BIOLOGY LABORATORY MANUAL
FOR NON-SCIENCE MAJORS

Safety Precautions

1. Do not:
    a. be late for class. You will miss important instructions.
    b. eat or drink in the laboratory
    c. ingest any reagents or chemicals used in the laboratory
    d. pour chemicals down the sink unless instructed otherwise
2. Dispose of laboratory materials as instructed. Take note of the location of:
    a. Regular trash can
    b. Biohazard bag
    c. Sharps container for disposal of glass slides and small sharp objects
3. Take note of the location of the:
    a. Fire extinguisher
    b. Eye wash and emergency shower
    c. Emergency power shut off button
    d. Location of security phone
    e. Safety glasses cabinet
4. Report all spills, unsafe conditions, or accidents to the instructor.
5. Wash your hands before leaving the laboratory
6. Keep work area neat and organized. Clean up when done with the laboratory exercise.
7. Push in your chair before you leave.

"I understand all of the safety procedures and information presented and I have been given an opportunity to ask questions concerning this safety information."

UNFOLDING THE MYSTERY OF LIFE: BIOLOGY LABORATORY MANUAL FOR NON-SCIENCE MAJORS

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Preface

This laboratory manual is intended for use in a biology laboratory course taken by non-science majors, pre-biology, and pre-allied health majors.

Laboratory exercises provide students with experience in basic laboratory skills, gathering and organizing data, measuring and calculating, hypothesis testing, analysis of data, writing, and laboratory safety. The skill sets are designed to promote the development of critical thought and analysis. Students work with living and preserved specimens, and laboratory reagents and equipment.

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Name _____ Date _____

Lab Time _____

Contents

Lab Exercise 1

THE METRIC SYSTEM OF MEASUREMENT

Purpose

To apply the metric system to the measurement of weight, distance, volume, and temperature

1.1 Universal Standards

The metric system was designed in France in the 18th century. Before then, it was common for units of length, area, and weight to vary from one country to another and even within the same country. Length could be measured in feet, miles, spans, cubits, hands, furlongs, palms, rods or chains. The metric system brought order to the confusing systems of weights and measures then being used in Europe. In 1875, most industrialized countries signed the Treaty of the Meter which formed the International Bureau of Weights and Measures. We now call this system the International System of Units. Although the US uses this system for scientific measurement, most people still use the more complicated system that involves inches, feet, miles, cups, pints, quarts, ounces, and pounds. The United States, Burma, and Liberia are the only countries that do not routinely use the metric system

Measuring Distance

The meter is the standard unit of linear measure. One meter (39.4 inches) is roughly equivalent to one yard (3 feet). 1 kilometer (km) is 0.6 miles. A finger is measured in cm (2.54 cm in 1 inch). A micrometer (micron) is not visible by unaided human eye. Most cells are in the micron range. To see an object much smaller, an electron microscope must be used.

Measuring mass

The **gram** is the basic unit for measuring mass. There are 0.454 kilograms in a pound.

Measuring temperature

The basic unit of temperature is degrees Celsius. Body temperature is 37 ^{o}C.

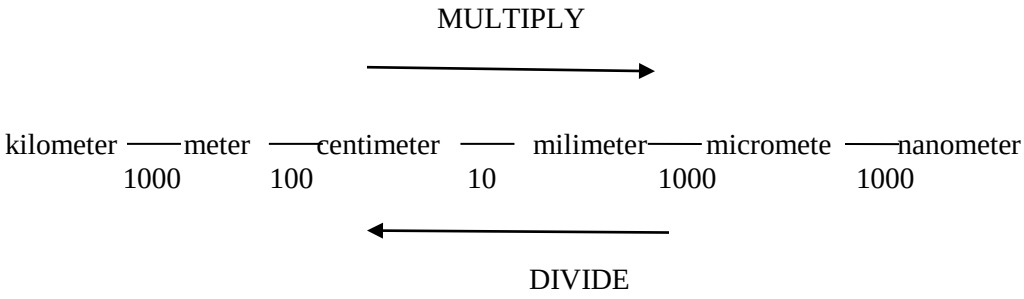
Measuring Volume

Volume is a three dimensional space occupied by a gas, liquid, or solid.

