GAVILAN COMMUNITY COLLEGE

# Principles of Biology Lab Manual

## Biology 10

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# Introduction

Welcome to the **Lab Manual** for **Principles of Biology** (Bio 10) at Gavilan College! The lab exercises in this Lab Manual will correlate closely with lecture topics. We will begin with the **basics** and learn about the scientific process, using microscopes to view tiny organisms, how cells eat, and why they are as small as they are. After these introductory labs, we will take a closer look at key cellular processes: how enzymes work, how cells acquire energy, and how cells divide. The next few labs are about how organisms inherit traits from their parents, and how an organism's genetic makeup leads to these traits. Finally, the course ends with a few labs on evolutionary biology and ecology.

Each lab exercise begins with a list of objectives. These **objectives** describe what you should be able to do through completing that lab exercise. After the objectives is the **Introduction** that states the purpose of the lab and provides relevant background information. The **Materials & Methods** section describes the materials and protocols used to complete the lab activities. This section contains questions you will answer as you do the lab work.

Labs are a valuable learning experience in many ways:

1. They allow you to learn to organize and follow protocol to conduct an experiment.
2. They provide you with hands-on experience to make concepts more clear.
3. Labs can actually be fun! Actively participate and get to know your classmates.

In order to do well in and learn from the lab portion of this course, you must put in time **before, during, and after** lab sessions.

**Before lab:**

- Read the lab!
- Jot down some notes while you read
- Complete any pre-lab work your instructor may assign

**During lab:**

- Show up, do the lab work, and turn in your lab! (your instructor may not allow you to make up a missed lab)
- Take notes during lab lecture and discussion
- Ask questions if you are unclear on something

**After lab:**

- Think about the lab! This is a crucial part of connecting the lab work you did with the concepts the lab deals with.
- Complete any post-lab work your instructor may assign

Thank you for your attention. Let's have a great semester!

Patrick Yuh

# Lab Safety

It is necessary for you to follow a few rules in order to ensure the safety of students and the proper care of lab equipment. The following are basic lab rules (please especially note **the ones in bold**):

- **Do not eat, drink, or smoke during a lab exercise.**
- Report any spills to your instructor. Some chemicals must be discarded in a special manner.
- If you accidentally get a chemical in your mouth, spit out the chemical, rinse your mouth with water, and inform your instructor.
- Wash your hands before leaving lab.
- Wear safety glasses if heating solutions with glassware and a Bunsen burner, or if instructed to do so.
- Wear gloves and/or a lab coat if instructed to do so.
- Note the location of the fire extinguisher, eye wash, and first aid kit.
- **Follow lab protocol exactly unless instructed otherwise.**
- **Treat your work area like a lab space, not a desk. Keep it free of unused items.**
- Before leaving lab, thoroughly wash (with lab detergent and brush if needed) and rinse ALL lab ware and place in drying racks.
- **Return things from where you got them, usually the lab cart or a station within the room.**
- Use equipment only according to specified instructions unless instructed otherwise.

# LAB 1 The Scientific Process

## Objectives

At the end of the lab, you should be able to:

1. Describe the steps in the scientific process.
2. Explain what characterizes a question that can be answered by the scientific process, and identify these types of questions.
3. Explain what characterizes a good scientific hypothesis, and identify scientific hypotheses.
4. Define and give examples of independent, dependent, and standardized variables.
5. Identify the variables in an experiment.
6. Explain what control treatments are and why they are used.
7. Explain what replication is and why it is important.

## Introduction

The scientific process (also known as the scientific method) is an approach to problem solving and learning more about the world in which we live. This process includes distinct steps: asking questions and proposing answers; designing and doing experiments; interpreting data to refine the proposed answer. The more experiments are done, the more data can be gathered to support the proposed answer.

These are the overall steps of the scientific process:

1. **Observe** the world around you
2. Ask a **question** about something you observe
3. Propose an **answer** to your question – this is called a **hypothesis**
4. Make testable **predictions** assuming your hypothesis is correct
5. Design and do an **experiment** to **gather data** that may support or refute your hypothesis
6. **Refine** your hypothesis and do **more experiments** to gather more data
7. **Communicate** your findings to your peers in the scientific community

**Figure 1.1** on the next page shows these steps visually. Note that it is not identical to the list of steps above, but describes the same process. The practice of science is an ongoing process; thus, there is no real "last step". There are always more questions to ask, more hypotheses to formulate, and more experiments to do.

In this lab, you will become familiar with the steps of the scientific process by

1) performing a short investigation, and
2) focusing on three steps of the process: the question; the hypothesis; and the experiment.

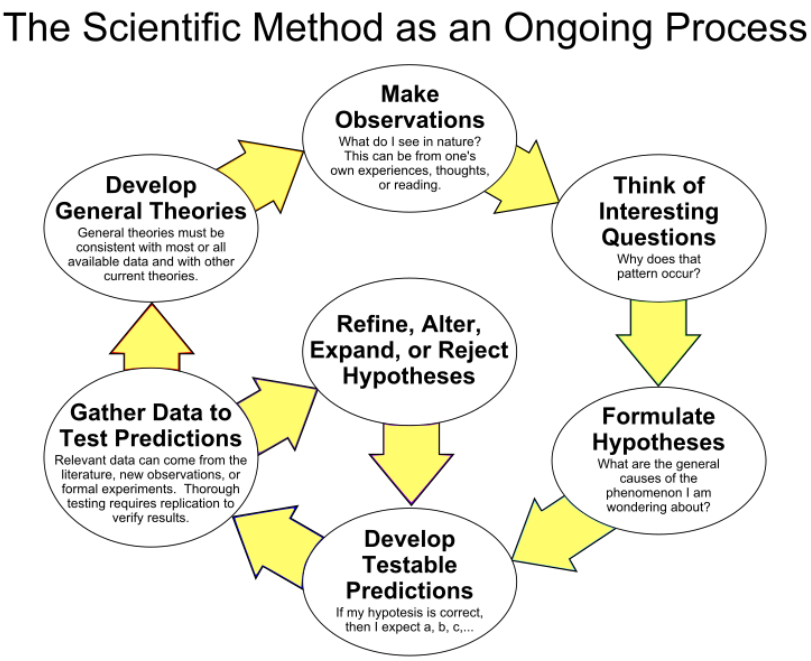


**Figure 1.1. The steps of the scientific process.**

## Materials & Methods

### ACTIVITY 1: THE "BLACK BOX" INVESTIGATION

**Materials**

- one sealed box
- one empty box
- one bag of objects

The sealed box contains one or more of the objects in the bag. Your goal: figure out what is inside this box **without** opening it. You may open the empty box and take out the objects in the bag.

Usually there is a procedure for you to follow. For Activity 1, you come up with your own procedure as you work through the activity.

Gather the materials for this activity. Do not shake the sealed box as you bring it to your seat.

1. State the **question** you are asking in this activity.

2. Before you do anything, make a guess about what the **answer** is.

3. You will need **evidence** from an **experiment** to support or refute your answer. What experiment can you do to **test** whether your answer is correct? List the steps you will take in your experiment.

4. **Perform your experiment** to test your answer. If your answer from #2 seems incorrect, **revise** it and **repeat** the experiment. Keep doing this until you think you have the right answer, and write it here.

5. Experiments can work well. Which objects do you know for sure are not in the sealed box, and why?

6. Experiments are rarely perfect. What are the **limitations** of your experiment on getting the right answer?

7. Can you prove your final answer is right without opening the box? Why or why not?

8. Do the objects in the bag have properties other than sound that you cannot detect through your experiment?

9. You may now open the sealed box by pulling on the loose tab. Was your final answer right or wrong? If wrong, how did your experiment lead you astray?

## ACTIVITY 2: ASKING A SCIENTIFIC QUESTION

Every scientific investigation begins with an **observation** that leads to a **question** the scientist wants to answer. However, just because a question ***can*** be answered doesn't mean it can be answered ***scientifically***. Consider this question: *Do more people behave immorally when there is a full moon than at other times of the month?* The phase of the moon is ***well-defined*** and ***measurable***, but morality is not. Thus, no experiment can be performed to measure morality. Also, questions that lack ***objective*** answers are not considered scientific.

**How can you tell whether a question is scientific?**

**Discuss the following questions with your group. Are they scientific questions (can they be answered by the scientific process)? Write yes or no, and explain briefly.**

1. What is in the sealed box from Activity 1?

2. Are serial killers evil by nature?

3. Why is grass green?

4. What is the best recipe for chocolate cookies?

5. How do you get the maximum yield from a peanut plant?

6. Does watching television cause children to have shorter attention spans?

**ACTIVITY 3: PROPOSING A SCIENTIFIC ANSWER (i.e. HYPOTHESIS)**

After asking a scientific question, the next step is to answer it. A **hypothesis** is simply an answer to a question. It is sometimes described as an educated guess. As with scientific questions, scientific hypotheses have certain properties.

A good hypothesis is **specific**.

- The hypothesis should naturally lead to a single experiment, not several.
- Include more detail to make your hypothesis more specific.
- Example: for the question *"Why is my phone not working?"*, "the battery is dead" is a more specific hypothesis than "one of the parts is broken". The first hypothesis leads to one experiment, while the second hypothesis leads to many.

A good hypothesis is **reasonable**.

- The hypothesis should be consistent with what we already know is true.
- Example: for the same question as before, "my phone is possessed by demons" is not a reasonable hypothesis because demons have not been objectively shown to exist.

A good hypothesis is **testable**.

- The hypothesis must allow for experimental evidence to be gathered to test whether it is correct.
- Example: for the question *"Is there life on other planets?"*, "there are other inhabited planets in the universe" is a testable hypothesis. There are ways to find out if life exists on other planets (exploration rovers like Spirit and Opportunity on Mars, radio telescopes receiving signals).

A good hypothesis is **falsifiable**.

- The hypothesis must be able to be proven false.
- Example: for the same question as before, "there are other inhabited planets in the universe" is testable but NOT falsifiable. There is no way to test every planet in the universe for life.

We ***can prove*** a hypothesis ***false*** with experimental evidence that does NOT support the hypothesis. But we **cannot prove** a hypothesis **true** with experimental evidence that DOES support the hypothesis.

In Activity 1, you may have found that your hypothesis was wrong despite accurate observations and careful experimentation. Your hypothesis reflected the best evidence available *at the time*, from one particular investigation. Scientific knowledge is thus an accumulation of evidence in support of hypotheses; it cannot be regarded as absolute truth. Hypotheses are the best answers at the time, and future investigations may be able to prove a hypothesis false.

**What are the properties of a good scientific hypothesis?**

**Can a hypothesis be proven true?**

**Can a hypothesis be proven false?**

**Discuss the following questions with your group. Are they good scientific hypotheses? Write yes or no, and explain briefly.**

1. Plants absorb water through their leaves as well as their roots.

2. Mice require calcium for developing strong bones.

3. Dogs are happy when you feed them steak.

4. An active volcano can be prevented from erupting by throwing a virgin in during each full moon.

5. The higher the intelligence of the animal, the more easily it can be trained.

6. The earth was created by an all-powerful being.

7. HIV (human immunodeficiency virus) can be transmitted by cat fleas.

When a hypothesis continues to be supported by massive amounts of experimental evidence, it gets "promoted" and is regarded as a **scientific theory**. Be careful with this term! A scientific theory is NOT the same as an everyday theory. In fact, when people say they have a theory in casual conversation, they actually mean they have a hypothesis. Scientific theories are often treated as facts since they are so widely supported. Examples of scientific theories include the theory of evolution by natural selection, the cell theory, and the Big Bang theory.

A **scientific law** is related to a scientific theory. Laws describe *what* happens, while theories attempt to explain *why* it happens. Laws are often expressed as math equations or short statements. Examples of laws include the law of gravity, the laws of thermodynamics, and Mendel's laws of inheritance.

## ACTIVITY 4: DESIGNING A GOOD EXPERIMENT

### Part 1 – Variables

With a scientific question and a good scientific hypothesis, it's time to design a good experiment to test the hypothesis. Experiments have three types of **variables**. A variable is simply something whose value can vary. Let's use a question from Activity 2 to learn about variables: ***How do you get the maximum yield from a peanut plant?*** In order to answer this question with supporting evidence, the experiment you do will involve growing multiple peanut plants in slightly different conditions and measuring them in some way. So, what do you ***change***, what do you ***measure***, and what do you ***keep the same***?

**Independent variables** are what the investigator ***intentionally changes*** in the experiment. One hypothesis for this question might be: ***Peanut plants have a higher yield when grown in 8 hours of sunlight a day than 4 hours.*** To test this hypothesis, you would have to provide at least one plant 8 hours of sunlight a day, and at least one plant 4 hours of sunlight a day.

1. In this experiment, what is the independent variable?

2. List three other examples of independent variables that could be tested.

**Dependent variables** are what the investigator ***measures*** in the experiment. They are called dependent because their value depends on the value of independent variable. In our example, the question asks about ***maximum yield*** of a peanut plant.

3. List three properties of peanut plants that could be measured as dependent variables.

**Standardized** or **control variables** are what the investigator ***keeps the same*** in the experiment. This way, any changes in the dependent variable can be attributed *exclusively* to the independent variable.

4. In the experiment from before, what variables would have to be standardized or controlled?

In summary:

- a good scientific hypothesis leads to an experiment where the investigator intentionally changes one (and only one) independent variable
- this leads to a change in one (or more) dependent variables which the investigator measures
- all other potential variables must be kept constant as standardized variables

5. Why should an investigator have only one independent variable per experiment?

6. In Activity 1, what was the independent variable and the dependent variable(s)?

**Discuss the following experiments in your group. Circle the independent variable and underline the dependent variable.**

7. Height of bean plants given different levels of nitrogen fertilizer is recorded daily for two weeks.

8. Guinea pigs are kept at different temperatures for six weeks. Percent weight gain is recorded.

9. Light absorption by a pigment is measured for red, blue, green, and yellow light.

10. Batches of seeds are soaked in salt solutions of different concentrations, and germination is counted.

11. An investigator hypothesizes that the adult weight of a dog is higher when it has fewer littermates.

**Part 2 – Control Groups**

A good experiment also includes control groups and experimental groups. A **control group** is a group in which ***the independent variable is either eliminated or set to a standard value***. The control group provides a standard for comparison to the experimental group, which allows the investigator to determine how the independent variable is affecting the dependent variable.

Example: In #8 above, the investigator must compare plant growth with fertilizer to plant growth with no fertilizer. Plants that get no fertilizer make up the control group (the independent variable is ***eliminated***).

Example: In #9 above, temperature is the independent variable. Since it cannot be eliminated (you can't raise a guinea pig in "no temperature"), it is ***set to a standard value*** for the control group (typical room temperature).

Discuss the following experiments in your group. Describe an appropriate control group for each.

12. An investigator studies the amount of alcohol produced by yeast when it is grown with different types of sugars.
    Control group:

13. The effect of light intensity on photosynthesis is measured by collecting oxygen produced by the plant.
    Control group:

14. The effect of NutraSweet sweetener on tumor development in laboratory rats is investigated.
    Control group:

15. Subjects are given squares of paper that has been soaked in a bitter-tasting chemical. The investigator records whether each person can taste the chemical.
    Control group:

**Part 3 – How Reliable Are Your Results?**

A poorly designed experiment can get just about any kind of result imaginable. Let's wrap up our discussion of good experiments with two related topics: sample size and reproducibility.

In general, larger sample sizes are better than smaller ones. **Sample size** is simply how many things are included in the control and experimental groups. In the plant fertilizer example, what if you have four plants, each one gets a different amount of fertilizer, and one of the plants dies? It's a big leap to claim that the fertilizer killed the plant. If you had more plants in each group (say, 20), and one plant in one of the groups died, you would not think it was because of the fertilizer.

After an experiment is completed, it is essential to make sure that repeating the experiment gets the same results. This is known as **reproducibility**. No one will be convinced by one result from one experiment. If the results support a hypothesis, anyone should be able to repeat the same experiment and reproduce the results. In living systems, there is naturally a small amount of variation, so reproducibility does NOT mean getting exactly the same results every time. Rather, it allows us to see how much variation is present and get an average range from multiple trials.

## Extra Questions – your instructor may assign them

1. List the steps of the scientific process and briefly describe the purpose of each in your own words.

2. Come up with two questions of your own that can be answered through the scientific process.

3. For one of your questions above, develop a hypothesis and design an experiment for testing the hypothesis.

    a. Hypothesis:

    b. Describe your experiment:

    c. What is your independent variable?

    d. What is your dependent variable?

    e. What are your standardized variables (list at least three)?

    f. What is your control group?

# LAB 2 Microscopes

## Objectives

At the end of the lab, you should be able to:

1. Identify and describe the parts of a microscope.
2. Safely and properly handle a microscope and microscope slides.
3. Operate a compound light microscope: mount a slide on a microscope stage, adjust the light, and focus on a specimen.
4. Explain the microscopy concepts of field of view, image orientation, and depth of field.
5. Prepare a wet mount slide specimen.
6. Prepare a simple stain slide specimen.

## Introduction

The purpose of this lab is to become familiar with the parts and proper usage of a **compound light microscope**. Magnifying glasses have used lenses to enlarge images for millennia, but microscopes have only been in use since the late 16th century.

The first **compound microscopes** (i.e. microscopes with more than one lens) were believed to have been made in the Netherlands during the 1590s (**Fig. 2.1**). A pair of lenses was encased inside three sliding tubes, with 9x total magnification when fully extended.

In the late 1600s, Anton van Leeuwenhoek, also from the Netherlands, pioneered a **simple microscope** (i.e. a microscope with one lens) that could magnify specimens by 270x (**Fig. 2.2**). He achieved this by carefully grinding and polishing the lenses. He was among the first to see and describe bacteria, yeast, algae, and other organisms on a microscopic level.

Since the days of van Leeuwenhoek, many advances have been made in microscopy. In this class you will use a compound light microscope with a maximum total magnification of 1000x (**Fig. 2.3**). They will allow you to observe cells and even parts of cells, which are too small to see by the naked eye.

