Figure 13.3.13: Isopod

Myriopods

Millipedes



Millipede



Figure 13.3.14: Millipede



Figure 13.3.15: A centipede

Insects

Insects are the largest group of Arthropods, comprising over one million species. Insects are ubiquitous, filling virtually all ecological niches, including a variety of terrestrial and aquatic habitats. For this portion of the lab, you will view the external anatomy of a grasshopper. You will not have to dissect it, so it can be placed back in the jar when you are done.

Obtain a grasshopper and place it on a dissection pan/mat.

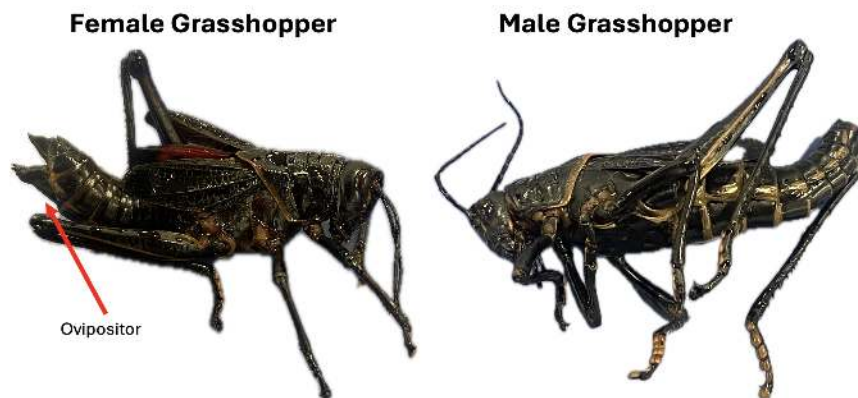


Figure 13.3.16: Female and Male Grasshoppers

1. Is this animal segmented (i.e., is its body divided into distinct segments)?

Determine whether your grasshopper is male or female by looking at its posterior abdomen (see photos above). Females will have a forked **ovipositor**, while males possess a **penis and claspers**.

2. Is your grasshopper male or female?

Observe the specialized sensory organs of the grasshopper. The animal possesses **antennae** that are used for _____. Locate the animal's **compound eyes** and its **tympanum**, an organ used for _____.

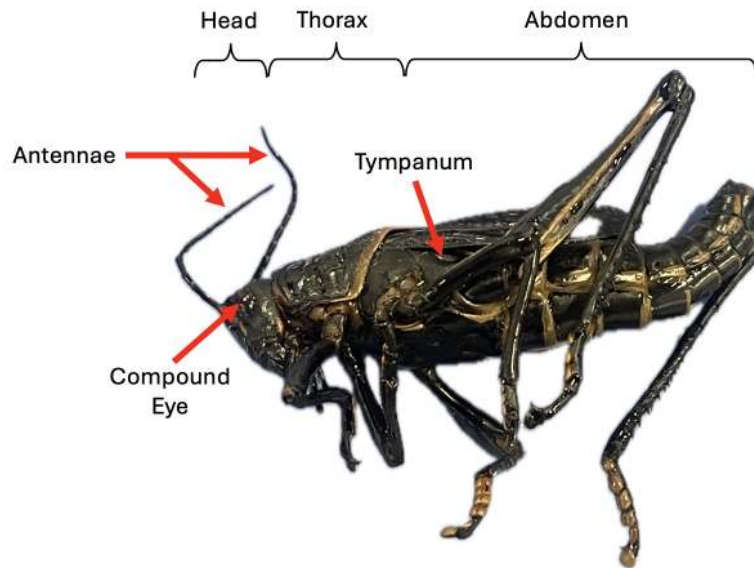


Figure 13.3.17: Grasshopper attributes

3. Does this animal exhibit cephalization?

Observe the animal's several pairs of **jointed appendages**.