> *Liquid volume* is measured in **liters**. One liter = 1.06 quarts.
> The *volume of a solid* material is determined by multiplying length X depth X width to obtain a cubic number (c^3)f

1.2 Metric system prefixes and the metric ladder

prefix	fraction	decimal	scientific notation
nano (n) = 1 billionth	1/1,000,000,000	0.000000001	10^{-9}
micro (μ) = 1 millionth	1/1,000,000	0.000001	10^{-6}
milli (m) = 1 thousandth	1/1,000	0.001	10^{-3}
centi(c) = 1 hundredth	1/100	0.01	10^{-2}
kilo (k) = 1 thousand	1000	1000	10^{3}



Remember:

- The metric system is based on units of 10. Count zeros. Consult the metric ladder as to how many spaces to move the decimal point.
- Move the decimal left when converting from a smaller to larger value (DIVIDE)
- Move the decimal right when converting from a larger to smaller value (MULTIPLY)

Examples:

a. One meter is how many kilometers? The question involves going from a smaller value (meter) to a larger value (kilometer). Move the decimal to the left (divide)

1.0 meter → .001 kilometers = 1 X 10^{-3} kilometers

b. How many millimeters are in 5 meters? The question involves going from a larger value (meters) to a smaller value (millimeters). Move 3 decimal places to the right (multiply).

5.0 meters → 5000 millimeters = 5 X 10^{3} millimeters

c. How many liters are in 80 nanoliters? The question involves converting a small value (nanoliters) to a larger value (liters). Move the decimal to the left 9 places

80. nanoliters → .00000008 liters = 8 X 10^{-8} liters

1.3 Measuring Distance

Most objects encountered in biology range from under a millimeter to meters in length or diameter.

1. Obtain a human bone. Handle with care. Consult the articulated skeleton to find out the location of the bone.

What part of the body is the bone from? _____

The scientific name of the bone is the _____

2. Measure the length of the bone in centimeters (cm).

The length of the bone = _____ cm = _____ millimeters (mm) = _____ m

3. Use the meter stick to measure the length of your arm or leg, thumb.

The body part, the _____=_____centimeters (cm) =_____ millimeters (mm)

1.4 Measuring Mass

An electronic scale is used to measure the mass of an object. The instructor will demonstrate its use.

1. Obtain a small object such as dice. The mass of the object is _____ grams (g) = _____milligrams (mg)

2. Tare the weigh boat. Empty the salt into the weigh boat. The mass of the salt is _____ g = _____ mg

Question

- You should consume less than 3 grams of salt/day. You weighed an amount that many in the United States consume daily. How does this compare with the recommended amount?

4. Conversions

1 pound = 0.454 kilograms **1 inch = 2.54 centimeters**
1 kg = 2.2 pounds **1 meter = 39.4 inches**

1 quart = 0.946 liters
1 liter = 1.057 quarts

A person weighs 90,000 grams (g). What is the mass in kilogram (kg)? _____ What is the mass in pounds? _____

What is your height in inches _____ = _____ centimeters = _____ meters

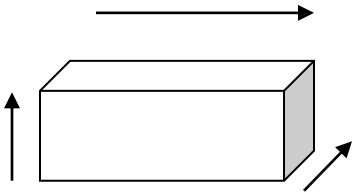
Could a normal person be 3 meters tall? ___ What is this height in feet/inches? ___

A bottle of water is 500 ml. Approximately how many cups of water does a person consuming this bottle drink? _____ cups

1.5 Measuring volume

The volume of an object is the amount of space it takes up.

Volume in cubic centimeters (cc or cm^3) = Length X Width X Depth



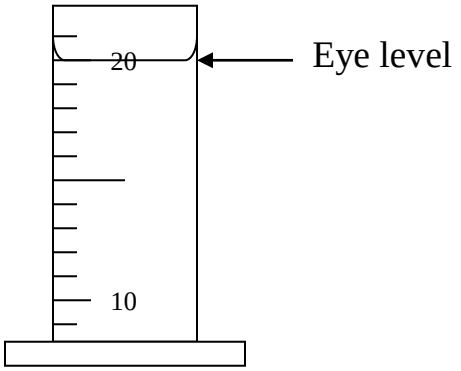
1. Measure the volume of a small item, such as dice, using a small metric ruler.

   The volume of the item is _____ cc (cubic centimeters or cm^3)

2. A <u>solid volume</u> of 1 cubic centimeter (cc) equals a <u>liquid volume</u> of 1 milliliter (ml). A person receives an injection of insulin (an important drug for diabetics).