**Figure 2.1. A compound microscope similar to those from the 1590s.**

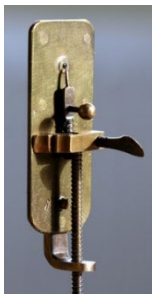

**Figure 2.2. A simple microscope made by van Leeuwenhoek.**

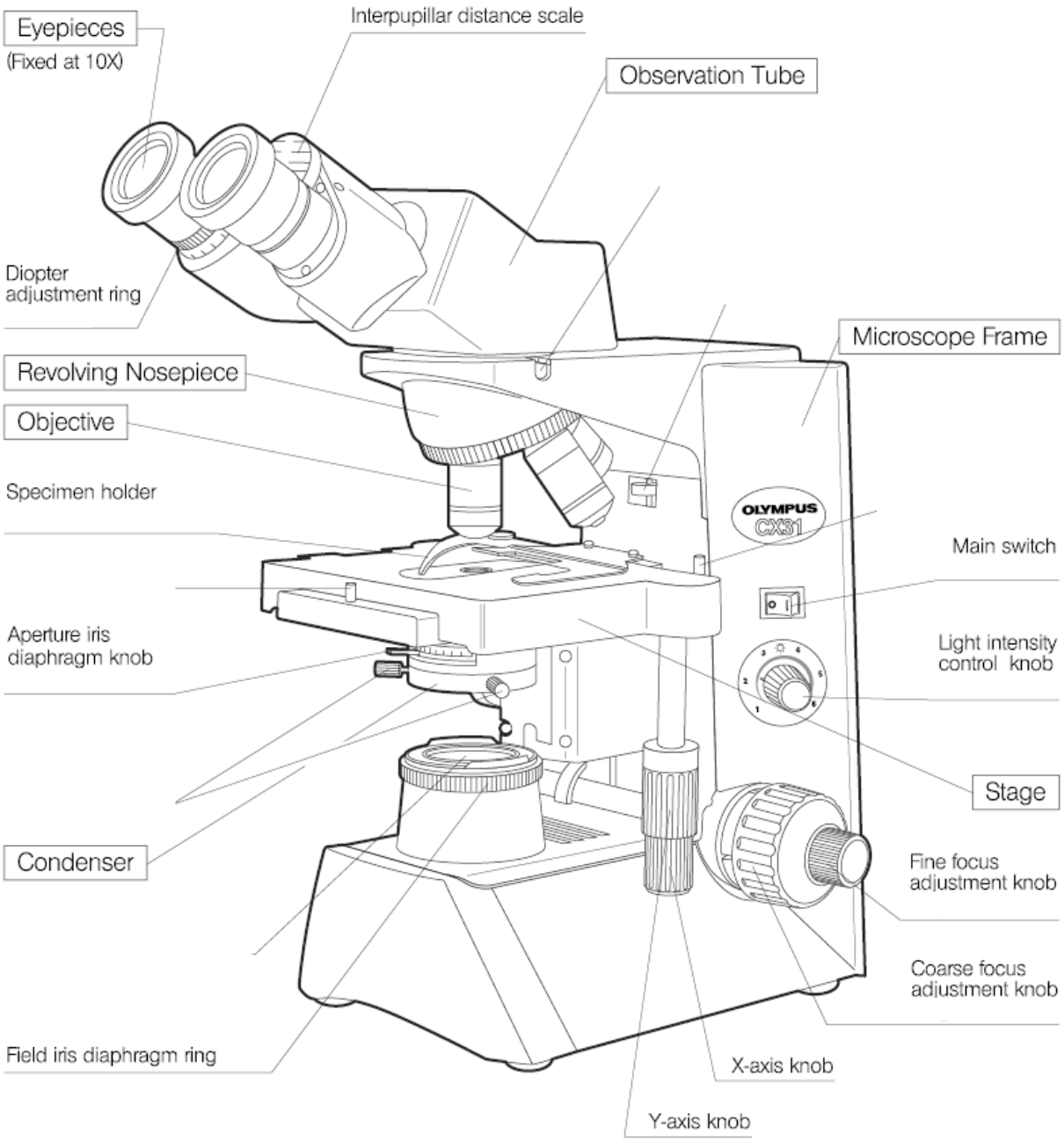


**Figure 2.3. A labeled diagram of the Olympus CX31 microscope (some labels are omitted for simplicity).**

**I want to do one of these three things… *So I should adjust these parts…***

| See through the microscope | Focus on my specimen | Adjust the light |
|---|---|---|
| ***Ocular lenses*** | ***Coarse focus knob (4x obj. ONLY!)*** | ***Dimmer switch*** |
| ***Objective lenses*** | ***Fine focus knob*** | ***Aperture iris diaphragm*** |
| | ***X- and Y-axis knobs*** | ***Condenser*** |
| | | ***Field iris diaphragm*** |

**Table 2.1. A helpful guide to using the microscope.**

## Materials & Methods

**Fig. 2.3** is a labeled picture of the microscope you will use. Refer to it as well as **Table 2.1** for three common microscope objectives, and the relevant parts that should be adjusted to achieve them.

Microscopes are very expensive instruments! The one you are using costs **several thousand dollars** and must be treated respectfully.

### ACTIVITY 1: MICROSCOPE PARTS & MAGNIFICATION

**Materials**

- one microscope

**Microscope Parts**

In this section you will familiarize yourself with the parts of a microscope.

1. The microscopes are located in the cabinet next to your seat. Using both hands, take out a microscope and remove the dust cover.

2. Take a moment to compare **Fig. 2.3** with your microscope. Begin identifying the parts.

3. As we watch this video (http://tinyurl.com/k96uwoh), describe these parts:
   a. aperture iris diaphragm knob

   b. stage

   c. stage clips or specimen holder

   d. X- and Y-axis knobs

   e. coarse focus knob

   f. fine focus knob

   g. revolving nosepiece

   h. objective lens

   i. ocular lens

**Magnification**

There are two lenses that magnify the image of the specimen: the ocular lens and the objective lens. Total magnification = magnification of ocular x magnification of objective.

1. The magnification of the ocular lens is: _____
2. The magnification of the four objective lenses is: _____, _____, _____, _____
3. The total magnification of an object viewed with the lowest power objective is: _____
4. The total magnification of an object viewed with the highest power objective is: _____

## ACTIVITY 2: IMAGE ORIENTATION & FIELD OF VIEW

In this activity you will learn how to set up a microscope slide and focus on the specimen. You will also become familiar with the microscope's field of view and the orientation of the image you see. We will watch this video as we go: http://tinyurl.com/h44b979.

**Materials**

- letter e slide

1. Plug in and turn on your microscope, and obtain a **letter e slide**. Clean it with a Kimwipe if needed.
2. **Mount** the slide on the microscope stage with the stage clips.
3. Adjust the **X- and Y-axis knobs** so that light **shines through** the specimen.
4. Sketch the specimen as it appears on the slide.

Letter e sketch, no magnification

5. **Focus** on the specimen with the 4x objective lens:
   a. Turn the **dimmer** to 2 or 3.
   b. Use the **coarse focus knob** to bring the stage all the way up.
   c. While looking through **both** ocular lenses, **slowly** bring the stage down until the letter comes into focus.
   d. For optimal viewing: adjust the **distance** between the **ocular lenses** to match the distance between your eyes, and the **focus** of the **right ocular lens** with the **diopter adjustment ring**.

Letter e sketch with 4x objective

6. Sketch the specimen while looking through the microscope.

While you are focused on the letter e with the 4x objective, you will see how the microscope image moves when you move the slide. This is known as **image orientation**.

7. Use the **X-axis knob** to move the slide **to the right**. As you do this, look through the microscope. In which direction does the letter e appear to move? _____

8. Use the **Y-axis knob** to move the slide **away from you**. As you do this, look through the microscope. In which direction does the letter e appear to move? _____

9. Handling only the **black rubber ring** of the revolving nosepiece, **rotate** to the **10x objective**.

10. **Refocus** on the letter e using **ONLY** the **fine focus knob**. Adjust the **dimmer** as needed.

11. Sketch the specimen while looking through the scope.

Letter e sketch with 10x objective

The **field of view** is the area you can see while looking through the microscope. When you went from the 4x to the 10x objective, did the field of view get larger or smaller? _____

12. **Switch back** to 4x, **remove** the slide from the stage, and **return** it to the **slide tray**.

## ACTIVITY 3: DEPTH OF FIELD & FOCUSING

In this activity you will see how the magnification affects the **depth of field**, and how **focusing** lets you see different slices of a specimen.

**Materials**

- colored threads slide

1. Obtain a **colored threads slide**. This slide has three threads layered on top of each other.

2. Focus on your specimen with the **4x objective**, like you did before. At 4x, can you focus on all three colored threads at once? _____

3. Focus on your specimen with the **10x objective**, like you did before. At 10x, can you focus on all three colored threads at once? _____

4. Sketch the specimen while looking through the scope.

Threads sketch with 10x objective

When you focus on a specimen, you are seeing a thin horizontal slice of it. This slice has a certain thickness, which is called the **depth of field**. As you increase the magnification, does the depth of field get thicker or thinner?

_____

## ACTIVITY 4: LIVE SPECIMENS – WET MOUNTS

Now that you are familiar with how to use and handle the microscope, it is time to look at some more interesting specimens.

**Materials**

- microscope slides
- coverslips
- Protoslo
- tape
- *Amoeba* culture
- *Paramecium* culture
- *Rotifer* culture
- *Blepharisma* culture
- *Elodea* plant
- *Rhizopus* plate

The **wet mount** is an easy way to prepare a live specimen for viewing. The steps are the same for any organisms already in water (the four cultures in the Materials list):

1. Place one drop of the culture onto the center of a clean slide.

2. Add one or two drops of Protoslo to slow down the organisms.

3. Place a coverslip on top of the specimen. To avoid getting air bubbles, place one side of the coverslip down at an angle so the underside gets wet, then let the coverslip fall onto the specimen.

4. View your specimen under the microscope. Start with the 4x objective. You will need to look around! Organisms tend to gather near the edge of the coverslip, but can be elsewhere too.

5. When you have found and focused on an organism, switch to the 10x objective. These are live, mobile organisms, so you may have to chase after them. If you wish, switch to the 40x objective.

**Sketch your specimens below. If you are familiar with the parts, label them on your sketches.**

*Amoeba* sketch with _____x objective

Parts: cell membrane; cytoplasm; nucleus; pseudopod

*Rotifer* sketch with _____x objective

Parts: body; mouth; cilia

*Paramecium* sketch with _____x objective

Parts: cell membrane; cytoplasm; food vacuole; nucleus; cilia

*Blepharisma* sketch with _____x objective

Parts: cell membrane; cytoplasm; food vacuole; nucleus; cilia

Preparing a wet mount of *Elodea* is very similar:

1. Place one drop of water onto the center of a clean slide.

2. Use tweezers to remove one *Elodea* leaf and place it in the drop of water.

3. Place a coverslip on top of the specimen. To avoid getting air bubbles, place one side of the coverslip down on the slide. Move it towards the liquid until the underside is wet, then let the coverslip fall onto the specimen.

4. View your specimen under the microscope. Start with the 4x objective. You will see many cells at once. **Make a first sketch below.** If you are familiar with the parts, label them on your sketches.

5. When you have found and focused on some cells, switch to the 10x objective. Focus on a single cell and **make another sketch with the 10x or 40x objective.**

| *Elodea* sketch with 4x objective | *Elodea* sketch with _____x objective |
|---|---|
| Parts: cell wall; chloroplast; central vacuole | Parts: cell wall; chloroplast; central vacuole |

6. The leaf is made up of many layers of cells, one on top of another (like the colored threads). Adjust the fine focus knob and you will see your cells go out of focus, and other cells come into focus. You have just moved the focus from one layer to another layer. Keep adjusting the focus until there are no more cells. Now go back the other way, and count how many cell layers you see in the leaf.

   How many layers of cells are in the leaf? _____

Preparing a wet mount of *Rhizopus* does not require water:

1. Remove the lid from a *Rhizopus* plate. Use a piece of tape to grab some *Rhizopus*.

2. Place the tape on a slide. Do not add a coverslip.

3. View your specimen under the scope. Start with the 4x objective. You will see many cells at once.

4. When you have found and focused on some cells, switch to the 10x objective. **Make a sketch with the 10x or 40x.** If you are familiar with the parts, label them on your sketch.

| *Rhizopus* sketch with _____x objective |
|---|
| Parts: spore; sporangium; sporangiophore |

## ACTIVITY 5: LIVE SPECIMENS – SIMPLE STAINS

Most cells have no color, which makes them difficult to see under a microscope. Applying a stain will make it easier to see them. In this activity you will stain cheek cells with methylene blue, and potato cells with iodine.

**Materials**

- microscope slides
- coverslips
- toothpick
- cheek cells
- methylene blue
- knife
- potato
- iodine

**Cheek cells**

1. Place one drop of water onto the center of a clean slide.
2. Gently **scrape** the inside of your cheek with a toothpick.
3. **Stir** the toothpick in the drop of water so that it becomes cloudy.
4. **Add one drop** of methylene blue.
5. Place a coverslip on top of the specimen.
6. Soak up excess liquid by touching a Kimwipe to the edges of the coverslip.

Cheek cells sketch with _____x objective

Parts: cell membrane; nucleus; cytoplasm

7. View your specimen under the scope. Start with the 4x objective. You should see many cells.
8. When you have found and focused on some cells, switch to the 10x objective. **Make a sketch with the 10x or 40x.** If you are familiar with the parts, label them on your sketch.
9. Dispose of the slide in the Bleach Water container at the back of the room.

**Potato cells**

1. Place one drop of water onto the center of a clean slide.
2. Obtain a paper-thin slice of potato, the thinner the better.
3. Place the potato slice in the drop of water on the slide.
4. Place a coverslip on top of the specimen.
5. View your specimen under the scope. Start with the 4x objective. You should see many cells.
6. When you have found and focused on some cells, switch to the 10x objective. **Make a sketch with the 10x or 40x.** If you are familiar with the parts, label them on your sketch.

| Potato cell sketch with _____x objective<br>**before iodine**<br><br>Parts: cell wall; amyloplast; vacuole | Potato cell sketch with _____x objective<br>**after iodine**<br><br>Parts: cell wall; amyloplast; vacuole |
|---|---|

7. Leave the slide on the stage and do not adjust the focus. Carefully add one drop of iodine next to one edge of the coverslip. The iodine should seep under the coverslip and stain the cells. Ask your instructor for help if you need!

8. Now look at the cells again and **make another sketch**. If you do not see the staining, add another drop of iodine.

9. Dispose of the slide in the Bleach Water container at the back of the room.

***When you are finished looking at all the specimens, put away your microscope properly!***

- *remove slide*
- *switch to 4x objective*
- *bring stage all the way down and forward*
- *turn dimmer all the way down*
- *power off*
- *unplug and wrap cord neatly around back*
- *put on dust cover*
- *return to cabinet with both hands*
- ***if you have not returned your microscope properly, you will suffer the consequences***

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# LAB 3 Cell Diffusion & Osmosis

## Objectives

At the end of the lab, you should be able to:

1. Explain the processes of diffusion and osmosis.
2. Define the terms concentration gradient, selectively permeable, hypertonic, hypotonic, and isotonic.
3. Describe what happens when cells are in hypertonic, hypotonic, and isotonic environments.
4. Describe the processes of plasmolysis, crenation, and hemolysis.

## Introduction

Have you ever sprayed perfume at one end of a room, and after a while your friend at the other end of the room notices it? How about adding cream to your coffee? At first the coffee is mostly black, but lightens over time. These are examples of **diffusion**, which is the movement of molecules from an area where they are more concentrated to an area where they are less concentrated.

Diffusion occurs because of the ***constant*** and ***random*** movement of molecules. When there is a difference in concentration in one area (such as two sides of a room), we say there is a **concentration gradient**. For example, at first the perfume is highly concentrated in one part of the room and less concentrated in another part. Over time, the perfume will ***diffuse down its concentration gradient*** until the molecules are equally distributed throughout the area.

Diffusion is essential for life. All cells must eat, drink, and expel waste to survive. Cells are mostly water, and live in a watery environment. Any substance dissolved in a liquid is called a **solute**. This means solutes must diffuse into, and out of, cells. For this to happen, they must pass through the **cell membrane**, or **plasma membrane**. Cell membranes are described as **selectively permeable**, meaning they only allow certain solutes to pass through them. Smaller and/or nonpolar things (lipids, gases) are permeable, while larger and/or polar things (ions, sugars, proteins) are not. (You will soon learn in lecture that cells have a way to allow some of these impermeable things to pass through.)

What happens when the solute is ***not*** permeable to the plasma membrane? It cannot diffuse and stays on one side, either inside or outside the cell. Imagine a cell in a very sugary solution. The sugar cannot pass through the membrane, so it stays outside the cell. However, everything else that *can* diffuse *will* diffuse, including water itself.

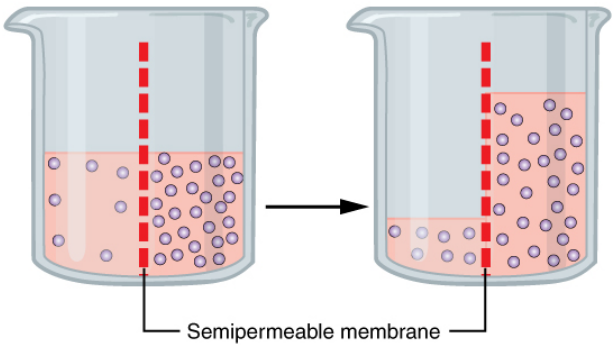


**Figure 3.1. Osmosis is the diffusion of water down its concentration gradient, when the solute is not permeable.**

The diffusion of water across a membrane is called **osmosis** (**Fig. 3.1**). It occurs when the concentration of an ***impermeable*** solute is higher on one side and lower on the other side. Since the solute cannot diffuse down its concentration gradient, the water will do so instead. This means a cell can lose water, gain water, or stay the same, depending on the concentration gradient. These three possible environments are known, respectively, as **hypertonic**, **hypotonic**, and **isotonic**. See **Table 3.1**.

| Environment | Solute concentration outside the cell | Solute concentration inside the cell | Net flow of water |
|---|---|---|---|
| Hypertonic | higher | lower | out of the cell |
| Hypotonic | lower | higher | into the cell |
| Isotonic | same | same | no net flow |

**Table 3.1. The three osmotic environments.**

One difference between plant and animal cells is their cell barriers. Plant cells have a plasma membrane surrounded by a rigid cell wall, and animal cells have only a plasma membrane. Both cell types are affected by solute concentration. In hypertonic environments, plant cells experience **plasmolysis**, a process where the plasma membrane pulls away from the cell wall; red blood cells experience **crenation**, a process where cell shape becomes spiky and irregular. In hypotonic environments, plant cells swell up, which helps maintain their structure; red blood cells burst, or lyse, in a process called **hemolysis**.

In this lab, you will observe two demos, one showing diffusion and another showing osmosis. You will also observe osmosis happening in live plant and animal cells.

## Materials & Methods

### ACTIVITY 1: DIFFUSION DEMO

Your instructor will demonstrate the process of diffusion by adding food coloring to a beaker of water.

**Describe what is happening in the beaker at a molecular level.**

### ACTIVITY 2: OSMOSIS DEMO

Your instructor will demonstrate the process of osmosis with a chicken egg. Eggshells contain calcium carbonate which makes them hard. When calcium carbonate is exposed to acid, it dissolves and carbon dioxide is formed. ***The remaining membrane is selectively permeable.*** Eggs were soaked in vinegar (acid) to dissolve the shell, and then placed in corn syrup (a hypertonic environment) to extract excess water. One of the eggs was weighed and placed in distilled water (water with no solutes).

**What is the initial and final weight of the egg?**

**Describe what has happened with the egg at a molecular level.**

**What would happen if the egg was placed back in a hypertonic environment?**

## ACTIVITY 3: OSMOSIS IN PLANT CELLS

In this activity you will observe the effect of osmosis in onion skin cells placed in solutions with different solute concentrations. The solute here is sodium chloride (table salt, NaCl).

**Materials**

- onion skin cells
- distilled water
- 0.9% and 10% NaCl solutions
- microscope slides
- coverslips

1. **Work in pairs** for Activity 3.
   Obtain TWO small pieces of onion skin (about this big: ▢ ).
   You want ONLY the top layer. The best way is to snap a piece of onion and peel off the layer.