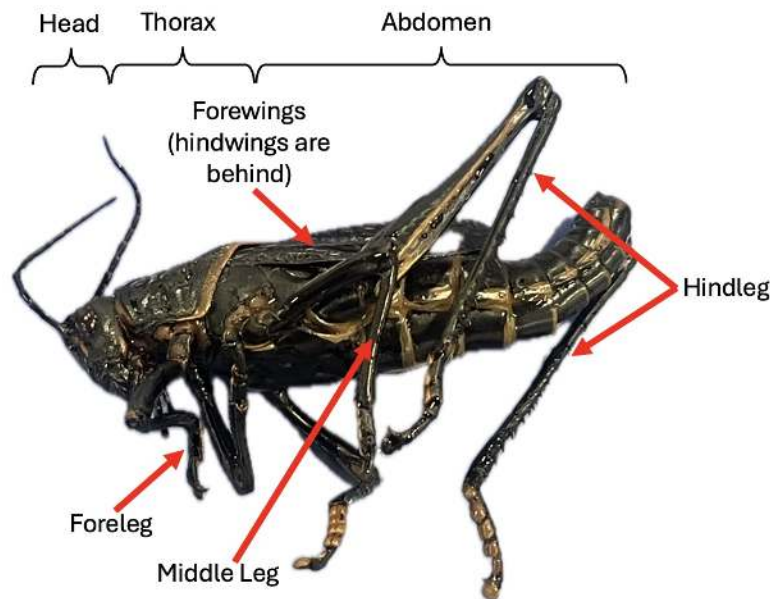


Figure 13.3.18: Additional grasshopper attributes

4. How many sets of appendages do you see?

5. How many segments are present on each limb?

6. Are the appendages adapted for different types of locomotion? How so?

Locate the respiratory structures along the grasshopper's abdomen. These small openings are called **spiracles**, which connect to **tracheae** and are used to bring oxygenated air into the body.

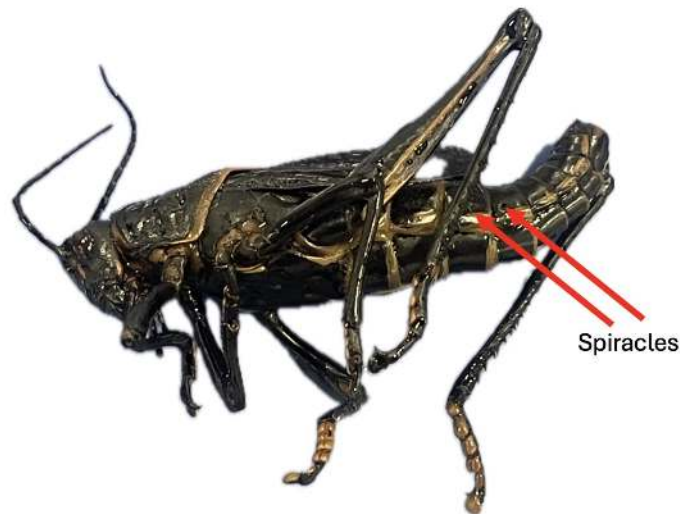


Figure 13.3.19: Grasshopper spiracles

7. Compare and contrast the anatomy of the crayfish and grasshopper in the table below.

Question	Crayfish	Grasshopper
What type of sensory organs does the animal have?		
Does the animal exhibit cephalization?		
Does the animal have a segmented body plan?		
What organ does the animal use for respiration?		
What type(s) of locomotion does the animal use?		
What are the reproductive organs of the animal called?	Male = Female =	Male = Female =

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CHAPTER OVERVIEW

Laboratory 14: Vertebrates

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[14.3: Mammal \(Rat\) Dissection](#)

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14.1: Introduction to Laboratory 14

Dissection is one of the most engaging and insightful ways to explore the inner workings of living organisms. In this lab, we will investigate the anatomy of two vertebrates—a **frog (amphibian)** and a **rat (mammal)**—to observe how different animals share a common structural blueprint despite leading very different lives.