   The volume of the injection is 1.25 cubic centimeters (cc) = ____ milliliters (ml)

3. Fill a 50 ml graduated cylinder with 20 ml H_20. Bend down so *meniscus* is at eye level. A meniscus is the curvature of the surface of the water. Measure at the lowest point of the meniscus.



3. Use the **displacement method** to determine the volume of the item that you measured in step 1. Examine the meniscus to insure accuracy.

   The volume of the item is _____ ml = _____ cc (cubic centimeters)

Question: What is displacement?

1.6 Measuring temperature

$$\text{Celsius (C)} = \frac{5\,(^0F - 32)}{9} \qquad \text{Fahrenheit (F)} = (^0C \times 1.8) + 32$$

1. Obtain a thermometer and record the temperature of the following environments:

   - Room temperature _____ º C
   - Ice water temperature _____ º C
   - Skin temperature _____ º C

2. Calculate

   - Water boils at 212 º F or _____ º C.
   - Body temperature is 37 º C or _____ º F
   - Water freezes at 32 º F or _____ ºC

TRY IT

1. How many decimal positions are moved to convert a meter to a kilometer? ___
2. What metric measure would you use for your height? _____
3. How many millimeters are in 9 centimeters? _____
4. How many meters are in 90 millimeters _____
5. How many grams are in 1 kilogram? _____
6. How many kilograms are in one microgram? _____
7. 1800 milliliters = _____liters
8. 1,200,000,000 nanograms (ng) = _____grams
9. 1,200,000,000 ng _____milligrams _____g _____kg
10. What is one metric measurement used for volume? _____
11. Body temperature is _____degrees Celsius
12. A block measures 3 X 4 X 2 cm.
    What is its volume in cubic centimeters(cc)?_____ in milliliters (ml)? _____

*Replace all items used in the laboratory and push in your chair when done
*Have your work checked by the instructor.
***Pick up the homework assignment due at the beginning of the next lab**

Laboratory Exercise 1
NOTES

Lab Exercise 2

MICROSCOPY

Purpose

- To gain experience in the care and use of the compound light microscope
- To prepare wet mounts of animal and plant tissues
- To observe living protozoa and vinegar eels

Light microscopes use light rays as an energy source. The **compound light microscope** is used to view objects in the micrometer (micron) range. These microscopes can magnify an object up to 1000 times.

2.1 Parts of the microscope

Become familiar with the location and function of the following parts.

1. **Arm** and **Base**

2. **Ocular lens** – magnifies by 10X

3. **Revolving nosepiece** – contains **3 objective lenses**

   scanning objective lens magnifies by 4X
   low power objective lens magnifies by 10X
   high power objective lens magnifies by 40X