2. **Prepare your specimens.**
   a. Place each piece of onion skin on a slide.
   b. Add one drop of 0.9% NaCl solution to each.
   c. Place a coverslip on each specimen.

3. Each person will view one specimen with the 4x, 10x, and 40x objectives. **Sketch with 10x or 40x.**
   When you are done sketching, leave the slide on the stage and do not adjust the focus.

4. **Change the environment** of your specimens.
   a. To one specimen, carefully add two drops of **10% NaCl** next to one edge of the coverslip. It should seep under the coverslip.
   b. To the other specimen, do the same but with **distilled water**.

5. View both specimens with the 4x, 10x, and 40x objectives. **Sketch with 10x or 40x.**

6. **Clean up:** slides go in the Bleach Water container; return everything else where you got them.

**Select the correct terms for each environment.**

Onion skin cell sketch with _____x objective
**0.9% NaCl**

- ☐ HYPERTONIC
- ☐ HYPOTONIC
- ☐ ISOTONIC
- ☐ CONTROL GROUP

Onion skin cell sketch with _____x objective
**10% NaCl**

- ☐ HYPERTONIC
- ☐ HYPOTONIC
- ☐ ISOTONIC
- ☐ CONTROL GROUP

Onion skin cell sketch with _____x objective
**distilled water**

- ☐ HYPERTONIC
- ☐ HYPOTONIC
- ☐ ISOTONIC
- ☐ CONTROL GROUP

**Describe what happened with the onion cells when you added 10% NaCl.**

**Describe what happened with the onion cells when you added distilled water.**

## ACTIVITY 4: OSMOSIS IN ANIMAL CELLS

In this activity you will observe the effect of osmosis in red blood cells placed in solutions with different solute concentrations. The solute here is sodium chloride (table salt, NaCl).

**Materials**

- red blood cells
- distilled water
- 0.9% and 10% NaCl solutions
- microscope slides
- coverslips

Your instructor will prepare the specimens (red blood cells in one of the three solutions) one at a time, in this order: 0.9% NaCl; 10% NaCl; distilled water.

1. Work in pairs for Activity 4. Obtain your first specimen.
   a. From your instructor, obtain one drop of red blood cells in **0.9% NaCl**.
   b. Place a coverslip on your specimen.

2. Each person will view the specimen with the 4x, 10x, and 40x objectives. **Sketch with 10x or 40x.** When you are done sketching, remove the slide from the stage and set it aside.

3. Repeat step 1 to obtain cells in **10% NaCl *as soon as*** they are ready.

4. Repeat step 2 with your new specimen.

5. Repeat step 1 to obtain cells in **distilled water *as soon as*** they are ready.

6. Repeat step 2 with your new specimen.

7. **Clean up:** slides go in the Bleach Water container.

**Select the correct terms for each environment.**

Red blood cell sketch with _____x objective
**0.9% NaCl**

- ☐ HYPERTONIC
- ☐ HYPOTONIC
- ☐ ISOTONIC
- ☐ CONTROL GROUP

Red blood cell sketch with _____x objective
**10% NaCl**

- ☐ HYPERTONIC
- ☐ HYPOTONIC
- ☐ ISOTONIC
- ☐ CONTROL GROUP

Red blood cell sketch with _____x objective
**distilled water**

- ☐ HYPERTONIC
- ☐ HYPOTONIC
- ☐ ISOTONIC
- ☐ CONTROL GROUP

**Describe what happened with the red blood cells when you added 10% NaCl.**

**Describe what happened with the red blood cells when you added distilled water.**

# LAB 4 Limits of Cell Size

## Objectives

At the end of the lab, you should be able to:

1. Explain the relationship between surface area and volume as objects get larger.
2. Explain how this relationship influences the size and shape of cells.

## Introduction

In Lab 3 you learned about diffusion and osmosis, the processes by which cells get most of their nutrients. Cells must be able to take in nutrients quickly enough to feed themselves. They must pass through the plasma membrane (also called absorption), and then diffuse through the cytoplasm to where they are needed. Absorption and diffusion take time. The rates of absorption and diffusion limits how large a cell can become. The purpose of this lab is to see how cell size and shape affect the rate of absorption.

All cells have some amount of surface area (the plasma membrane) and volume (the space inside). If we assume that cells are shaped like spheres, we can find their surface area (SA) and volume (V):

$$SA = 4\pi r^2 \qquad V = \frac{4}{3}\pi r^3$$

Looking at these equations, you can see that as a cell gets bigger, its surface area goes up with $r^2$, but its volume goes up with $r^3$. In other words, volume goes up ***faster than*** surface area as a cell gets bigger. The rate of absorption depends on surface area. This means there is a point at which the volume is too big for the surface area to support. In other words, if a cell gets too big, nutrients will not be absorbed fast enough to meet its metabolic needs.

In this lab, you will perform a cell feeding simulation to see how a "nutrient" is absorbed into four "cells" of different sizes and shapes.

## Materials & Methods

For this simulation, the "cells" are made of a carbohydrate called agar, and have been filled with a blue pH indicator. When exposed to acid, the blue color disappears. The agar by itself appears light yellow. Thus, the more yellow the cell gets, the more it has been fed. The more blue remains, the more it has not been fed.

**Materials**

- four simulated cells in bowl
- 0.01 M hydrochloric acid
- ruler
- calculator

1. Each group will get one bowl with four cells. Their dimensions are in **Table 4.1**.

2. Your instructor will come by and immerse your cells with the acid "nutrient".

3. Allow Cell 1 to lose most of its blue color. This will take ~10 minutes. While you are waiting, calculate the volume of the four cells in **Table 4.1**.

4. When Cell #1 looks like this (  ), pour the acid into the sink.

5. Rinse the cells in water twice. Bring them back to your bench.

6. Lay out the cells on a paper towel.

7. **As soon as** Cell 1 is completely yellow, complete Table 4.2 by measuring the **blue parts** in Cells 2-4. Begin answering the questions on the next page.

| Cell | Cell shape | Diameter (d) | Length (l) | Cell volume (V) |
|---|---|---|---|---|
| 1 |  | 0.8 cm | 8 cm | |
| 2 |  | 1.1 cm | 2 cm | |
| 3 |  | 1.1 cm | 4 cm | |
| 4 |  | 1.6 cm | 2 cm | |

Volume of a cylinder:

$$V = \pi r^2 l$$

$$r = \frac{1}{2}d$$

**Table 4.1. Dimensions of the four cells.**

| Cell | Cell Shape | Diameter ($d_f$) | Length ($l_f$) | Volume ($V_f$) | Percent of cell "fed" |
|---|---|---|---|---|---|
| 1 |  | 0 cm | 0 cm | 0 $cm^3$ | 100% |
| 2 |  | | | | |
| 3 |  | | | | |
| 4 |  | | | | |

**Table 4.2. Measurements of the blue parts at the end of the simulation.**

$$\%\ of\ cell\ fed = \frac{V - V_f}{V} \times 100$$

1. Which cell is able to feed itself the fastest?

2. Which cell fed the slowest?

3. Do these two cells have the same volume?

4. How do they compare in shape?

5. Calculate the ratio of surface area to volume ($SA/V$) for all cells. The surface areas of the cells are

| Cell 1: | Cell 2: | Cell 3: | Cell 4: |
|---|---|---|---|
| $SA = 21.1\ cm^2$ | $SA = 8.8\ cm^2$ | $SA = 15.7\ cm^2$ | $SA = 14.1\ cm^2$ |

6. Which cell has the largest $SA/V$ ratio?

7. How does surface area affect the rate of absorption?

8. How might a cell increase its surface area without changing its volume?

9. Which of the four cells is best at acquiring enough nutrients to survive?

10. Why would a growing cell eventually have to divide into two cells?

11. Why are cells as small as they are?

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# Lab Review

## Introduction

At this point in the labs, you have learned about some key tools and techniques in studying biology:

1. The scientific process
2. Microscopes
3. Diffusion and osmosis in cells

Since Lab 4 is fairly short, we will use the rest of our lab time this week to review these things. The first activity gives you a chance to participate in real science by collecting data and applying the scientific process. The second activity reviews how compound light microscopes work, and the effects of osmosis on cells.

## Materials & Methods

**ACTIVITY 1: RESEARCH AT GORONGOSA NATIONAL PARK**
***(adapted from HHMI Biointeractive: www.biointeractive.org)***


This activity connects you to actual ecological research in Gorongosa National Park in Mozambique. Gorongosa was once a thriving national park, until decades of war decimated the park's large animal populations and halted tourism. A long-term restoration project is underway to restore the park's ecosystem, revive tourism, and support local communities.

Gorongosa's researchers are using different approaches to track the recovery the park's wildlife, including ***remote trail cameras***. These cameras are equipped with motion sensors that snap photos when an animal moves in front of them. The trail cameras have collected hundreds of thousands of photos that are now available on WildCam Gorongosa, an online citizen science platform.

Scientists in Gorongosa National Park use the data generated through WildCam Gorongosa to answer a variety of questions about animal communities and their habitats. In this activity, you will ***collect actual data*** from WildCam Gorongosa and use those data to ***create graphs*** and ***answer an ecological question of your choice***.

**Materials**

- laptop with Internet access

You will complete this activity in pairs, and pool your data on a shared spreadsheet with Google Sheets.

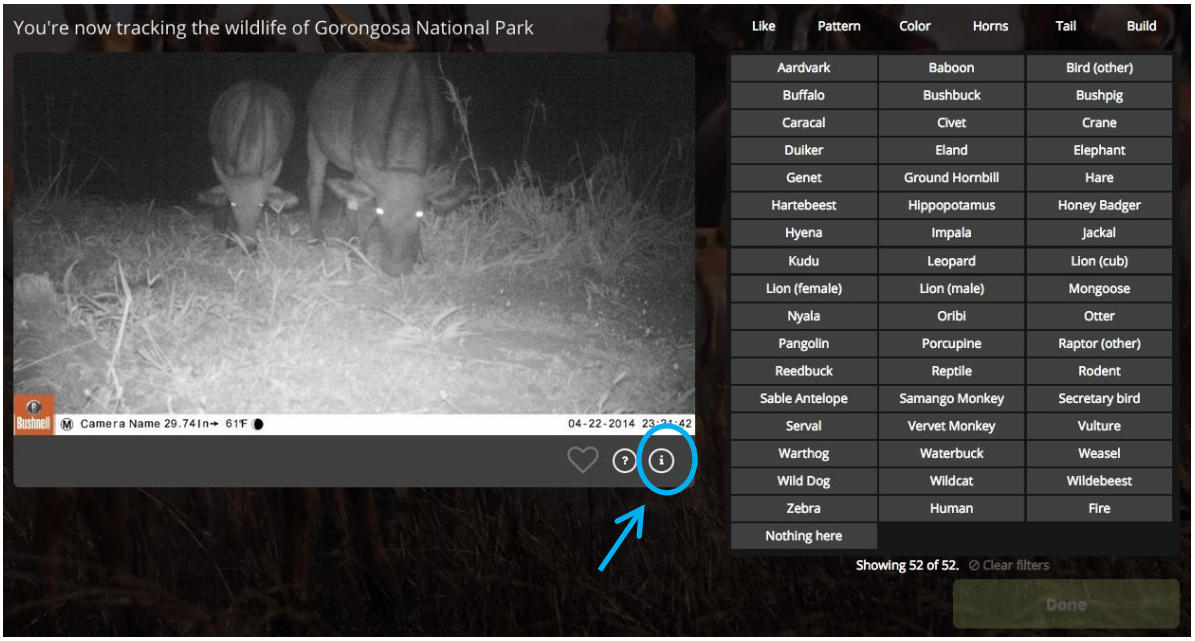
**Part 1 – Collect Data**

1. Open the class spreadsheet in which you will collect your data. Your instructor will provide you with the link.

2. Open WildCam Gorongosa: http://www.wildcamgorongosa.org/#/classify

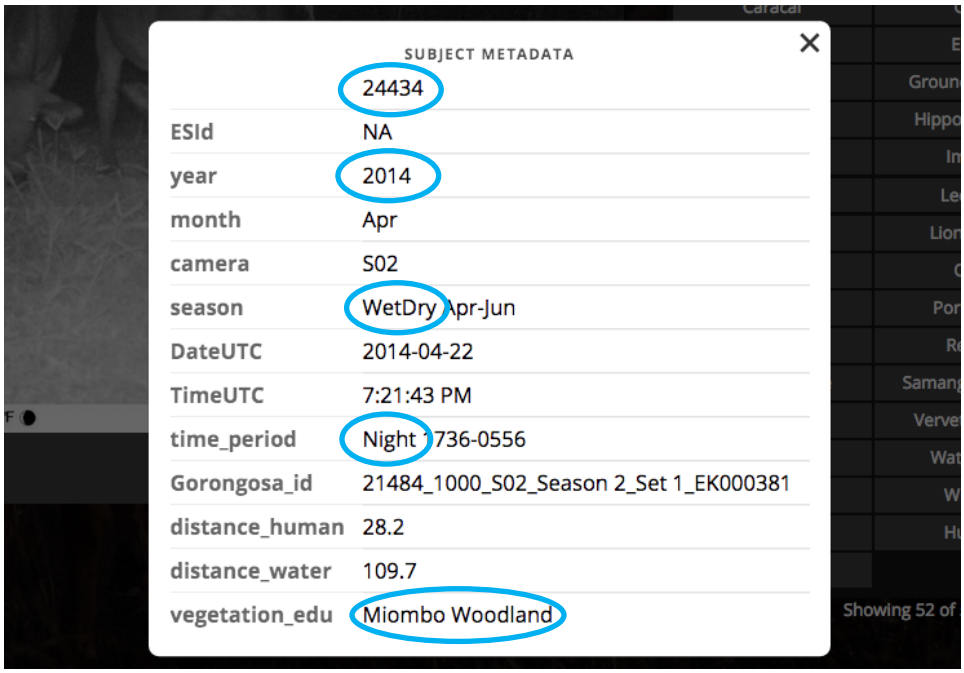
3. Your task is to collect data on individual animal "sightings". A sighting is a unique species that you see in a photo. For example, a photo with two warthogs in it is one sighting and a photo with two warthogs and one impala counts as two sightings; a photo with no animals in it does not count as a sighting.

4. For each photo with an animal in it, click the "i" icon below the photo (see image below).



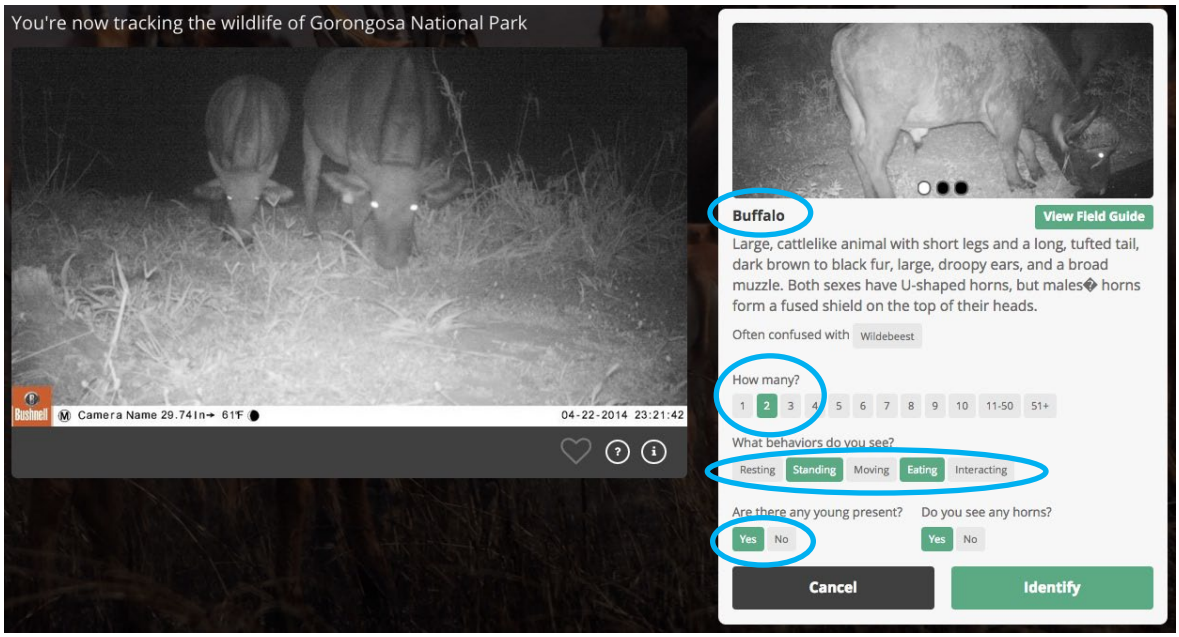

5. Clicking on the "i" will open a window with different types of data associated with the photo, as shown in the image below**. Record the following data in the spreadsheet (see image below):**

   - **Image ID**: the number at the top
   - **Year**
   - **Season**: only record the season name, not the months. The options are Wet, Dry, WetDry (which refers to the transition between the wet and dry seasons), or DryWet (which refers to the transition between the dry and wet seasons).
   - **Time period**: only record Night or Day, not the times.
   - **Vegetation**: this is the type of vegetation where the photo was taken




6. Close the metadata window and on the website, **identify the species** you see in the photo. As you do this, **record the following data in the spreadsheet (see image below):**

- **Animal**: the name of the species
- **Number**: how many of that species appear in the photo
- **Resting**: record a "1" if you selected this behavior, or a "0" if you did not
- **Standing**: record a "1" if you selected this behavior, or a "0" if you did not
- **Moving**: record a "1" if you selected this behavior, or a "0" if you did not
- **Eating**: record a "1" if you selected this behavior, or a "0" if you did not
- **Interacting**: record a "1" if you selected this behavior, or a "0" if you did not
- **Young**: record a "1" if you selected Yes, or a "0" if you selected No



7. Click “Identify”. If there is another species in the same photo, repeat steps 5-7 and record those data in the next row in the spreadsheet. If there are no other species in the photo, select “Done”.

8. Repeat steps 4-7 until you have collected data on **five** animal sightings (in other words, until you have filled out five lines in the spreadsheet). The class will have about 50-75 sightings.

**Part 2 – Analyze Data**

You will now use the data you collected to answer this question: “Which vegetation type in Gorongosa has the highest abundance of animals?”

1. In Google Sheets, **create a bar chart** that shows ***vegetation type*** on the X-axis (use the Vegetation column) and the ***total number of animals*** on the Y-axis (use the Number column).

2. What does this graph tell you about the abundance of animals in different habitats?

Now, think of a basic scientific question you can answer by comparing two columns in the spreadsheet.

3. What is your scientific question?

4. Rewrite your question as a good scientific hypothesis.

5. How would you test this hypothesis using data from the spreadsheet?

6. In Google Sheets, create a chart that will allow you to test your hypothesis.

7. What does your graph show?

8. Was your hypothesis supported or rejected? Why?

## ACTIVITY 2: MICROSCOPE HIGHLIGHTS & OSMOSIS

In this activity you will review some key points on how microscopes work, and the effects of osmosis on cells.

### Part 1 – Microscopes

#### Field of view

What is it?