Although frogs and rats may look quite different on the outside, both are members of the vertebrate group and share many **homologous structures** inherited from a common ancestor. By carefully examining both external and internal anatomy, students will develop a deeper understanding of how structure relates to function and how evolution shapes the body plans of animals to suit their environments.

Through side-by-side dissection and observation, students will:

- Identify and compare major **organ systems** (digestive, circulatory, respiratory, etc.).
- Observe **homologous structures** and consider how form and function have diverged over time.
- Gain hands-on experience with anatomical terminology, scientific observation, and dissection techniques.

This lab is not just about identifying organs—it's about seeing the connections between species, understanding evolutionary adaptations, and appreciating the complexity and unity of life.

Questions to consider while completing this lab:

- What structures do frogs and rats have in common?
- How are similar organs adapted to support different lifestyles?
- What can these comparisons tell us about the evolutionary relationships between amphibians and mammals?

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14.2: Amphibian (Frog) Dissection

Objective/Purpose:

To observe and identify the major external and internal anatomical features of a frog as a representative model of amphibian systems (which you will later compare to mammalian anatomy).

Materials Needed:

- Preserved frog specimen
- Dissection tray and pads
- Dissection tools: scalpel, scissors, forceps, probe, pins
- Gloves, a lab coat, and safety goggles
- Disinfectant and cleanup materials
- Lab notebook for diagrams and notes

Caution

- Always wear gloves and goggles when handling specimens.
- Handle the scalpel and scissors carefully.
- Be respectful—this is a real animal that was once alive.
- Dispose of waste properly and clean tools thoroughly afterward.

Amphibian (Frog) Dissection Directions:

Step 1: Identify External Features

Place the frog on the dissection tray, observing both its dorsal and ventral sides.

Observe the following:

- **Head, eyes, tympanic membranes** (eardrums)
- **Mouth, nostrils/external nares**
- **Forelimbs** (front legs, with four toes) and **hindlimbs** (with five toes and webbing)
- **Cloaca** (opening for waste and reproductive fluids)
- **Sex:**
 - Males: may have enlarged thumb pads or vocal sacs
 - Females: typically larger body size; visible eggs if dissected

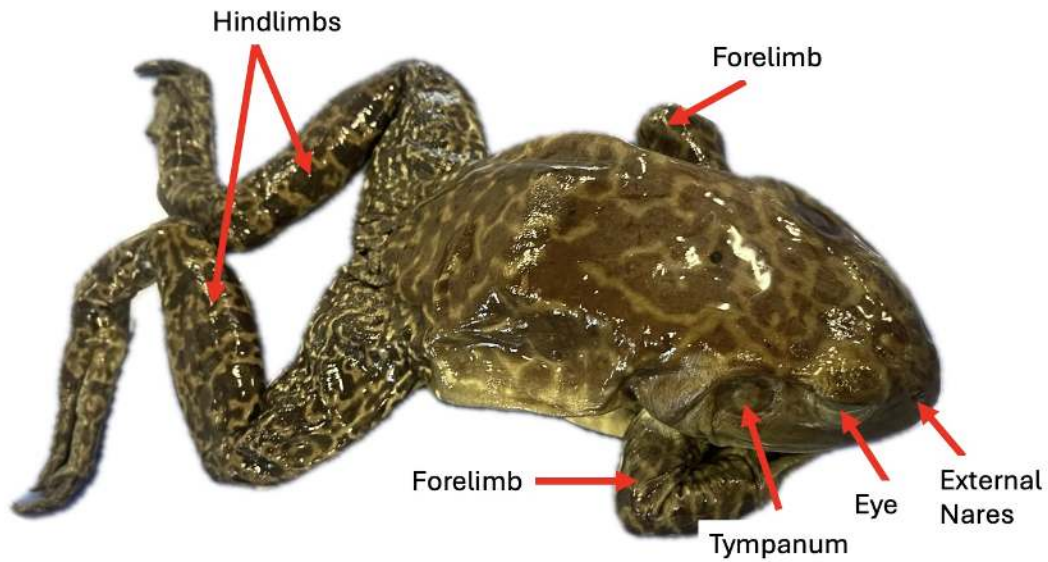


Figure 14.2.1: Frog attributes

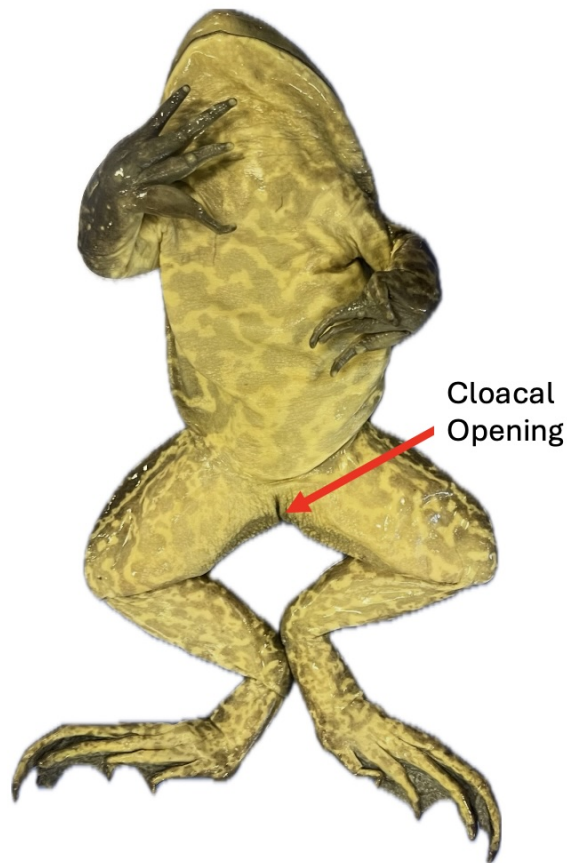


Figure 14.2.2: A frog's cloacal opening.

Step 2: Make the First Incision

- Place the frog in the dissection tray, ventral side up (belly up)
- Use scissors to make a shallow cut through the skin from the cloaca up to the chin.
- Make lateral cuts near the hind legs and arms to form flaps.
- Peel back and pin the skin open (or remove it).



Figure 14.2.3: Making the first incision.

Step 3: Open the Muscle Wall

- Carefully cut through the abdominal muscles along the same midline path.
- Make lateral cuts and pin back the muscle flaps to reveal the internal organs.



Figure 14.2.4: Opening the muscle wall.

Step 4: Examine the Digestive System Organs

- **Liver:** Large, dark, multi-lobed (easily mistaken for lungs; lungs are smaller and more posteriorly located)
- **Gallbladder:** A small green sac under the liver
- **Stomach:** The curved organ on the left side
- **Small intestine:** A long, coiled tube
- **Large intestine (colon):** Leads to the cloaca
- **Fat bodies:** Yellowish, finger-like structures