Fill out this table on how the field of view and amount of light change as you go from lower to higher magnification.

| Total magnification | Field of view | Amount of light |
|---|---|---|
| 40x |  | |
| 100x | | |
| 400x | | |
| 1000x | | |

What should you do to adjust for the amount of light?

#### Image orientation

How do you move the slide on the microscope stage?

When you move the slide, how does your field of view move?

#### Depth of field

What is it?

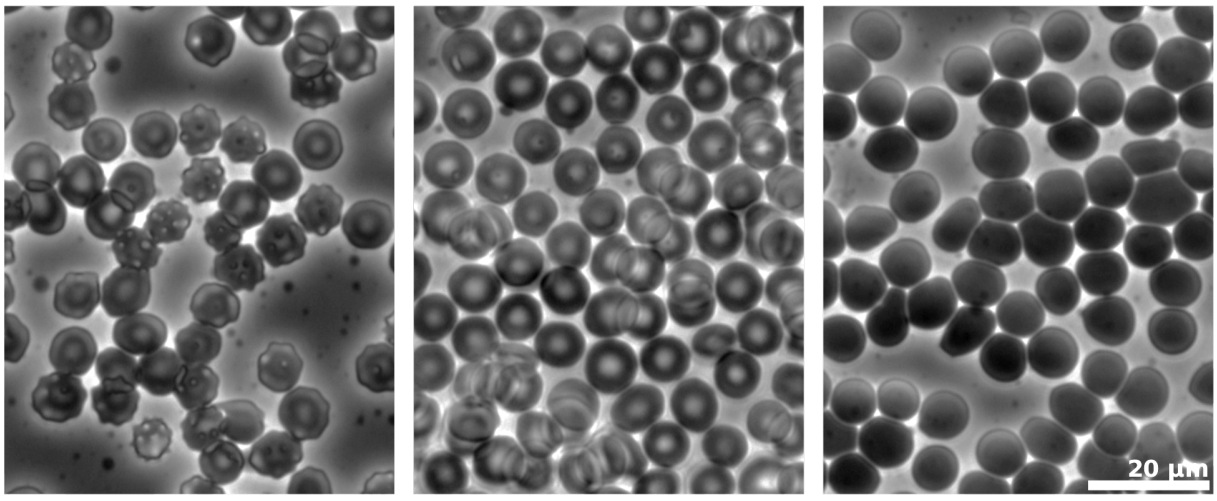
Sketch the depth of field at the various magnifications.

| Total magnification | Depth of field |
|---|---|
| 40x | |
| 100x | |
| 400x | |
| 1000x | |

How does this inform the way you focus on a specimen?

**Osmosis**

Here are three microscope images of red blood cells. Which ones are in a hypotonic environment, a hypertonic environment, and an isotonic environment?




Describe these three environments. In other words, what would you find in them?

# LAB 5 Enzymes & Catalysts

## Objectives

At the end of the lab, you should be able to:

1. Define the terms catalyst and enzyme.
2. Explain the properties of catalysts and enzymes.
3. Explain how and why surface area, high temperature, and pH affect an enzyme's ability to catalyze a reaction.

## Introduction

Life is essentially a highly organized series of chemical reactions. In Labs 3 and 4, you saw that cells acquire materials (i.e. eat and drink) from their environment to survive. Cell use these materials as an energy source, and to build more cell parts. This requires chemical reactions; think of digesting starch or building muscle mass.

These reactions can occur spontaneously, meaning they happen by themselves. If things bump into each other at the right speed and angle, it is possible to make or break a bond. However, the rate of these spontaneous reactions is usually far too slow to keep the cell alive. Recall the food coloring demo in Lab 3: diffusion was happening by itself, but not very quickly. Cells need better control of ***when, where, and how fast*** a reaction will happen. How do they do this? Enzymes!

Enzymes are more broadly known as catalysts. All enzymes are catalysts, but not all catalysts are enzymes. It is like saying that all apples are fruits, but not all fruits are apples. A **catalyst** is a molecule that speeds up the rate of a specific reaction, without itself changing. In other words, catalysts are reusable and can **catalyze** a reaction many times. How do they do this? See **Fig. 5.1**. They act like matchmakers. They bring the reactants together and interact with them in a way that lowers the **activation energy** of the reaction. This makes it much more likely that the reaction will proceed.

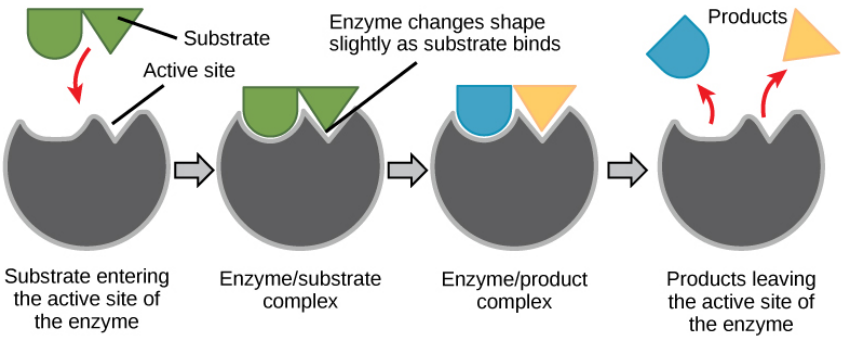



**Figure 5.1. The steps of an enzyme-catalyzed reaction.**
**Note that other enzymes can bind two substrates and make bonds between them.**

Almost all of a cell's catalysts are **enzymes**, which have one more important feature: ***they are proteins.*** Like all proteins, enzymes work due to their 3-D shape, or structure. This shape is held together by bonds and forces that are sensitive to changes in ***temperature, pH, and salt concentration***. Enzymes are organic molecules; thus, they are organic catalysts. Inorganic catalysts also exist, and they are not sensitive to changes in temperature, pH, or salt concentration.

In this lab, you will investigate the activity of an inorganic catalyst called manganese dioxide ($MnO_2$) and an enzyme called peroxidase. They both catalyze the same reaction:

$$\mathbf{2\ H_2O_2 \rightarrow 2\ H_2O + O_2}$$

2 hydrogen peroxide → 2 water + oxygen gas

## Materials & Methods

### ACTIVITY 1: INORGANIC CATALYST

In this activity you will test the ability of manganese dioxide ($MnO_2$), an inorganic catalyst, in different environmental conditions. You will test it at ***two temperatures*** and ***three pH levels***.

**Materials**

- test tubes in a test tube rack
- metal test tube holder
- squeeze bottle of water
- hot water bath with boiling chips
- manganese dioxide ($MnO_2$)
- hydrogen peroxide ($H_2O_2$)
- pH 4, 7, 10 buffer solutions

1. **Set up a hot water bath.**
   a. The larger beaker should be about half full of water, with boiling chips at the bottom.
   b. Place the beaker on the hot plate.
   c. Plug in the hot plate. Use the left dial to turn it on and set it to 200°.
   d. Proceed to step 2.
   e. When the water is almost boiling, turn it down to 100° and keep the water at a low boil.

2. **Set up tube 5 first.**
   a. Use the squeeze bottle to fill tube 5 with distilled water to the **first** marked line on the tube.
   b. Add a small amount of **$MnO_2$**.
   c. Place the tube in the hot water bath and let boil for 8 minutes. Start time when the hot water bath begins boiling. Proceed to step 3 while it boils.

3. **Set up tubes 1-4.** The final ingredients for each tube are listed in **Table 5.1**.
   a. You will add the ingredients in order ***from left to right*** based on **Table 5.1** (see next steps).
   b. Add the appropriate **pH buffer** solution to the **first** marked line on the tube.
   c. Add **peroxide (or water for tube 1)** to the **second** marked line on the tube.
   d. Add a small amount of **$MnO_2$**. Try to keep the amounts the same as you used in tube 5.
   e. In **Table 5.1**, record the relative speed of the reaction in each tube.

4. **Go back to tube 5.**
   a. After 8 minutes, use the test tube holder to remove the tube and **carefully pour out the water** at the sink. It's fine if you cannot pour it all out – just **do not pour out the $MnO_2$!**
   b. Add the **pH 7 buffer** solution to the **first** marked line on the tube.
   c. Add **peroxide** to the **second** marked line on the tube.
   d. In **Table 5.1**, record the relative speed of the reaction.

5. **Clean up once you record your results.**
   a. At the sink with the mesh strainer, carefully empty the contents of each tube. The strainer will catch the $MnO_2$ and the rest will flow through.
   b. Rinse out your tubes a few times with water.
   c. Put them back in your tube rack.

| Ingredients in each test tube | | | | | | Speed of reaction |
|---|---|---|---|---|---|---|
| pH | $H_2O_2$ | $H_2O$ | Boiled $MnO_2$ | $MnO_2$ | TEST TUBE | *None* *Slower* *Faster* ⟷ |
| 7 | | ✓ | | ✓ | 1 | |
| 4 | ✓ | | | ✓ | 2 | |
| 7 | ✓ | | | ✓ | 3 | |
| 10 | ✓ | | | ✓ | 4 | |
| 7 | ✓ | | ✓ | | 5 | |

**Table 5.1. Inorganic catalyst reaction ingredients (left) and results (right).**

**Questions**

1. In tube 1, why did you test the catalyst ($MnO_2$) with water? Hint: the peroxide you use is in solution, meaning it is mixed with water.

2. How can you tell if a reaction is happening, and how fast it is happening?

3. The bubbles you saw are the product of the reaction, so they are made of…

4. Does the $MnO_2$ get used up as the reaction keeps going?

5. If the $MnO_2$ was getting used up, how would you be able to tell?

6. Do your results agree with the fact that $MnO_2$ is an inorganic catalyst? Explain.

## ACTIVITY 2: ENZYME (ORGANIC CATALYST)

Hydrogen peroxide is a toxic waste product of metabolism, as it is very reactive with other molecules. Most cells have an enzyme called ***peroxidase*** that catalyzes the breakdown of $H_2O_2$.

In this activity you will test the ability of peroxidase from ***liver*** or ***potato*** cells in different environmental conditions. You will test it at ***two temperatures, three pH levels, and two surface area conditions***.

Your instructor will assign either liver or potato to each group.

**Materials**

- same items as Activity 1, plus…
- mortar and pestle
- sand
- fresh liver or potato
- cutting board and knife
- tweezers

1. **Set up a hot water bath.** Repeat step 1 from Activity 1 if needed.

2. **Set up tube 12 first.**
   a. Use the squeeze bottle to fill tube 12 with distilled water to the **first** marked line on the tube.
   b. Add a small whole piece of liver or potato.
   c. Place the tube in the hot water bath and let boil for 10 minutes. Start time when the hot water bath begins boiling. Proceed to step 3 while it boils.

3. **Set up tubes 6-11.** The final ingredients for each tube are listed in **Table 5.2**.
   a. You will add the ingredients in order ***from left to right*** based on **Table 5.2** (see next steps).
   b. Add the appropriate **pH buffer** solution to the **first** marked line on the tube.
   c. Add **peroxide (or water for tube 6)** to the **second** marked line on the tube.
   d. Add a small amount of **sand** to **tubes 6** and **7**.
   e. Obtain **four** small **whole pieces** of liver or potato. Try to keep the pieces the same size as the one in tube 12.
   f. Add one piece each to **tubes 8, 9,** and **10**. Record results in **Table 5.3**.
   g. Grind up the fourth piece with a mortar and pestle. Add a small amount of sand to help you grind it into smaller pieces. Add this to **tube 11**. Record results in **Table 5.3**.

4. **Go back to tube 12.**
   a. After 10 minutes, use the test tube holder to remove the tube and **carefully pour out the water** at the sink. It's fine if you cannot pour it all out – just **do not pour out the liver or potato!**
   b. Add the **pH 7 buffer** solution to the **first** marked line on the tube.
   c. Add **peroxide** to the **second** marked line on the tube.
   d. In **Table 5.3**, record the relative speed of the reaction.

5. **Clean up once you record your results.**
   a. Turn off your hot plate and unplug it.
   b. At a sink, carefully empty the contents of each tube.
   c. Rinse out your tubes a few times with water.
   d. Put them back in your tube rack.

| | Ingredients in each test tube | | | | | | |
|---|---|---|---|---|---|---|---|
| **TEST TUBE** | **pH** | **$H_2O_2$** | **$H_2O$** | **Sand** | **Whole piece** | **Ground up piece** | **Boiled whole piece** |
| 6 | 7 | | ✓ | ✓ | | | |
| 7 | 7 | ✓ | | ✓ | | | |
| 8 | 4 | ✓ | | | ✓ | | |
| 9 | 7 | ✓ | | | ✓ | | |
| 10 | 10 | ✓ | | | ✓ | | |
| 11 | 7 | ✓ | | | | ✓ | |
| 12 | 7 | ✓ | | | | | ✓ |

**Table 5.2. Peroxidase reaction ingredients.**

| ***Speed of reaction: LIVER*** | | | | ***Speed of reaction: POTATO*** | | |
|---|---|---|---|---|---|---|
| ***None*** | ***Slower*** | ***Faster*** | **TEST TUBE** | ***None*** | ***Slower*** | ***Faster*** |
| | | | 6 | | | |
| | | | 7 | | | |
| | | | 8 | | | |
| | | | 9 | | | |
| | | | 10 | | | |
| | | | 11 | | | |
| | | | 12 | | | |

**Table 5.3. Peroxidase reaction results for liver (left) and potato (right).**

## Questions

### *Tubes 6 and 7*

1. What was the purpose of tube 6? (Tube 6 had sand and water.)

2. What was the purpose of tube 7? (Tube 7 had sand and peroxide.)

***Surface area***

3. Which has more surface area, a whole piece of something or the same-sized piece ground up?

4. Explain your results for tubes 9 and 11.

5. How does the available surface area of enzymes affect their activity?

***Temperature***

6. Explain your results for tubes 9 and 12.

7. How does high temperature affect the activity of an enzyme?

***pH***

8. Explain your results for tubes 8, 9, and 10.

9. How does pH affect the activity of an enzyme?

***Peroxidase vs. $MnO_2$***

10. Explain how your results for tubes 5 and 12 support the claim that liver and potato **do not** contain $MnO_2$.

# LAB 6 Metabolism

## Objectives

At the end of the lab, you should be able to:

1. Describe the overall process of aerobic glucose metabolism.
2. Describe the overall process of anaerobic glucose metabolism (fermentation).
3. Describe the differences between aerobic and anaerobic metabolism.
4. Explain why some lentils respired while others did not.
5. Explain why some sugars are easier for yeast to ferment than others.

## Introduction

Cells build and use thousands of enzymes, which require specific environments (temperature, pH, salt concentration) to work. Some of these enzymes break down food molecules, which releases energy that is used to build ATP, the main energy carrier in cells. Glucose ($C_6H_{12}O_6$) is the major food molecule cells break down for energy. It is broken down to carbon dioxide ($CO_2$) in three stages:

**Glycolysis**

- occurs in the cytoplasm
- glucose is broken down to pyruvate
- ATP and NADH are made

**Krebs cycle**

- occurs in mitochondria
- pyruvate is broken down to $CO_2$
- ATP, NADH, and $FADH_2$ are made

**Electron transport chain (ETC)** and **chemiosmosis**

- occurs in mitochondria
- ETC uses oxygen ($O_2$) and electrons from NADH and $FADH_2$
- (much more) ATP is made

**The overall reaction: $C_6H_{12}O_6 + 6\ O_2 \rightarrow 6\ CO_2 + 6\ H_2O$ + 36-38 ATP**

In this case, oxygen is required in the ETC, so this type of glucose metabolism is known as **aerobic**. Most multicellular animals must make ATP this way to meet its energy demands. However, there is a way to make ATP without oxygen, also known as **anaerobic** glucose metabolism. One type of this is called **fermentation**, and occurs in certain organisms when there is not enough oxygen around. There are two stages in fermentation:

**Glycolysis**

- occurs in the cytoplasm
- glucose is broken down to pyruvate
- ATP and NADH are made

**Fermentation (in yeast)**

- occurs in the cytoplasm
- pyruvate is broken down to ethanol and $CO_2$
- $NAD^+$ is made

**The overall reaction:**

**$C_6H_{12}O_6 \rightarrow 2\ C_2H_6O + 2\ CO_2 + 2\ ATP$**

In this lab, you will see use lentils and yeast to detect aerobic and anaerobic glucose metabolism.

## Materials & Methods

### ACTIVITY 1: AEROBIC METABOLISM WITH LENTILS

In Activity 1 of this lab, you will test the ability of lentils to perform aerobic glucose metabolism (you can use the verb ***respire*** as a shorthand). Looking at the overall reaction, the easiest product to detect is $CO_2$ as it is the only one the cells do not want. You will detect this gas by mixing it with a pH indicator called **phenol red**, which is ***yellow below pH 7*** and ***red above pH 8***. When $CO_2$ mixes with water, it acts like an acid. The lentils have been prepared in three ways: dry (ungerminated); wet (germinated); and boiled (germinated). Each group will test one type of lentil.

**Materials**

- one respiration apparatus (**Fig. 6.1**)
- phenol red
- one type of lentil: dry (ungerminated); wet (germinated); or boiled (germinated)

1. Obtain one respiration apparatus (**Fig. 6.1**) and the type of lentil your group has been assigned.
   - Dry or wet lentils: fill the beaker about halfway
   - Boiled: take one beaker of boiled lentils
2. Set up the apparatus according to **Fig. 6.1**.
   - Make sure the two rubber stoppers make a good seal.
   - Add enough water to the test tube to cover the end of the small glass tube.
   - These two steps ensure that all gases stay inside the beaker.
3. Let the experiment run for one hour.
4. After one hour, replace the water in the test tube with the phenol red solution.
5. Fill the small beaker with tap water.

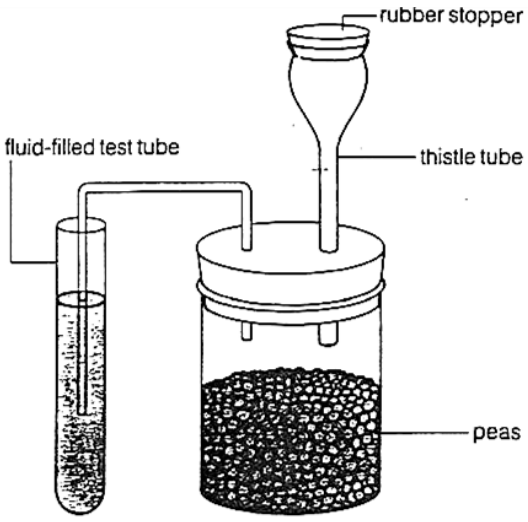



**Figure 6.1. A respiration apparatus.**

6. Remove the smaller rubber stopper on the thistle tube, and **slowly** pour in the tap water. The water will push the gases through the glass tube and into the phenol red solution.