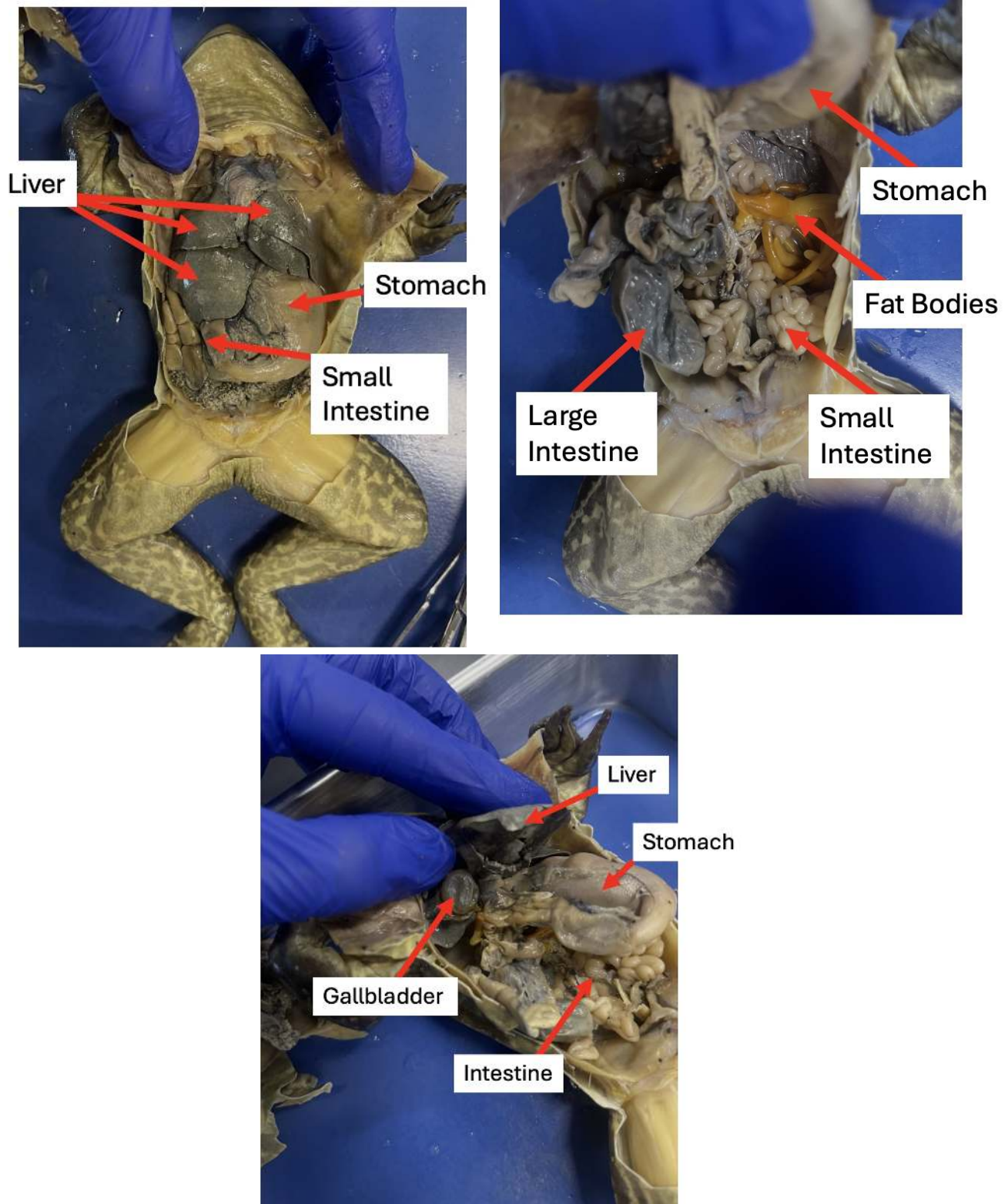


Figure 14.2.5: The digestive system organs of a frog.

Step 5: Examine the Respiratory and Circulatory Systems

- **Lungs:** Small, spongy sacs on either side near the back
- **Heart:** A triangular structure with 3 chambers (2 atria, 1 ventricle) located above the liver
- **Blood vessels:** May be faintly visible; use the probe to trace

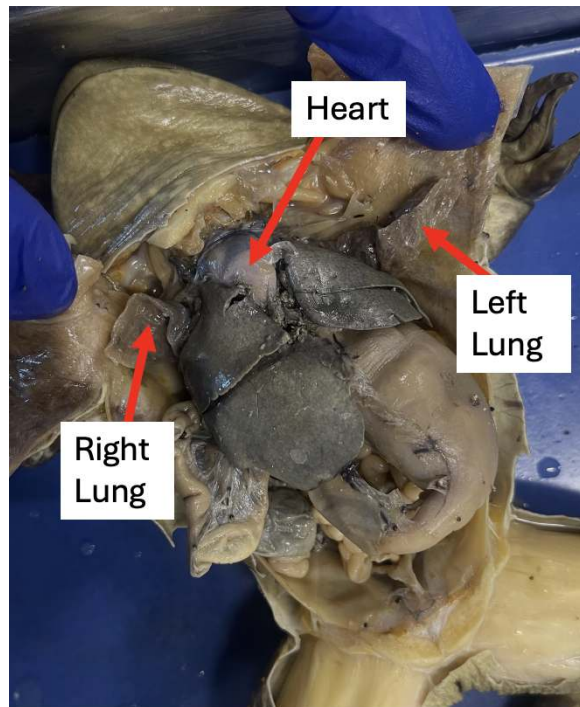


Figure 14.2.6: The respiratory and circulatory systems of a frog.

Step 6 (Optional): Examine the Urogenital System

- **Kidneys:** Flat, dark organs near the spine
- **Bladder:** A small sac near the cloaca

Male:

- Testes: Small, white organs above the kidneys
- Sperm ducts: May run along kidney sides

Female:

- Ovaries: May contain black-and-white speckled eggs
- Oviducts: Long, coiled tubes

Step 7: Lab Skills Check (Part 1)

- Ensure everyone in your group can identify all digestive, respiratory, and circulatory structures.
- Call your instructor over. Each member of the group will be asked to identify organs at random to earn points for the skills check.

Do not dispose of your frog just yet! Keep it to observe during mammal (rat) dissection to compare structures/locations.

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14.3: Mammal (Rat) Dissection

Objective/Purpose:

To observe and identify the major external and internal anatomical features of a rat as a representative model of mammalian systems.

Materials Needed:

- Preserved rat specimen
- Dissection tray and pads
- Dissection tools: scalpel, scissors, forceps, probe, pins
- Gloves, lab coat, and safety goggles
- Disinfectant and cleanup materials
- Lab notebook for diagrams and notes

Caution

- Always wear gloves and goggles when handling specimens.
- Handle the scalpel and scissors carefully.
- Be respectful—this is a real animal that was once alive.
- Dispose of waste properly and clean tools thoroughly afterward.

Definitions: Directional Terms

Anterior = toward the head

Posterior = toward the tail

Dorsal = back side

Ventral = belly side

Mammal (Rat) Dissection Directions:

Step 1: Identify External Features

- Lay the rat **ventral side up** (belly facing up) on the dissection tray.
- Pin the limbs gently to the tray for stability if necessary.
- Examine and note:
 - Head, snout, ears, eyes, tail, and whiskers (vibrissae)
 - Limbs and digits (forelimbs and hindlimbs)
 - External genitalia (to determine sex)

Step 2: Make the First Incision

- Using scissors, make a **shallow cut** just above the genital opening and cut up the **midline** toward the throat.
- Cut through the **skin only**—not the muscle layer yet.

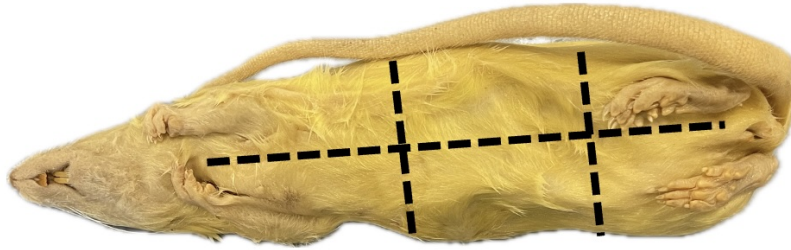


Figure 14.3.1: Position of incisions.

Step 3: Peel Back the Skin

- Use forceps and a probe to separate skin from the muscle layer.
- Make **lateral cuts** across the chest and pelvis and pin back the skin flaps.