7. Record the color of the phenol red solution in **Table 6.1**.

8. Clean up:
   - Lentils go in the bucket
   - Water and phenol red can go down the sink
   - Rinse and return the apparatus

| Type of lentil | Color of phenol red? | Was there $CO_2$? | Did the lentils respire? |
|---|---|---|---|
| Dry (ungerminated) | | | |
| Wet (germinated) | | | |
| Boiled (germinated) | | | |

**Table 6.1. Results of lentils respiration experiment.**

**Questions**

1. Which lentils were respiring?

2. Do lentils respire before germination?

3. What do you think is the difference in the lentils before and after germination?

4. Originally, what color was the phenol red solution? Does this mean it is acidic or basic?

5. Explain why boiling the lentils after germination caused the results that happened.

## ACTIVITY 2: ANAEROBIC METABOLISM WITH YEAST

In Activity 2, you will test the ability of yeast to perform fermentation with different sugars as their food source. Yeast are single-celled fungi that must eat to survive (unlike plants). Some species eat fruit sugar (fructose), others can eat the sugar in plant fiber (cellulose), and so on. They can respire and ferment, depending on whether they get enough oxygen. If we keep oxygen levels low, they will have to rely on anaerobic metabolism to make ATP.

**Materials**

- one fermentation tube
- ¼ teaspoon of yeast
- graduated cylinder
- Parafilm
- incubator
- one 10% sugar solution: glucose; galactose; fructose; maltose; sucrose; lactose

1. In the proper graduated cylinder, add 20 mL of your assigned sugar solution and ¼ teaspoon of yeast.
2. Take a piece of Parafilm, remove the paper, and stretch it over the top of the graduated cylinder.
3. Invert the graduated cylinder several times to mix the yeast.
4. Slowly pour the mixture into a fermentation tube. Every so often, tilt the tube back to get rid of any air bubbles in the long narrow part of the tube. It should look like **Fig. 6.2** when done.
5. Label your tube in some way so you can identify it.
6. Place your tube in the incubator for 20 minutes.
7. After 20 minutes, take your tube out of the incubator. We will record your results in **Table 6.2** as a class.
8. Clean up: rinse out your tube, remove labels, and return it.

**Figure 6.2. A filled fermentation tube.**

| Type of sugar | Amount of bubbles (0 = none, 5 = lots) | Did the yeast ferment? |
|---|---|---|
| Glucose | | |
| Galactose | | |
| Fructose | | |
| Maltose | | |
| Sucrose | | |
| Lactose | | |

**Table 6.2. Results of yeast fermentation experiment.**

**Questions**

1. Which sugar did you test?

2. Glucose, galactose, and fructose are monosaccharides.
   Maltose, sucrose, and lactose are disaccharides (made up of two monosaccharides).
   Which two monosaccharides make up each of the three disaccharides? You will need to look this up.

3. Which sugar was fermented the least?

4. Pyruvate is used in fermentation. What does it get broken down to?

5. Explain why these yeast could ferment some sugars but not others.

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# LAB 7 Photosynthesis

## Objectives

At the end of the lab, you should be able to:

1. Describe the overall process of photosynthesis.
2. Describe chloroplast structure in plant cells.
3. Describe the role of pigments (such as chlorophyll) in photosynthesis.
4. Explain how pigments and filters work.

## Introduction

In Lab 6 you saw that germinated lentils need to break down glucose for energy, releasing $CO_2$ as a byproduct. How did the lentils get that glucose? They made it themselves through **photosynthesis**, the process of using light energy, water, and $CO_2$ to make glucose and other food molecules.

**The overall reaction: $6\ CO_2 + 6\ H_2O$ + light energy → $C_6H_{12}O_6 + 6\ O_2$**

Essentially, light energy is captured and stored in the bonds of glucose. Where is light energy captured? Most cells in the leaves of plants contain an organelle called a chloroplast (**Fig. 7.1**). **Chloroplasts** contain stacks of disk-shaped membranes called grana (singular: granum). These disk-shaped sacs are called **thylakoids**, and their membranes contain several kinds of pigments, including **chlorophyll**.

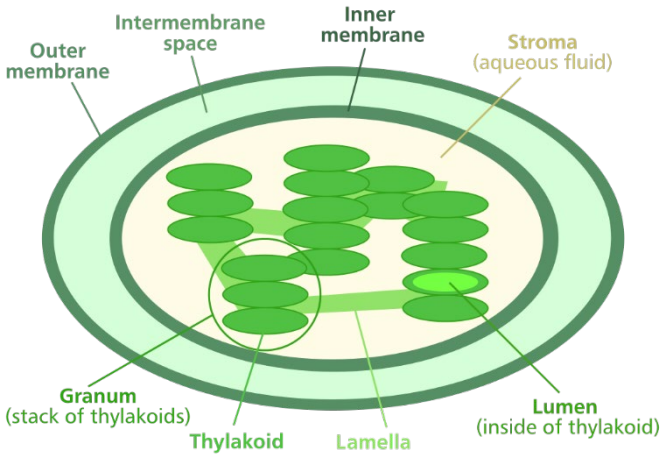



**Figure 7.1. Generalized structure of a chloroplast.**

**Pigments** are molecules that absorb light at specific wavelengths, or colors, and reflect light at other colors (**Table 7.1**). For example, chlorophyll *a* ***absorbs*** mostly red and blue light; in other words, it captures red and blue light energy to make glucose. Chlorophyll *a* also ***reflects*** green and some blue light; this means it appears blue-green. Since chlorophyll *a* and *b* are the predominant pigments in most

plants, most leaves appear green. Other plant pigments include xanthophylls (yellow), carotenoids (orange), and anthocyanins (purple).

| Pigment | Color of pigment | Colors of light most absorbed |
|---|---|---|
| **Chlorophyll *a*** | Blue-green | Red, blue-violet |
| **Chlorophyll *b*** | Yellow-green | Red-orange, green-blue |
| **Xanthophyll** | Yellow | Blue |
| **Carotenoids** | Orange | Green-blue |
| **Anthocyanin** | Purple near pH 7 | Yellow-green near pH 7 |

**Table 7.1. The major plant pigments.**

In this lab you will observe a demo showing the requirements for photosynthesis. You will also isolate pigments from various leaves, and take a closer look at the visible light spectrum.

## Materials & Methods

### ACTIVITY 1: PHOTOSYNTHESIS DEMO

Your instructor will set up a demo showing the three requirements for photosynthesis: $CO_2$, water, and light. This demo works similarly to the lentils part of Lab 6: in that activity, you detected the ***presence*** of $CO_2$ with *phenol red*, a pH indicator that is ***yellow below pH 7*** and ***red above pH 8***. In this demo, we will detect the ***absence*** of $CO_2$ with *bromothymol blue (BTB)*, a pH indicator that is ***yellow below pH 6*** and ***blue above pH 8***. **Table 7.2** shows the four tubes that your instructor will set up.

**Materials**

- four tubes with screw caps, 20x150 mm
- test tube rack
- two sprigs of *Elodea*, ~10 cm long
- aluminum foil
- lamp
- bromothymol blue
- NaOH
- straw

| | Ingredients in each test tube | | | | | *Results* | |
|---|---|---|---|---|---|---|---|
| **TEST TUBE** | ***Elodea*** | **BTB** | **$H_2O$** | **$CO_2$** | **Light** | ***Color change?*** | ***Did photosynthesis happen?*** |
| 1 | ✓ | ✓ | ✓ | ✓ | ✓ | | |
| 2 | | ✓ | ✓ | ✓ | ✓ | | |
| 3 | ✓ | ✓ | ✓ | ✓ | | | |
| 4 | | ✓ | ✓ | ✓ | | | |

**Table 7.2. Activity 1 demo.**

1. Fill each tube halfway with water and add 25 drops of BTB solution to each.
2. Use the straw to gently exhale into the liquid in each tube. This will dissolve $CO_2$ into the liquid. Exhale until the liquid is a pale yellow.
3. Add a sprig of *Elodea* to tubes 1 and 3, cut end down. Screw on caps.
4. Wrap foil around tubes 3 and 4.
5. Place four tubes in tube rack and turn on the lamp so it shines on all tubes.
6. Place a large beaker of water between the rack and the lamp, such that the light shines through the water directly on to the tubes. This will control for different amounts of heat from the light bulb reaching the exposed tubes (1 and 2) versus the covered tubes (3 and 4).
7. After 30 minutes, turn off the lamp and record the results.


**Figure 7.2. Setup for Activity 1 (© Josi Taylor).**

**Questions**

1. What is the purpose of tubes 2 and 4 (the ones without the plant)?

2. What is the purpose of tubes 3 and 4 (the ones covered in foil)?

3. What were the standardized variables (the ones kept constant) in this experiment?

4. What were the independent variables?

## ACTIVITY 2: PAPER CHROMATOGRAPHY

In Activity 2, you will use a technique called chromatography to see which pigments are in three kind of leaves. Chromatography is essentially a way of separating a mixture of molecules into individual components. For example, there is a jar with different colored objects and you want to separate the red marbles from everything else in the jar. You will be doing paper chromatography today. Here is how paper chromatography works:

- the mixture of molecules is applied onto the paper
- the paper is dipped in a solvent that it slowly absorbs
- the solvent moves up the paper and through the mixture of molecules, dissolving them and carrying them along
- the ***more soluble*** molecules stay dissolved in the solvent ***longer***, and are seen ***higher*** on the paper
- the ***less soluble*** molecules stay dissolved for a ***shorter time*** and are seen ***lower*** on the paper

**Materials**

- spinach, purple plum, golden euonymus leaves
- a penny
- chromatography paper
- large test tubes
- cork stopper
- thumbtack
- test tube rack
- chromatography solvent (95:5 pet ether:acetone)

Your group will be assigned one or more types of leaf (spinach, purple plum, golden euonymus).

1. **Apply the pigments to the paper.**
   a. Avoid excessive handling of the paper: grab it by its sides, and do not bend it.
   b. Lay it flat on your bench, over a paper towel.
   c. Place a leaf over the bottom of the paper (the pointy end is the bottom).
   d. About 1 inch from the bottom, roll the coin across the leaf to apply its pigments to the paper. Roll the coin back and forth at least 20 times to get lots of pigment.

2. **Set up the chromatography (Fig. 7.3).**
   a. Use a thumbtack to pin the top of the paper to the bottom of the cork.
   b. Add 5-10 mL of solvent to the large test tube.
   c. Insert the paper into the test tube and seal it with the cork.
   d. You want the pointy end (~½ inch) to be in the solvent, but NOT touching the pigments. If it is not, try again with more solvent.
   e. Leave the test tube in the tube rack and wait until the solvent is about 1 inch from the top.

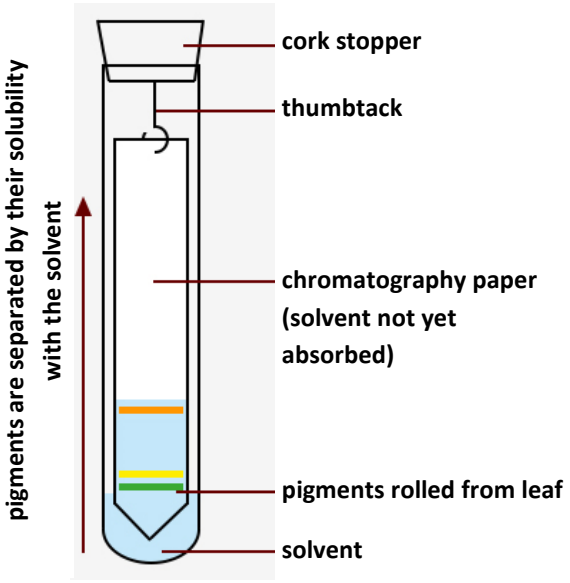



**Figure 7.3. A paper chromatography setup.**

3. **Observe your results, repeat, and clean up.**
   a. When the solvent is within an inch from the top of the paper, remove the paper from the tube and cork and re-stopper the tube to prevent the solvent from evaporating.
   b. Observe your results. Let the paper fully dry before handling.
   c. Repeat the relevant steps (steps 1 and 2, except 2b) for another leaf.
   d. When done, dispose of excess solvent in special waste container.
   e. Do not wash anything – simply return them.

**Questions**

1. Based on your spinach leaf results, list the four pigments in order from least to most soluble in the solvent, and from least to most nonpolar.

| | | | |
|---|---|---|---|
| Most soluble | _____ | Least nonpolar | _____ |
| | _____ | | _____ |
| | _____ | | _____ |
| Least soluble | _____ | Most nonpolar | _____ |

2. Compare the color of a spinach leaf to the pigments on the paper. Which pigments are the most abundant? Does this make sense, given the color of the leaf?

3. Given your answer to #2, which pigment do you think is the most abundant in the golden euonymus?

4. Does the golden euonymus chromatography result support your hypothesis for #3?

5. The golden euonymus leaves must still have chlorophyll. Why is this?

6. The purple plum leaf has anthocyanin. Based on where this pigment ended up on the paper, what can you say about how soluble it is in the solvent, and how polar it is?

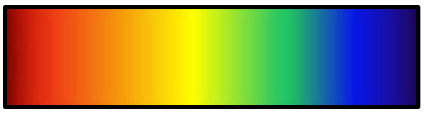
## ACTIVITY 3: LIGHT AND PIGMENTS

In Activity 3, you will take a closer look at the visible light spectrum and how pigments and filters work. Visible or "white" light (from the sun or a typical light bulb) is a mixture of many different colors, or wavelengths, of light. When sunlight hits water droplets in the air, the colors separate into a range from red to violet. This rainbow shows all the colors we can see. As described in **Table 7.1**, plants do not use all colors of visible light equally; some colors are more useful for photosynthesis than others. This is due to the pigments present in a given leaf. Pigments and filters work similarly. Pigments absorb some colors, and reflect other colors (these colors bounce off the pigment). Filters absorb some colors, and let other colors pass through them (they are transmitted through the filter).

**Materials**

- lamp
- spectroscope
- red, green, and magenta filters
- spinach, golden euonymus, and plum leaf extracts
- spinach leaf

1. First observe white light through the spectroscope. It should resemble the spectrum on the right.



2. Now place the **red filter** between the lamp and spectroscope, and observe. Repeat with the **green** and **magenta** filters**. Sketch the spectra you see in the boxes, and label the most prominent colors.**

| Red filter spectrum | Green filter spectrum | Magenta filter spectrum |
|---|---|---|
| | | |

3. Do the same with the **spinach leaf, golden euonymus**, and **plum leaf extracts**. **Sketch the spectra you see in the boxes, and label the most prominent colors.**

| Spinach leaf spectrum | Golden euonymus spectrum | Plum leaf spectrum |
|---|---|---|
| | | |

4. Compare the spectra for the leaf extracts. How do you explain any differences you see?

5. Based on your observations, which colors of light are ***most*** and ***least*** important for photosynthesis?

# LAB 8 Mitosis & Meiosis

## Objectives

At the end of the lab, you should be able to:

1. Identify the phases of mitosis.
2. Describe the major events of mitosis and meiosis.
3. Explain the purpose of mitosis and meiosis.
4. Describe how animal and plant cell mitosis are similar and different.

## Introduction

All living things must make more cells for its own survival, and possibly for creating offspring. Multicellular organisms must make new cells for growth and repair. In a process called **mitosis**, one body cell (known as a **somatic cell**) makes copies of everything, ensures they are distributed equally, then divides ***once*** into ***two*** **daughter cells**. It is critical that all the duplicated DNA (condensed into **chromosomes**) is distributed equally to the two daughter cells so they remain genetically identical. Mitosis in plant and animal cells is very similar but not identical (see **Table 8.1**).

| | **Animal cells** | **Plant cells** |
|---|---|---|
| **Mitotic spindle formation** | requires a pair of centrioles | does not require centrioles (plant cells do not have them) |
| **Cytokinesis** | during telophase, the plasma membrane begins to form a **cleavage furrow** that eventually pinches the cell in two | during telophase, vesicles form and fuse to make a **cell plate** that eventually splits the cell in two |

**Table 8.1. Differences between animal and plant cell mitosis.**

In addition to mitosis, many species must also make special reproductive cells to generate offspring. This happens through a process called **meiosis**, in which one **germ cell** (also known as a sex cell) makes copies of everything, ensures they are distributed equally*, then divides ***twice*** into ***four*** **daughter cells**. There are two events during meiosis that shuffle the chromosomes around; thus, these four daughter cells are genetically distinct. They also contain half the amount of DNA as the original germ cell (before it duplicated its DNA).

Mitosis and meiosis are very visual processes; as such, this lab is decidedly visual in nature. All three parts to this lab involve viewing the different phases of mitosis and meiosis, with an emphasis on mitosis.

*even though the four daughters end up genetically different, the cell must still ensure the chromosomes themselves are distributed equally

## Materials & Methods

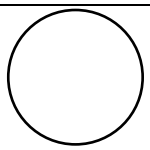
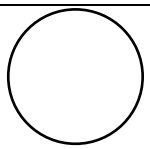
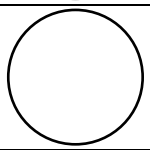
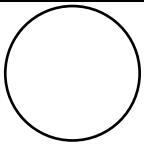
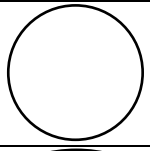
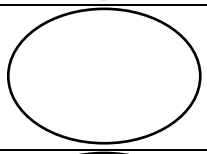
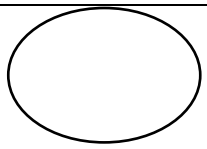
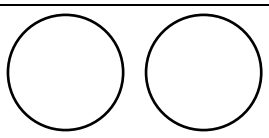
### ACTIVITY 1: VIDEOS

**Materials**

- Amoeba Sisters videos: MITOSIS MEIOSIS
- Crash Course videos: MITOSIS MEIOSIS

Your instructor will show you at least one mitosis video and one meiosis video. As you watch the video on mitosis, start answering the question below.

**Describe the major events of these parts of the cell cycle, and sketch what a cell would look like. Assume that this cell has TWO chromosomes before DNA duplication.**

| | Sketch of cell | Major events |
|---|---|---|
| **Before DNA duplication** |  | |
| **After DNA duplication** |  | |
| **Prophase** |  | |
| **Prometaphase** |  | |
| **Metaphase** |  | |
| **Anaphase** |  | |
| **Telophase** |  | |
| **Cytokinesis** |  | |

## ACTIVITY 2: MICROVIEWER

**Materials**

- microviewer and filmstrips

In Activity 2 you will use a microviewer (essentially a magnifying glass) to look at filmstrips containing images of animal and plant cells in some stage of mitosis.

**The filmstrip frames are numbered. Which phase of mitosis is shown in the following frames?**

| | Animal cell filmstrip | | Plant cell filmstrip |
|---|---|---|---|
| #1 | _____ | #2 | _____ |
| #3 | _____ | #3 | _____ |
| #6 | _____ | #5 | _____ |
| #7 | _____ | #7 | _____ |

**Compare the animal cell in metaphase with the metaphase sketch you made in Activity 1. What differences do you notice?**

## ACTIVITY 3: WHITEFISH AND ONION CELL COUNTS

**Materials**

- microscope, whitefish blastula (animal) slide, and onion root tip (plant) slide

In Activity 3 you will view two slides with the microscope. These specimens are good for viewing cells in mitosis because these cells are actively dividing (i.e. continuously going through the cell cycle).

**By now you should be able to identify cells in any phase of the cell cycle. Focus on each slide with the 40x objective, and count how many cells are in each phase. Do this for all cells in one field of view. Record your group totals as well.**

| | Cell cycle stage | I | P | M | A | T | Total |
|---|---|---|---|---|---|---|---|
| Whitefish (animal) | # of cells (individual) | | | | | | |
| | # of cells (group) | | | | | | |
| | % of cells in each stage (group) | | | | | | |
| Onion root (plant) | # of cells (individual) | | | | | | |
| | # of cells (group) | | | | | | |
| | % of cells in each stage (group) | | | | | | |

## Extra Questions – your instructor may assign them

1. In what phase of mitosis do sister chromatids separate?

2. The Introduction mentions that "there are two events during meiosis that shuffle the chromosomes around". What are these two events called?

3. Complete the table below.

| | Mitosis | Meiosis |
|---|---|---|
| **# of cell divisions** | | |
| **# of cells produced** | | |
| **Are daughter cells identical?** | | |
| **# of sets of chromosomes in daughter cells** | | |

4. Based on your answers in Activity 3, in which phase of the cell cycle do cells spend most of their time? (circle one: interphase, prophase, metaphase, anaphase, telophase)

5. What is one advantage of sexual reproduction? Here, reproduction means producing offspring.

6. What is one advantage of asexual reproduction? Here, reproduction means producing offspring.

# LAB 9 Genetics

## Objectives

At the end of the lab, you should be able to:

1. Define all terms in bold on this page.
2. Explain the inheritance of traits controlled by one gene:
   a. when one allele is dominant and the other is recessive;
   b. when one allele is dominant and the other is recessive, and the gene is X-linked;
   c. when two alleles are codominant to each other.

## Introduction

In this lab we turn to genetics and inheritance. Recall that a **gene** is a set of instructions used to build a protein (**Fig. 9.1**). We have two copies of each of our ~21,000 genes. We inherited one copy from mom and the other copy from dad. The combination we receive (the **genotype**) determines the outward expression or appearance of that gene (the **phenotype**). Different versions of the same gene are known as **alleles**; thus, from each of our parents we inherit ***one allele for each gene***.

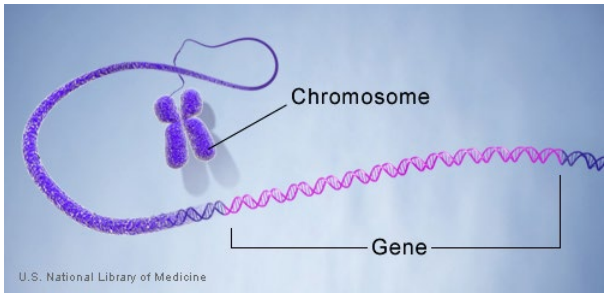



**Figure 9.1. Genes are part of chromosomes.**

Generally, alleles and phenotypes can be described as **dominant** or **recessive**. What do these terms mean? A dominant allele is one where you only need one copy of it to show the dominant phenotype. A recessive allele is one where you need two copies of it to show the recessive phenotype. **Fig. 9.2** has an example: in pea plants, purple flowers are dominant to white flowers. A plant with purple flowers can be **homozygous dominant** (AA) or **heterozygous** (Aa), but a plant with white flowers must be **homozygous recessive** (aa). Capital letters are used to write dominant alleles; lower-case letters for recessive alleles.


**Figure 9.2. Pea plants have either purple flowers or white flowers.**

Some things to keep in mind regarding the term ***dominant***:

- dominant alleles/traits ***are not better than*** recessive alleles/traits
- dominant phenotypes ***are not always more common than*** recessive phenotypes
- some diseases ***are caused by*** dominant alleles (i.e. not all disease alleles are recessive)

## Materials & Methods

**ACTIVITY 1: THE GENETICS OF TASTE**

In Activity 1 you will determine whether you can taste a molecule called phenylthiocarbamide, or PTC. The ability to taste PTC is determined by one gene with two common forms, or alleles. Tasting (T) is dominant to non-tasting (t). By doing a quick taste test, you can figure out your phenotype and possible genotypes for this trait. More information on this gene and its evolutionary significance is available at https://learn.genetics.utah.edu/content/basics/ptc/.

**Materials**

- PTC and control test strips

1. Place a **control** test strip on the tip, back, and sides of your tongue. Repeat with a **PTC** test strip.

**Questions**

1. Complete the table below. Circle your phenotype and possible genotypes.

| What are the three PTC tasting genotypes? | What are their phenotypes? |
|---|---|
| | |
| | |
| | |

2. Jack is a PTC non-taster, and his mother and father are both PTC tasters.
   a. Complete a Punnett square to determine the expected phenotypic ratio among Jack's siblings.

   b. In this family, what are the chances that a child will be able to taste PTC?

   c. What are the chances that a child will not be able to taste PTC?

## ACTIVITY 2: COLOR BLINDNESS

In Activity 2 you will consider red-green color blindness, a recessive trait determined by one gene with two alleles. This gene is located on the X chromosome, which changes its inheritance patterns. Traits determined by genes on the X chromosome are called **X-linked traits**.

Things to keep in mind:

- females have two X chromosomes (XX); males have one X and one Y (XY)
- write the alleles like this: $X^B$ = color vision allele; $X^b$ = color blind allele

### Materials

- color blindness test images

1. Look at the images and see if you can read the number or pattern in each.

### Questions

1. Complete the table below. Circle your phenotype and possible genotypes.

| Possible phenotypes | What is their genotype? |
|---|---|
| Female with color vision, homozygous | |
| Female with color vision, heterozygous | |
| Female with color blindness | |
| Male with color vision | |
| Male with color blindness | |

2. A female has color vision. How would you figure out her genotype?

3. Would you expect to find more color blind males or females in the population? Why?

## ACTIVITY 3: ABO BLOOD TYPES

In Activity 3 you will consider the ABO blood groups in humans. There are four blood types in the ABO group: A, B, AB, and O (the phenotypes). As with most genes, people have two copies of the ABO blood type gene. There are two important differences here:

1. There are three different alleles: A, B, and o. Sometimes these are written as $I^A$, $I^B$, and i.
2. A and B are both dominant to o, and they are ALSO **codominant** to each other.

What is codominance? It is a phenomenon in which a heterozygote with two different codominant alleles shows both phenotypes. One allele does not mask the other; they both "show through" in the phenotype.

For the ABO blood group, the actual phenotype is some combination of glycoproteins on the surface of the red blood cells. An A allele means having type A glycoproteins, a B allele means having type B glycoproteins, and an o allele means having no glycoproteins.

**Complete this table showing all possible genotypes and phenotypes for the ABO blood group.**

| Genotype | Heterozygous or homozygous? | What is their phenotype? (blood type) |
|---|---|---|
| AA | | |
| | heterozygous | A |
| | homozygous | B |
| Bo | | |
| | | AB |
| oo | | |

## ACTIVITY 4: LET'S HAVE A BABY

In Activity 4 you are going to experience the process of inheritance first-hand. How? By having a baby (on paper) with your lab partner! You will determine your genotype for five traits plus sex, and track which alleles your baby inherits from its two parents. These traits are described in **Table 9.1**.

### Materials

- popsicle sticks and stickers

| Trait | Mode of inheritance |
|---|---|
| **PTC taster** | PTC tasting (TT or Tt) is dominant to non-tasting (tt) |
| **widow's peak** (hair comes to a point on the forehead) | having a widow's peak (WW or Ww) is dominant to not having one (ww) |
| **attached earlobes** (ears do not form a free-hanging lobe) | free earlobes (EE or Ee) is dominant to attached earlobes (ee) |
| **tongue rolling** | tongue rolling (RR or Rr) is dominant to non-rolling (rr) |
| **freckles** | having freckles (FF or Ff) is dominant to not having freckles (ff) |
| **sex** | females are XX, males are XY |

**Table 9.1. A description of the traits you are tracking in Activity 4.**

1. Determine your phenotypes and genotypes for the traits in **Table 9.1**. Write them below.

| Trait | Your phenotype | Your possible genotypes |
|---|---|---|
| PTC taster | | |
| widow's peak | | |
| attached earlobes | | |
| tongue rolling | | |
| freckles | | |
| sex | | |

2. If you have any dominant phenotypes, ***assume you are heterozygous*** for those traits.

3. For each trait:
   a. Get one popsicle stick and two stickers (6 sticks and 12 stickers total).
   b. Write one allele on one side, and the other allele (for the same trait) on the other side.
   c. Write your initials on the stickers and put one on each side of every stick. See **Fig. 9.3** for what a stick should look like.

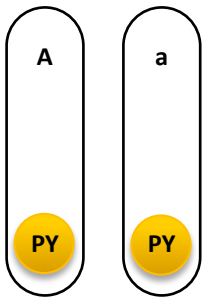



**Figure 9.3. Example of two sides of the same stick labeled for a heterozygous genotype (Aa). Initials are on a sticker at the bottom (my initials are PY).**

4. Time to make a baby! Stand up, hold your sticks up high, and drop them onto the lab bench. Your lab partner should do the same.

5. Write down your results in the table below.

| Trait | Child's genotype | Child's phenotype |
|---|---|---|
| PTC taster | | |
| widow's peak | | |
| attached earlobes | | |
| tongue rolling | | |
| freckles | | |
| sex | | |

**What is your child's name?**

**Does your child take after you more, or the other parent? Briefly explain.**

## ACTIVITY 5: DOES SUNSCREEN PROTECT MY DNA, PART 1

***(adapted from the Genetic Science Learning Center, https://teach.genetics.utah.edu/content/dna/)***

In Activity 5 you will set up an experiment and view the results next week. This experiment deals with DNA, the molecule of heredity. Genes are made of DNA. Some genes code for proteins that repair DNA when it gets damaged by things such as ultraviolet (UV) radiation. Overexposure to UV radiation can negatively affect your health in many ways, aside from the usual sunburns:

- increased risk of developing skin cancer
- premature aging of the skin, causing it to become thick, wrinkled, and leathery
- cataracts in the eyes that can lead to blindness
- suppression of the immune system

Sunscreen is an easy way to protect yourself from the sun's UV rays. It acts like a shield to absorb the UV radiation before it enters your skin cells. In this activity, you will use baker's yeast (*Saccharomyces cerevisiae*, the same one you used in Lab 6) to investigate the effect of UV and various sunscreens on their growth. To be continued in the next lab!

**Materials**

- three Petri dishes
- UV-sensitive yeast
- clear tape
- one type of sunscreen
- one Sharpie

YOUR GROUPS FOR ACTIVITY 5 ARE EACH ROW OF LAB BENCHES

1. **Clean your hands and work area.**
   a. Wash your hands with soap and water.
   a. Wipe your hands and your work area with an alcohol wipe.

2. **Get one Petri dish and label it, along the outside edge of the bottom side (see Fig. 9.3).**
   a. Write your row number and the day and time of your lab (e.g. Row #1 Mon 8:00).
   b. Label each dish with the condition the yeast will be growing (e.g. no sun, sun, or sun + sunscreen).
   c. Decide which sunscreen you will test.

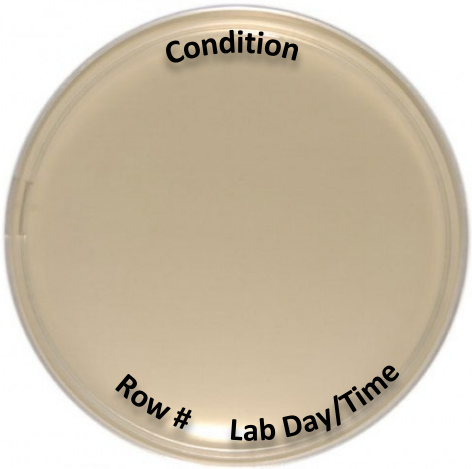


**Figure 9.3. Label your Petri dish like this.**

3. **Spread yeast cells on the culture medium in the Petri dish.**
   a. Your instructor will dispense 1 mL of yeast solution onto your medium.
   b. Gently tilt and rotate your dish to spread the liquid across the surface. Make sure the entire surface is covered.
   c. If there are places the liquid does not cover, use the rounded end of a sterile toothpick to move the liquid over them.

4. Place the Petri dish in a **dark place** with the **lid on** for 10-20 minutes to let the liquid soak into the medium.

5. **Tape the lid to the bottom**. Use small pieces of clear tape along the sides. Do not put tape on the lid!

6. **Set up your "no sun" and "sun + sunscreen" dishes.**
   a. For your "no sun" dish: tape a piece of dark paper on the lid. Keep the tape on the sides.
   b. For your "sun + sunscreen" dish: spread a thin, even layer of sunscreen on the lid.

7. **Expose the Petri dish to the sun or a UV light.**
   a. Your instructor will tell you how long to expose your Petri dish.
   b. If you are using the sun, leave the plate so the surface is aimed directly at the sun.

8. **Let the yeast grow until next week.**
   a. Wipe off the sunscreen from the lid.
   b. Place the Petri dishes upside down in the plate rack.

The yeast will grow for several days, and you will look at the results next week.

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## Genetics Practice Problems

**Name & Lab Section:**

1. In cats, short hair is dominant to long hair (angora). Set up ***Punnett squares*** for the following crosses, AND indicate the ***phenotypic ratio*** for them.

   SS x ss  Ss x Ss  ss x ss  Ss x ss

2. In humans, brown eyes are usually dominant to blue eyes. Suppose a blue-eyed man marries a brown-eyed woman whose father had blue eyes. ***What proportion*** of their children is expected to have ***blue eyes***?

3. In pea plants, yellow pea color is dominant to green. What are the expected ***phenotypic ratios*** of the offspring in the following crosses?

   Homozygous yellow x homozygous green

   Heterozygous yellow x homozygous green

   Heterozygous yellow x heterozygous yellow

   Heterozygous yellow x homozygous yellow

4. Two long-winged flies were mated. The offspring consisted of 77 with long wings and 24 with short wings. Are short wings a dominant or recessive trait? What are the genotypes of the parents?

5. Tongue rolling is a dominant trait. A man-woman couple can roll their tongues, but their son cannot. What are the genotypes of all three individuals? Support your answers with a Punnett square.

6. In corn plants, purple kernels are dominant to yellow, and smooth kernels are dominant to wrinkled. A double heterozygous plant is crossed to a heterozygous purple, wrinkled plant. What are the phenotypic ratios for their offspring?

7. Hemophilia is a recessive X-linked disorder ($X^H$ = healthy allele, $X^h$ = hemophilia allele). A girl has hemophilia. What are the possible genotypes of her parents?

8. Snapdragon flowers occur in three color variations: white, red, and pink. Color is controlled by one gene with two incompletely dominant alleles: red (R) and white (W). A heterozygote (RW) has pink flowers. Determine the genotypes and phenotypes for the following crosses:

   RR x WW RR x RW RW x WW

9. The ABO blood types are determined by one gene with three alleles: A and B are codominant to each other, and also dominant to o. This results in four phenotypes: type A, type B, type AB, and type O. Determine the genotypes and phenotypes for the following crosses:

   AA x oo Ao x Bo

10. Mendel believed that hereditary factors were always either dominant or recessive. How might he have changed his view had he performed the crosses with snapdragons in question 8?

# LAB 10 Gene Expression

## Objectives

At the end of the lab, you should be able to:

1. Describe the structure and function of DNA.
2. Explain the overall process of gene expression.
3. Describe the effect of UV radiation on DNA.

## Introduction

How does a genotype cause a phenotype? In this lab we see how the information in a gene (made of DNA) is used to build a protein. This process is known as **gene expression** and has two major steps:

1. **Transcription (DNA → RNA):** an enzyme unwinds the double helix, reads one strand of DNA, and builds the complementary strand using RNA nucleotides
2. **Translation (RNA → protein):** an enzyme reads the RNA and builds the corresponding strand of amino acids, which folds up into a protein

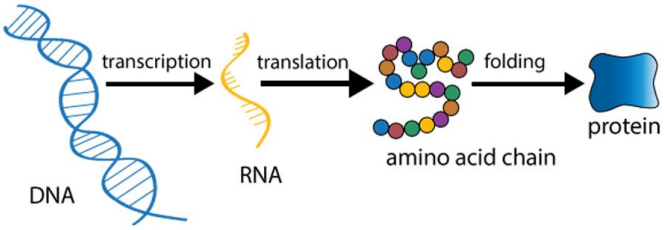



**Figure 10.1. An overview of gene expression.**

TRANSCRIPTION: In the nucleus, the cell's machinery copies the gene sequence into messenger RNA (mRNA), a molecule that is similar to DNA. Like DNA, mRNA has four nucleotide bases - but in mRNA, the base uracil (U) replaces thymine (T).

TRANSLATION: The protein-making machinery, called the ribosome, reads the mRNA sequence and translates it into the amino acid sequence of the protein. The ribosome starts at the sequence AUG, then reads three nucleotides at a time. Each three-nucleotide codon specifies a particular amino acid. The "stop" codons (UAA, UAG and UGA) tell the ribosome that the protein is complete.

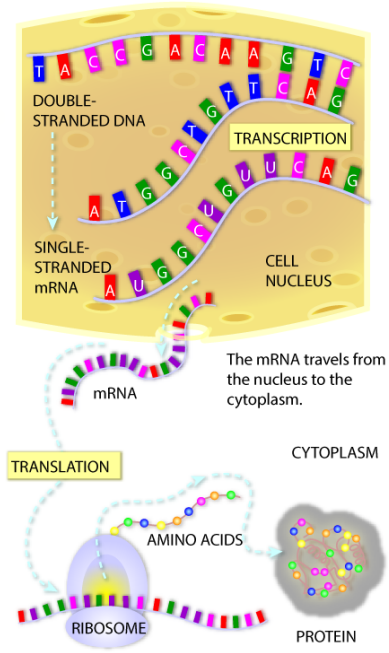



**Figure 10.2. A more detailed overview of gene expression.**

The first step is called **transcription**, and part of it is shown in **Fig. 10.3**:

1. An enzyme called **RNA polymerase** binds to the beginning of a gene in a region called the **promoter**
2. RNA polymerase begins unwinding the double helix and ***transcribing*** RNA—that is, reading one strand to build the complementary strand with RNA nucleotides
3. After it transcribes a region called the terminator, RNA polymerase releases the RNA and transcription stops

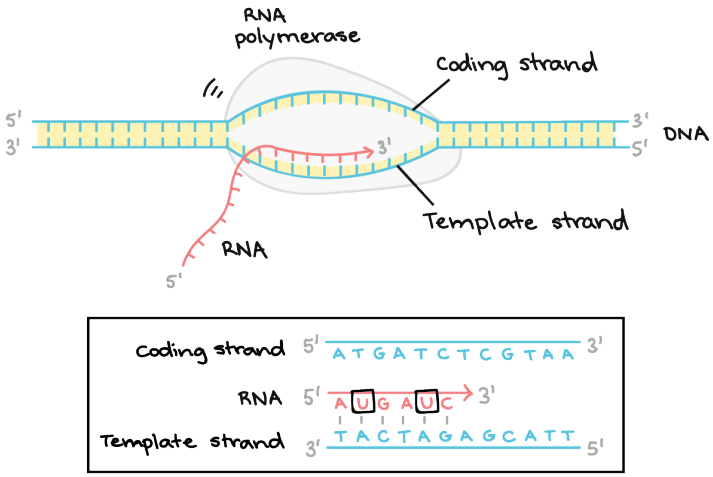



**Figure 10.3. An overview of transcription.**

The second step is called **translation**, and part of it is shown in **Fig. 10.4**:

1. An enzyme called a **ribosome** binds to the beginning of the RNA strand and begins reading
2. When it reads the first AUG, it begins ***translating*** the RNA—that is, reading one triplet (also known as a **codon**) and translating the nucleotide language into amino acids
3. Each new codon corresponds to another amino acid
4. After the ribosome reads a stop codon, it falls off the RNA and translation stops

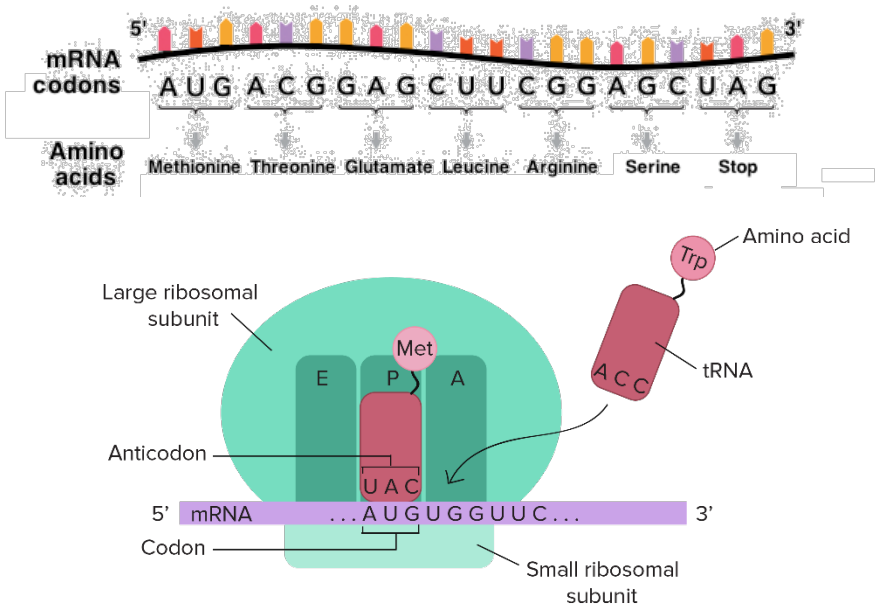



**Figure 10.4. An overview of translation (above), with the ribosome included (below).**

## Materials & Methods

### ACTIVITY 1: STRAWBERRY DNA EXTRACTION

In Activity 1 you will extract DNA from a strawberry. This is a very easy procedure that you can do at home with household ingredients! Being able to hold DNA in your hands and see it may help you make connections with the importance of this molecule that lives in all our cells.