Step 4: Open the Abdominal Cavity

- Carefully cut through the abdominal muscles along the same midline path.
- Avoid puncturing underlying organs.
- Gently pull and pin back muscle flaps to expose the internal organs.



Figure 14.3.2: Open abdominal cavity.

Step 5: Identify Major Abdominal Organs

- Begin identifying and observing:
 - **Liver:** Large, dark brown, just under the diaphragm
 - **Stomach:** A curved sac under the liver
 - **Small intestine:** Long, coiled
 - **Large intestine:** Shorter, wider; leads to the anus
 - **Spleen:** Small, dark structure near the stomach
 - **Kidneys:** Toward the back wall of the cavity, bean-shaped

- **Bladder:** A small sac in the lower pelvic region

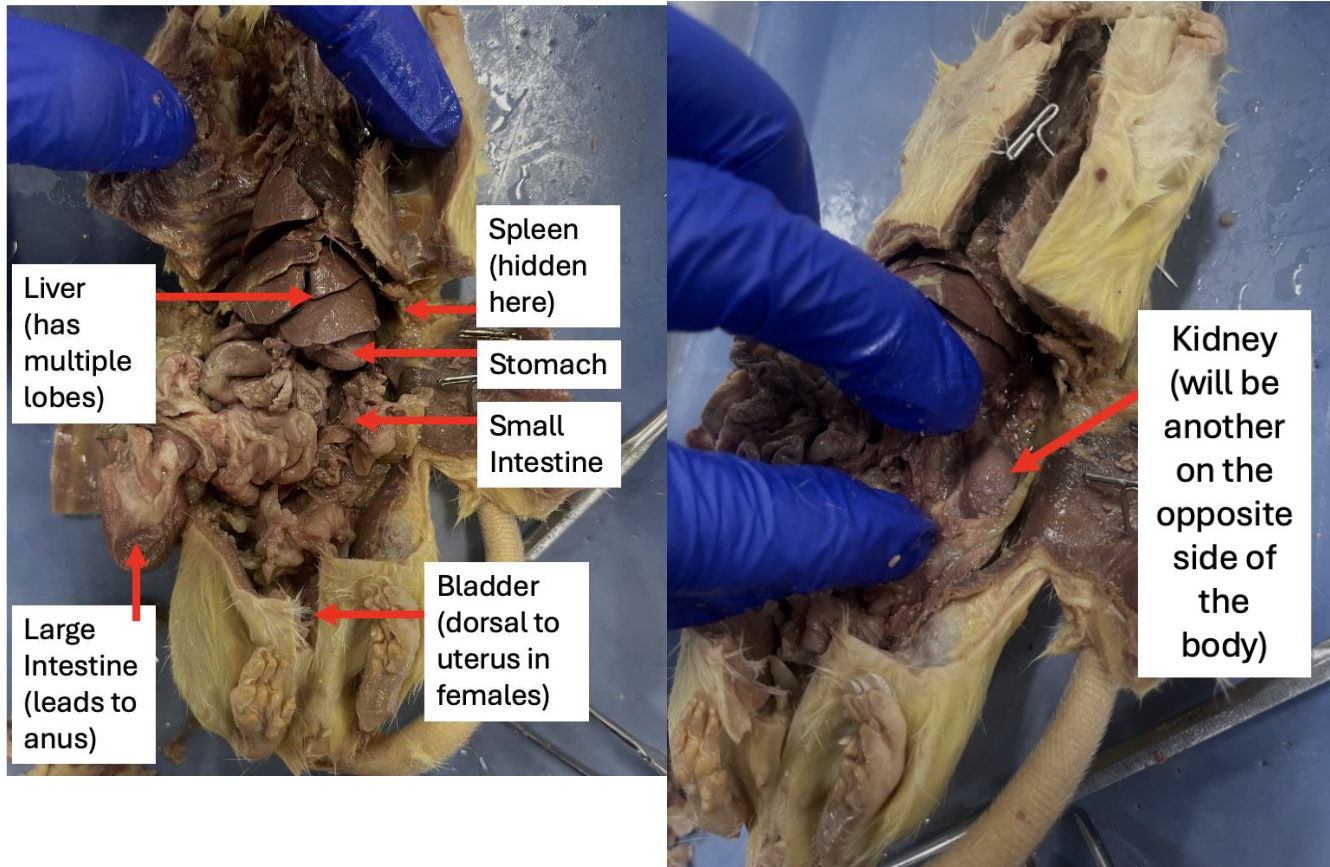


Figure 14.3.3: Major abdominal organs of a rat.

Step 6: Examine the Thoracic Cavity

Cut through the **ribs** on either side using scissors to expose:

- **Heart:** Between the lungs, in the pericardial sac
- **Lungs:** On either side of the heart, spongy tissue
- **Trachea:** Windpipe, transports air toward the lungs
- **Diaphragm:** A thin muscle separating the thoracic and abdominal cavities

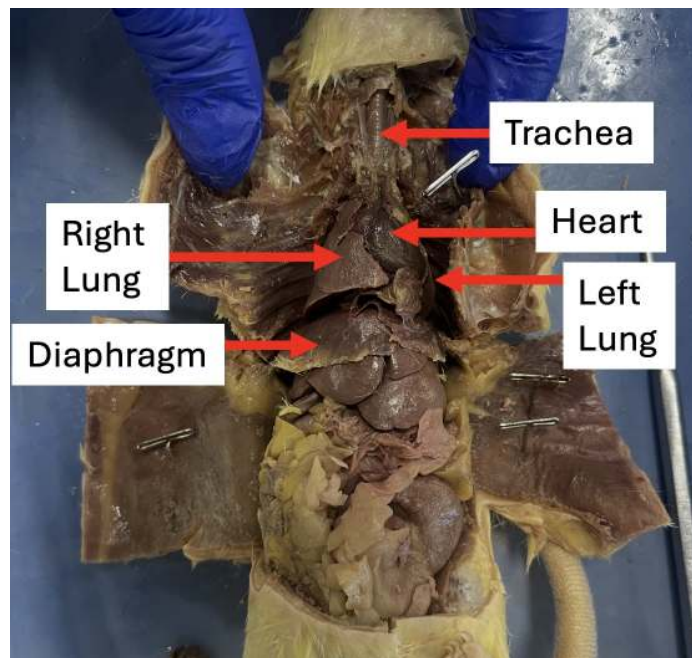


Figure 14.3.4: Respiratory and circulatory organs of a rat.

Step 7: Lab Skills Check

- Ensure everyone in your group can identify all of the organs in the thoracic and abdominal cavities.
- Call your instructor over. Each member of the group will be asked to identify organs at random to earn points for the skills check.

Step 8: Clean Up

- Remove pins and dispose of specimens properly.
- Clean and dry dissection tools.
- Clean/disinfect the dissection tray and work area.
- Wash hands thoroughly.

Follow-Up Questions

1. How does the skin of a frog differ from the skin of a rat? What are the functional differences between these types of skin?
2. Frogs and rats are both vertebrates. What does this tell you about their evolutionary relationship?
3. How do the limbs of a frog compare to those of a rat? How are their limbs suited for their respective lifestyles?
4. Amphibians have a three-chambered heart, while mammals have a four-chambered heart. What are the advantages of a four-chambered heart?
5. Describe how the lungs of amphibians and mammals differ. How does this reflect differences in lifestyle and habitat?
6. How does the length and complexity of the digestive tract in frogs compare to that of mammals?
7. Identify at least three homologous structures in the frog and the mammal. What do these similarities tell you about their evolutionary history?
8. What anatomical features of the frog show adaptations to an amphibious life?
9. What features of the mammal show adaptations to terrestrial life?
10. What are some characteristics unique to mammals?

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