**Materials**

- one strawberry
- mortar and pestle
- cell lysis solution
- beaker
- cheese cloth
- 15 mL conical tube
- long cotton swab
- 70% ethanol
- 1.5 mL tube

1. Place one strawberry in a mortar and begin mashing it with a pestle for about 20 seconds.

2. Add 5 mL of cell lysis solution and continue grinding for another 30 seconds.

3. Place a single layer of cheese cloth over a beaker. Pour the contents from the mortar into the beaker. The cheese cloth will filter out larger pieces. You may need to use your hands to squeeze the liquid through the cloth. When done, throw away the cheese cloth.

4. Collect 2 mL of the strawberry filtrate in a clear 15 mL conical tube.

5. Carefully add 5 mL of ice-cold ethanol down the side of the tube, taking care not to mix the layers. The DNA should precipitate quickly.

6. Use the handle of a long cotton swab to fish out the DNA and place it in a smaller 1.5 mL tube.

7. Rinse the mortar and pestle and leave it on the drying tray.

8. Review the model of DNA.

**Questions**

1. Describe the appearance of the strawberry DNA.

2. What do you think is the purpose of the cell lysis solution?

3. Label all parts of the DNA double helix on the image.

**ACTIVITY 2: GENE EXPRESSION**

In Activity 2 you will first go through a brief computer simulation of gene expression. Then you will go through the steps of gene expression on paper by answering the questions.

1. With either your own or a school laptop, go through the gene expression simulation at https://learn.genetics.utah.edu/content/basics/transcribe/.

2. Now go through a similar process here. Below is the sequence of a DNA double helix. ***Reading the top strand***, transcribe a messenger RNA (mRNA) molecule.

AGGCTACGGTGCCCAGTAGAGCCTGCACTCTTAGCT

mRNA:

TCCGATGCCACGGGTCATCTCGGACGTGTGAATCGA

3. Your mRNA will now be translated. Refer to the genetic code in **Fig. 10.5**.
   a. Copy your mRNA strand below for convenience.
   b. Find and underline the first AUG you see. This is the start codon and the ribosome begins translation here.
   c. Underline every codon (three bases) after the AUG. The ribosome reads each codon to know which amino acid should come next.
   d. Translate the mRNA. Write the 3-letter abbreviation for each amino acid below each codon.

mRNA:

protein:

4. How many amino acids is your protein? _____

5. As you can see in **Fig. 10.4**, a molecule called a **transfer RNA (tRNA)** brings the amino acids to the ribosome. A tRNA forms base pairs with the mRNA codon with its **anticodon**.
   a. The start codon is AUG. What is the anticodon in its corresponding tRNA? _____

   b. If a tRNA anticodon is CCA, what amino acid does it bring to the ribosome? _____

6. Let's go back to DNA for a minute. If DNA analysis of a gene shows it has 20% adenine (A), what percent of the other bases does it have?

   Cytosine (C): _____ Guanine (G): _____ Thymine (T): _____ Uracil (U): _____

7. Now for a sense of size. Rank the following from smallest (1) to largest (6):
   gene, cell, chromosome, atom, nucleus, nucleotide

**ACTIVITY 3: DOES SUNSCREEN PROTECT MY DNA, PART 2**
***(adapted from the Genetic Science Learning Center, https://teach.genetics.utah.edu/content/dna/)***

Last week you set up an experiment testing the effect of various sunscreens on yeast. Recall that sunscreen is designed to prevent UV radiation from damaging DNA. Now that you are learning how one's genetic makeup (genotype) leads to a trait (phenotype), we can think about this experiment in more depth.

Like humans, yeast have genes that code for DNA repair proteins. Their job is to repair DNA damaged by UV (and other things). In order for the experiment to work, you used a yeast strain in which some of these genes were mutated. This means the repair proteins don't work, and these yeast are more sensitive to the effects of UV exposure. UV-damaged DNA is essentially mutated DNA, and if these new mutations occur in essential genes, the cell dies. By using these UV-sensitive yeast, we can observe how much DNA damage occurs when cells are exposed to, or protected from, UV radiation.

**Materials**

- yeast plate from last week

Your yeast have grown for several days. Take a look at your plate and answer these questions.

**Questions**

1. Do you see any differences between areas of your plate? If so, describe them.

2. Did one sunscreen protect the yeast cells better than others? If so, which one?

3. Why did the yeast grow more in some areas than others?

4. What can you conclude from the results of your experiment?

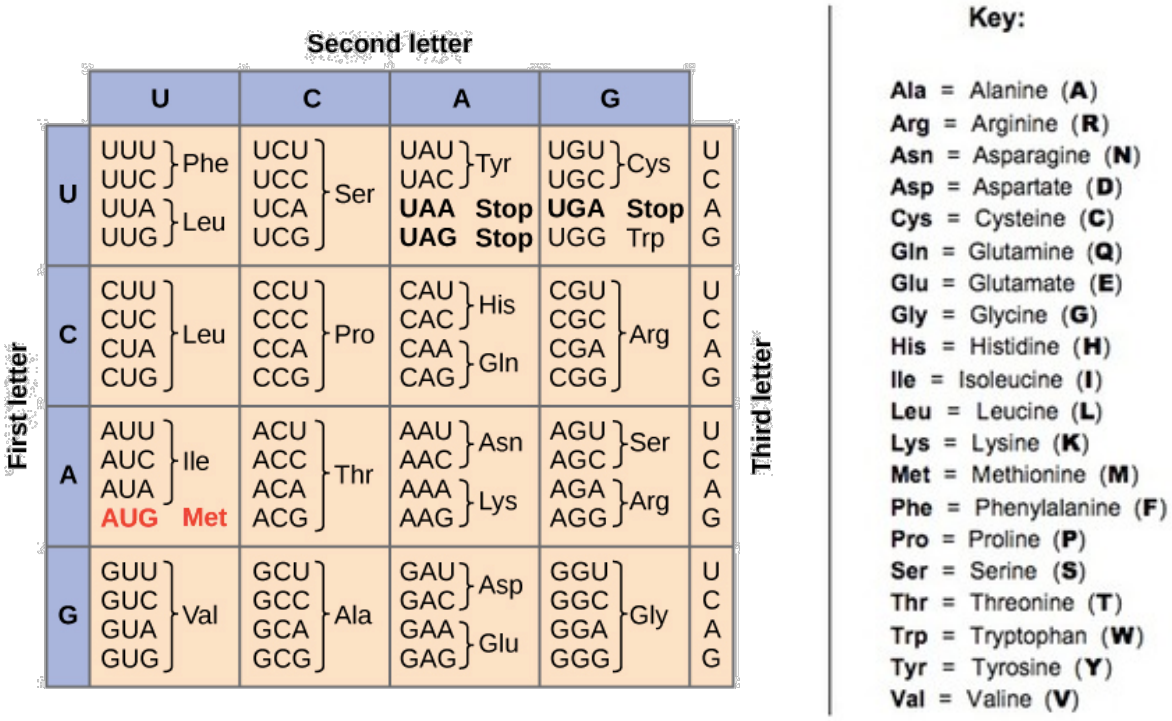



**Figure 10.5. The genetic code.**

LAB
# 11
# Evolution & Comparative Anatomy

## Objectives

At the end of the lab, you should be able to:

1. Define the terms evolution, variation, and natural selection.
2. Explain how comparative anatomy provides evidence to support evolutionary theory.
3. Describe the differences between homologous and analogous structures.
4. Construct arguments and make claims using evidence from class discussion and a short film.
5. Use data to make predictions about the effects of natural selection.

## Introduction

In the last part of the course, we turn to evolution and ecology. The simplest definition of **evolution** is ***change over time***. What's changing is the genetic makeup of a population. We will refine this definition in the next weeks. For now, know that it is the population that changes over time, NOT the individual.

Why does the genetic makeup of a population change over time? The simplest answer here is: because the environment changes. In any population, there are differences in traits (yellow peas and green peas, narrow beaks and broad beaks, etc.). These **variations** may be more or less beneficial to the individual. Those possessing traits which help them survive longer will reproduce more, potentially passing on those traits to the next generation. However, as their environment changes, so do the variations that will boost survival and reproduction. This means that populations constantly change in their genetic makeup, because of changes ***to their environment***. This is known as **natural selection**.

A key line of evidence to support the theory of evolution by natural selection is comparing the anatomy of different species. **Comparative anatomy** looks at the bones and organs of different species to learn more about them. Sometimes these structures are similar in different species, but often have different functions. These are called **homologous structures** and suggest the species evolved from a common ancestor. When two parts have similar functions but are structurally different, they are called **analogous structures**. They suggest a phenomenon known as ***convergent evolution***, in which two species from different ancestors evolved the same trait. See **Fig. 11.1** for examples.

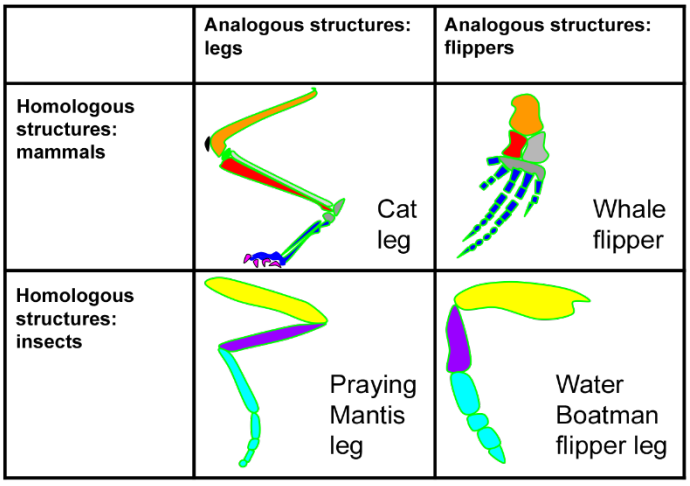



**Figure 11.1. Examples of homologous structures (rows) and analogous structures (columns).**

A special case of homologous structures can arise when one species has a smaller and sometimes non-functioning version of a structure. These are called **vestigial structures**, as they represent the vestiges of a structure that had an essential function in an ancestral species.

## Materials & Methods

### ACTIVITY 1: COMPARATIVE ANATOMY

In Activity 1 you will examine homologous and analogous structures in more detail. You will also consider vestigial structures.

**Materials**

- chicken, cat, and human skeletons with labeled keys

**Part 1 – Homologous Structures**

**Go to the various skeletons and write down the names of the labeled bones.**

**Chicken**

| | | |
|---|---|---|
| 1. | 5. | 8. |
| 2. | 6. | 9. |
| 3. | 7. | 10. |
| 4. | | |

**Cat**

| | | |
|---|---|---|
| 11. | 15. | 18. |
| 12. | 16. | 19. |
| 13. | 17. | 20. |
| 14. | | |

**Human**

| | | |
|---|---|---|
| 21. | 25. | 28. |
| 22. | 26. | 29. |
| 23. | 27. | 30. |
| 24. | | |

**Try to explain the differences between the three skeletons for the structures listed below. Consider where the animal usually lives, how it obtains food, and typical movements it makes.**

1. Leg bones (femur, tibia/fibula, tarsals)

2. Pelvic bones (ilium, ischium, pubis)

3. Upper limb bones (scapula and clavicle)

4. Arm bones (humerus, radius, ulna)

**Part 2 – Vestigial Structures**

**Vestigial structures are like homologous structures, only one of them is smaller and has no essential function. Say whether <u>the first</u> of each pair is a vestigial structure, and briefly explain why.**

1. <u>The human appendix</u> vs. other mammals' appendix (mammals that eat mostly plants have a much larger appendix which helps digest their food)

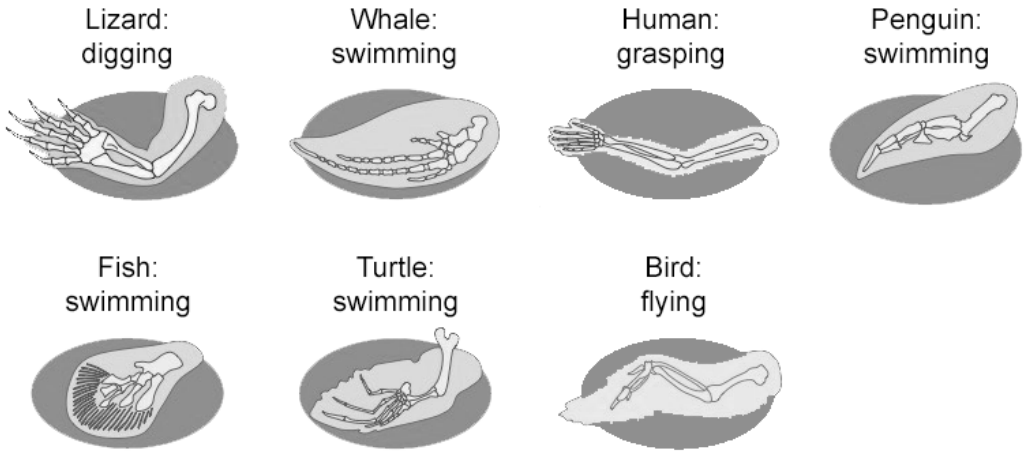
2. <u>Whale phalanges</u> (finger bones) vs. human phalanges (whale flippers are used for swimming, human hands are used for grasping)

3. <u>Modern human wisdom teeth</u> (third molars) vs. early human wisdom teeth (skulls of human ancestors had larger jaws with more teeth, which were possibly used to help chew plant material)

4. Please come up with an example of two homologous structures, one of which is vestigial. Briefly explain why it is vestigial when compared to the other structure.

**Part 3 – Analogous Structures**

**Look at the structures in the image below. Identify one pair of homologous structures and one pair of analogous structures. Explain why they are homologous or analogous.**




Homologous structures:

Analogous structures:

**ACTIVITY 2: GALÁPAGOS FINCHES, PART I**

In Activity 2 you will consider the effect of natural selection on one of the Galápagos Islands. There are 13 different species of finch on the Galápagos Islands off the coast of Ecuador. On one of the islands, Daphne Major, biologists Peter and Rosemary Grant have devoted many years to studying four of these bird species. The Grants have studied the effects of drought and periods of plenty on the finches, and the results of their experiments have had an enormous impact on evolutionary science.

Today, you will first analyze the characteristics of the 13 finch species found on the Galápagos Islands. Then you will begin watching a short film about the research conducted by the Grants. Based on the information in the film *The Origin of Species: The Beak of the Finch* (https://youtu.be/mcM23M-CCog) and your own observations, you will construct an argument and make predictions about the role of natural selection on the evolution of finch populations.

**Materials**

- set of 13 finch cards
- butcher paper
- blue painter's tape
- index cards & sticky notes
- color pens (optional)
- camera (optional)

Break into teams as instructed. Each team will get a set of 13 cards with pictures of different finch species. Begin Part 1, and record observations and answers as you go.

**Part 1 – What Do You Already Know?**

1. Working with your team, examine the cards of the Galápagos finches and arrange the species into groups based on their characteristics. Grouping species according to shared characteristics can provide clues to how they have evolved.

2. On a large poster board or butcher paper, use the tape to attach the cards according to the groups your team has created.
    a. Give each group an informative name and write that name down next to each group.
    b. On index cards, list the evidence for each grouping, and tape them next to the group.

3. Pause for a gallery walk. Walk around the class and examine the displays by the other teams. Pay attention to the following:
    a. How were other teams' groupings similar or different to those of your team?

    b. What evidence did your classmates use to justify their groupings?

    c. How does the evidence they provided support their groupings?

4. On your own, write one question about each team's presentation on sticky notes. Initial your feedback, and stick them next to the team's poster.

5. Based on what you observed during your gallery walk:
    a. Does your team want to make any changes to your own groupings? Make your changes and write down your rationale for revising (or not revising) them.

    b. What additional evidence would you need to better justify your team's groupings? Write this on your poster.

**Part 2 – Sorting Finch Groups**

1. Watch the first part of the film (https://youtu.be/mcM23M-CCog), from the beginning to 5:42. As you watch, answer the following:
    a. What do the different beaks tell us about the different finch species?

    b. What are the two hypotheses for how the Galápagos ended up with so many species of finches?

    c. What evidence did scientists use to rule out one of the hypotheses?

    d. Why is the supported hypothesis important for understanding the effects of natural selection on these species?

2. Let's discuss what you just watched.

3. Return to your finch groupings from Part 1. Work with your team and, if necessary, rearrange the bird groupings based on what you heard in the film. Then, use the information from the film to revise your names and evidence. Answer the following:
    a. What did you change?

    b. What evidence from the film convinced you to make the change?

    c. What do the different groups of finches that you created represent?

4. Put your names on your poster and leave them on the front counter. To be continued next week!

# LAB 12 Natural Selection

## Objectives

At the end of the lab, you should be able to:

1. Use data to make predictions about the effects of natural selection.
2. Produce a bar graph to illustrate predicted results and compare them to actual results.
3. Explain how speciation can occur.
4. Describe the conditions required for a Hardy-Weinberg equilibrium population.
5. Use the Hardy-Weinberg equations to calculate phenotype and genotype frequencies.

## Introduction

In Lab 12 you will continue examining the finch species of the Galápagos Islands. Recall from Lab 11 that the research of Peter and Rosemary Grant focuses on four of the 13 species of finch currently living on the islands. The rest of the film deals with how environmental changes led to **natural selection** on these finch populations.

It can be challenging to study the genetic makeup of a population, due to the myriad natural selection pressures that influence which traits are most favorable. British mathematician G. H. Hardy and German physician Wilhelm Weinberg came up with a mathematical model to find the allele and genotype frequencies of a population. In order for their model to hold, they had to make many assumptions about the population:

1. No mutations
2. No gene flow
3. Infinitely large population size (to prevent genetic drift)
4. Random mating
5. No natural selection

A population in which these five things are true is said to be at **Hardy-Weinberg equilibrium**. In reality, no population exists at Hardy-Weinberg equilibrium because none of these five things are true. However, it is still useful to assume they are, because it allows us to use the **Hardy-Weinberg (H-W) equations** to estimate the allele and genotype frequencies of the population. These estimates also provide a baseline against which to measure the amount of genetic change (in other words, the estimates serve as a control group).

The simplest case to consider is tracking one gene with two alleles, in which one is completely dominant (A) and the other is completely recessive (a). The two forms of the H-W equation, and their explanations, are in **Table 12.1**. It may be useful to think of the H-W equations like a Punnett square, ***but for a population*** (see **Figure 12.1**).

| $p + q = 1$ | $p^2 + 2pq + q^2 = 1$ |
|---|---|
| Deals with **allele frequencies** in a population | Deals with **genotype frequencies** in a population |
| ***p*** = the frequency of the **dominant** allele in the population | $p^2$ = the frequency of the **homozygous dominant** genotype in the population |
| ***q*** = the frequency of the **recessive** allele in the population | ***2pq*** = the frequency of the **heterozygous** genotype in the population |
| | $q^2$ = the frequency of the **homozygous recessive** genotype in the population |

**Table 12.1. The two forms of the Hardy-Weinberg equation and their significance.**

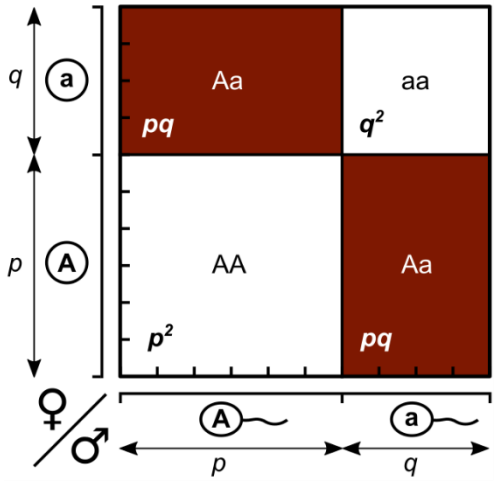



**Figure 12.1. The terms in the Hardy-Weinberg equations placed in a Punnett square.**

In Activity 2 of this lab you will run a simulation to see natural selection at work, and use the H-W equations on your simulated populations.

## Materials & Methods

### ACTIVITY 1: GALÁPAGOS FINCHES, PART II

In Activity 1 you will refine your groupings of 13 Galápagos finch species from last time, and finish watching the short film on the evolution of these finches.

**Materials**

- things from last time, plus:
- graph paper

**Part 3 – Examining Finch Beaks**

1. Get back into your teams from last time, and retrieve your poster and notes. Take a few minutes to review the groupings you made.

2. Watch the next part of the film (https://youtu.be/mcM23M-CCog), from 4:48 to 9:00. As you watch, answer the following:
    a. Describe the beak sizes of the medium ground finch population (species 12 on the cards).

    b. How did the population of medium ground finches on Daphne Major change as a result of environmental changes?

3. Make a prediction. On graph paper, create a bar graph on your own that shows the beak sizes of the medium ground finch population **before** and **after** the drought. Your graph should indicate the number of finches with each of four different beak sizes (see list below) before and after the drought. (Hint: You will create two bars for each category of beak size: one representing the populations before the drought, and one after the drought.) Include these categories of beak sizes:
    - medium ground finches with much smaller beaks
    - medium ground finches with smaller beaks
    - medium ground finches with larger beaks
    - medium ground finches with much larger beaks

4. Share your graph with your team, and provide feedback by asking at least two questions about your team members' graphs. Be ready to explain your graph.

5. Watch the next part of the film (https://youtu.be/mcM23M-CCog), from 9:00 to 11:12.

6. After you watch, answer the following:
    a. How did your graph compare to the graph in the film?

    b. If your graph was close to the one in the film, what part of your thinking was the same as that of the scientists in the film?

    c. Did anyone on your team have a graph that was similar to the one in the film?

    d. If no one on your team had a graph that was similar, what evidence did you not consider?

    e. Why did the drought have such an impact on the medium ground finch population?

7. Let's make a class prediction: if the drought had continued longer, what would you expect your beak graph to look like?

**Part 4 – Understanding Speciation**

1. Watch the last part of the film (https://youtu.be/mcM23M-CCog), from 11:12 to the end. As you watch, answer this question: How did one ancestral finch population give rise to 13 species, each with different characteristics?

2. With your team, create a graphic representation of the process that led to 13 different finch species. You may use the finch cards to make your graphic. Prepare your representation like a museum exhibit, so it will stand alone without your needing to explain it. However, you can include a written caption, as museum exhibits do.

3. Pause for a gallery walk. During the gallery walk, offer written feedback in the form of questions to at least three other teams' exhibits on index cards. Initial your feedback.

4. After the gallery walk, you may want to revise your presentation. What do you need to add? What do you need to take out? Make sure your presentation can stand up to peer review. This is your final chance to revise!

5. When you are done revising your presentation, turn it in to your instructor.

**ACTIVITY 2: NATURAL SELECTION SIMULATION**

In Activity 2 you will run a simulation on natural selection using pictures of birds. This simulation will also give you a chance to use the Hardy-Weinberg equations to estimate allele frequencies in your simulated population.

**Simulation:** you are bird-eating predators. In the bird population, there is a ***feather color trait*** determined by one gene with two alleles. ***Green feathers*** (G) are dominant to ***white feathers*** (g). Proceed through Part 1 in a sandy environment, then move to Part 2 in a forest environment.

**Materials**

- bird cards
- a small container
- a coin

**Part 1 – Sandy Environment (lighter coloring is more favorable)**

**Complete the tables as you go through each generation of the simulation. For Part 1, the birds' genotypes are on the back of the cards.**

***Generation 1:***

1. Populate your environment with 5 **green** birds and 5 **white** birds.

2. Fill out Gen 1 of the tables below. Flip the cards over for their genotypes.

3. As the predators, **randomly** remove 3 **green** birds and 2 **white** birds from the population.

4. The surviving birds have offspring. Place them in a small container.
   a. Randomly select two birds. They will have one offspring.
   b. Determine the ***genotype*** of the offspring by doing the cross.
      i. If the offspring has **only one** possible genotype, that is the genotype.
      ii. If the offspring have **two** possible genotypes, flip a coin **once**. Heads is the genotype with more dominant alleles, and tails is the genotype with fewer dominant alleles.
      iii. If the offspring have **three** possible genotypes, flip a coin **twice**. Two heads is the GG genotype, one head and one tail is the Gg genotype, and two tails is the gg genotype.
   c. Get a bird card with the correct genotype to represent the offspring. Place it on your bench.
   d. Put the two parents back in the container.
   e. Repeat steps a-d until you have **five** offspring. Place the survivors back on your bench.

***Generation 2:***

5. The birds on your bench now are Generation 2.

6. Fill out Gen 2 of the tables below. Flip the cards over for their genotypes.

7. As the predators, **randomly** remove 3 **green** birds and 2 **white** birds from the population. If you don't have that many green or white birds, remove as many as you have.

8. Repeat step 4 above to repopulate for Generation 3.

***Generations 3 & 4:***

9. Repeat steps 5-8.

10. Once you have Generation 4, fill out your Gen 4 numbers in the tables.

11. Return all of your **green** bird cards to the front.

| Generation | # of green birds | # of white birds | Total # of birds | % of green birds | % of white birds |
|---|---|---|---|---|---|
| 1 | | | | | |
| 2 | | | | | |
| 3 | | | | | |
| 4 | | | | | |

| Generation | $q^2$ | $q$ | $p$ | $p^2$ | $2pq$ | Does $p^2 + 2pq + q^2 = 1$? |
|---|---|---|---|---|---|---|
| 1 | | | | | | |
| 2 | | | | | | |
| 3 | | | | | | |
| 4 | | | | | | |

**Part 2 – Forest Environment (greener coloring is more favorable)**

**Complete the tables as you go through each generation of the simulation. This time, you do NOT know the birds' genotypes. You will need to calculate them based on their phenotypes.**

***Generation 1:***

1. Populate your environment with 5 **green** birds and 5 white birds.

2. Fill out Gen 1 of the first table below. Use those numbers to calculate the Hardy-Weinberg equation numbers.

3. As the predators, **randomly** remove 2 **green** birds and 3 white birds from the population.

4. The surviving birds have offspring. Place them in a small container.
   a. Randomly select two birds. They will have one offspring.
   b. Determine the ***phenotype*** of the offspring:
      i. If both parents are white birds, the offspring is a white bird.
      ii. If both parents are **green** birds, flip a coin **four** times. Four tails means the offspring is a white bird. Anything else, and the offspring is a **green** bird.
      iii. If one parent is **green** and the other is white, flip a coin **twice**. Two tails means the offspring is white; anything else means the offspring is **green**.
   c. Get a bird card with the correct phenotype to represent the offspring. Place it on your bench.
   d. Put the two parents back in the container.
   e. Repeat steps a-d until you have **five** offspring. Place the survivors back on your bench.

***Generation 2:***

5. The birds on your bench now are Generation 2.

6. Fill out Gen 2 of the tables below. Flip the cards over for their genotypes.

7. As the predators, **randomly** remove 2 **green** birds and 3 **white** birds from the population. If you don't have that many **green** or **white** birds, remove as many as you have.

8. Repeat step 4 above to repopulate for Generation 3.

***Generations 3 & 4:***

9. Repeat steps 5-8.

10. Once you have Generation 4, fill out your Gen 4 numbers in the tables.

11. Return all of the bird cards to the front.

| Generation | # of green birds | # of white birds | Total # of birds | % of green birds | % of white birds |
|---|---|---|---|---|---|
| 1 | | | | | |
| 2 | | | | | |
| 3 | | | | | |
| 4 | | | | | |

| Generation | $q^2$ | $q$ | $p$ | $p^2$ | $2pq$ | Does $p^2 + 2pq + q^2 = 1$? |
|---|---|---|---|---|---|---|
| 1 | | | | | | |
| 2 | | | | | | |
| 3 | | | | | | |
| 4 | | | | | | |

**One last question:**
**In the forest simulation, explain why the recessive allele does not disappear from the population.**

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## Hardy-Weinberg Practice Problems **Name & Lab Section:**

1. 1 in 1700 US Caucasian newborns have cystic fibrosis. The healthy allele (C) is dominant to the disease allele (c).

   *Phenotype and allele frequencies*

   a. What percent of the above population has cystic fibrosis? (recall: $q^2$ = homoz. recessive)

   b. When counting the phenotypes in a population, why is the homozygous recessive the most significant?

   c. Find $p$ and $q$ (the allele frequencies).

   *Genotype frequencies*

   a. What is the frequency of CC individuals ($p^2$)?

   b. What is the frequency of Cc individuals ($2pq$)?

   c. It has been found that a carrier (Cc) is better able to survive diseases with severe diarrhea. What would happen to the frequency of the c allele if there was an epidemic of cholera (another type of diarrhea producing disease)?

2. If 9% of an African population is born with a severe form of sickle-cell anemia (ss), what percentage of the population will be more resistant to malaria because they are heterozygous (Ss) for the sickle-cell gene?

3. Albinism is a rare genetically inherited trait that is only expressed in the phenotype of homozygous recessive individuals (aa). The most characteristic symptom is a lack of melanin pigment production. One in every 20,000 people has albinism.
   a. Find $p$ and $q$.

   b. Find the frequency of homozygous dominant, heterozygous, and homozygous recessive genotypes.

4. Within a population of butterflies, the color brown (B) is dominant over the color white (b). And, 40% of all butterflies are white.
   a. What percentage of butterflies in the population is heterozygous?

   b. What is the frequency of homozygous dominant butterflies?

# LAB 13 Ecology

## Objectives

At the end of the lab, you should be able to:

1. List and briefly describe some of the communities found in the duck pond.
2. Define and recognize examples of producers, consumers, herbivores, carnivores, omnivores and decomposers.
3. Describe the difference between trophic levels and food webs.

## Introduction

Ecology is the study of the interactions between living organisms and their environment. There is no vacuum in the biosphere. The behavior, appearance, population dynamics, biochemistry…just about any trait of a living organism that you can think of is affected by other organisms and the physical world in which they live. Such inclusiveness makes ecology a diverse and complex field, much too broad to be entirely addressed in a single lab session. Instead we will concentrate on two aspects of the interactions of organisms: the concept of community, and the way in which energy flows through a community.

There are several levels of organization considered in ecology. The first level is that of a **population**, a group of individuals of the same species. For instance, we can define ourselves, the group of *Homo sapiens* in this classroom right now, as a population. This lab room would be considered our habitat. A **habitat** is characterized by its physical features and the presence of certain other organisms, particularly plants. Here, our habitat consists of wood, plastics, and metals. Of course, we are not the exclusive occupants of this habitat. If we look closely we would find that we share this room with bacteria, nematode worms, and probably an insect or two. All of these populations in this particular habitat, including ourselves, make up a community.

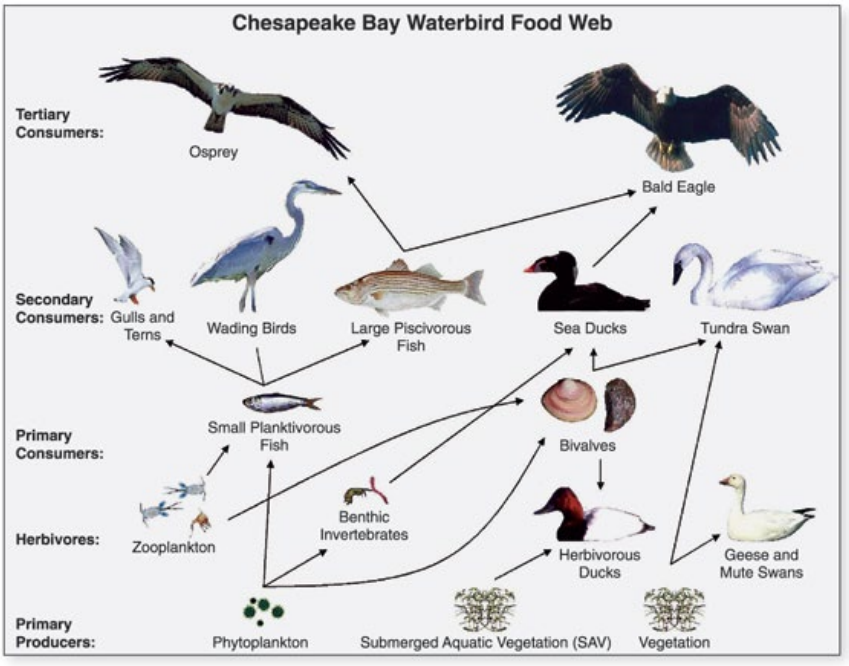


**Figure 13.1. A food web for the waterbirds of Chesapeake Bay.**

The **community** is of prime importance these days from a political aspect. In the past, preservation efforts have tended to focus on individual species; hence, the existence of the Endangered Species List. This effort is a bit misguided, however. Little is accomplished by preserving a single species from extinction if its natural habitat and the community of which it is a member is allowed to disappear. There is now a thrust in government towards the protection of entire communities or **ecosystems** (the combination of the community and its physical environment). To gain a better idea of what a community is, we will be taking a walk around the duck pond and discussing the communities.

## Materials & Methods

**ACTIVITY 1: THE DUCK POND**

**Materials**

- duck pond
- dissecting microscopes
- large culture dishes
- finger bowls
- tweezers

1. We will split into three groups for taking samples from the duck pond and identifying as many plants and animals as possible:

   **Group 1** will investigate the organisms living on the surface of the pond and around its immediate periphery (the mud zone).
   **Group 2** will look at the water column.
   **Group 3** will assess the sediments at the bottom of the pond.

   Samples will be brought back to the lab for macro- and micro-scopic examination.
   **We will pool our data at the end of the lab.**

2. List at least three physical characteristics of the duck pond that make it a unique habitat.

3. List at least three different pond organisms, and some adaptations they have which enable them to survive.

## ACTIVITY 2: THE FOOD WEB AND TROPHIC LEVELS

The primary source of energy for an ecosystem is the sun. All day long, the sun showers Earth with energy in the form of light (ultraviolet, visible, infrared, etc.). Only the **producers**, or **autotrophs**, are able to capture some of this light energy and convert it into chemical energy. They harness light energy to synthesize organic compounds, primarily carbohydrates, from inorganic compounds. The producers "feed" the rest of the ecosystem, which includes organisms known as consumers.

**Consumers** are also known as **heterotrophs** (literally "other feeder," an organism which must feed on another organism for nourishment). Consumers are divided into **trophic levels** based on their diet:

- **primary consumers** eat plants
- **secondary consumers** eat primary consumers or other secondary consumers
- **tertiary consumers** eat secondary consumers
- **decomposers** break down organic material into smaller molecules which are then recycled into the ecosystem; this group includes fungi, bacteria, and some protists

Together, all organisms in an ecosystem comprise a **food web**. See **Fig. 13.1** for an example.

**Below is a list of organisms in a forest community and their sources of energy in parentheses. Next to each write down its trophic level based on the definitions above.**

1. Human (raspberries, hickory nuts, deer, rabbits)
2. Black raspberry (sun)
3. Deer (plants)
4. Bear (raspberries, deer)
5. Coyote (deer, rabbits, raspberries)
6. Nematode (living hickory roots)
7. Bacteria I (raspberries)
8. Bacteria II (deer)
9. Bacteria III (dead trees, dead raspberries, dead humans, dead bears, dead deer)
10. Weasels (young rabbits)
11. Mosquito (blood of living humans, deer and bears)
12. Hickory tree (sun)
13. Cyanobacterium (sun)
14. Fungus I (raspberries)
15. Fungus II (dead hickory trees, dead raspberries)
16. Rabbit (raspberries)

**Now, pick at least ten of those organisms and put them together in a food web. Use Figure 13.1 as a model for your food web.**

**In ecosystems, there must be roughly ten times the mass of organisms in one trophic level to support the next "higher" trophic level. For example, 100 tons of plants (i.e. producers) will support 10 tons of rabbits and deer (i.e. primary consumers). What does this mean about ecosystems in general?**