4. **Stage** and **stage clips –** hold the slide for viewing

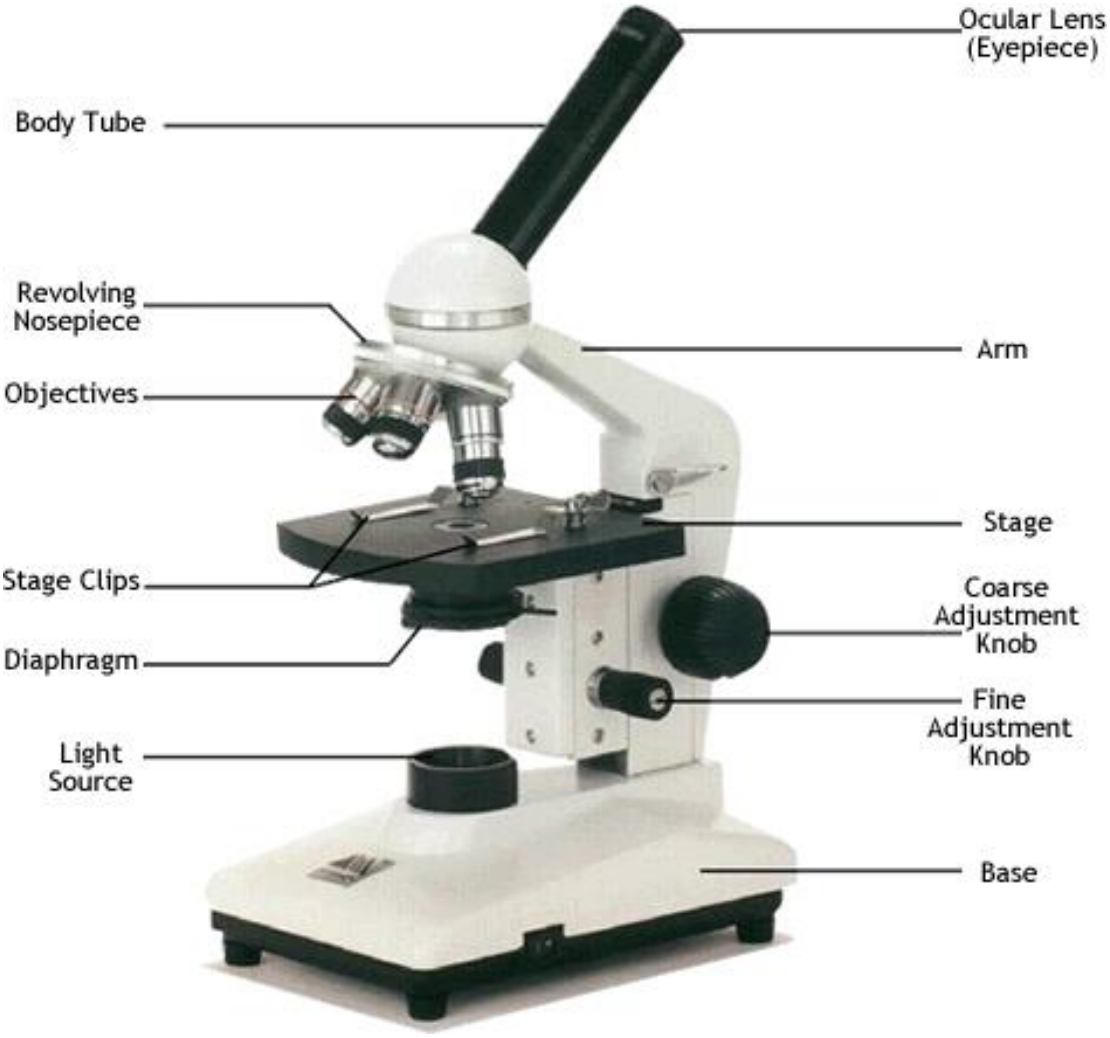
5. **Stage adjustment knobs** – located below the stage to control forward/reverse and side to side movement of the stage

6. **Coarse adjustment knob** – for focusing ONLY when using scanning objective lens

7. **Fine adjustment knob** –brings object into clearest focus

8. **Iris diaphragm** – controls the amount of illumination to improve contrast and resolution

The Compound Light Microscope



2.2 Care of the Compound Light Microscope

1. Only lens paper should be used to clean the lenses
2. Always carry the microscope by grasping the arm with one hand and supporting the base with the other
3. Do not force anything on the microscope
4. When finished working with the microscope:

    a. remove the slide
    b. rotate the nosepiece to scan
    c. turn the microscope off
    d. wrap the cord securely
    e. return to the cabinet

2.3 Focusing the Microscope - letter e slide

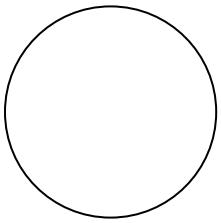
1. Rotate the nosepiece so that the scanning objective is over the light source.

2. Obtain a **letter e** slide from the instructor. Use the **stage clip** to secure the slide onto the mechanical stage. Use the **stage adjustment knobs** to bring the **letter e** under the scan objective and over the light source.

3. Use the **coarse adjustment knob** to bring the **letter e** into focus. Once the **letter e** is in focus, the image can be made sharper by using the **fine adjustment knob.**

Draw the "e" as you observe it with the microscope and describe the principle of **inversion**.

DRAWING

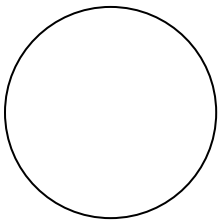


DESCRIPTION OF INVERSION

4. Move the slide so that the pointer is on the letter e. Rotate the **low power objective lens** into place and use the **fine adjustment knob** to bring the slide into focus.

5. Focus using the **high power objective lens** and the fine adjustment knob**.** *Do not use the coarse adjustment knob with the high power objective lens*. Record observations on high power.

Observations (draw):



6. Return the slide and use the revolving nose piece to return to the scan objective lens.

2.4 Total Magnification

Total magnification is calculated by multiplying the objective lens power X the ocular lens power. Complete the following table

Objective lens	Magnification	Total magnification
SCAN		
LOW POWER		
HIGH POWER		

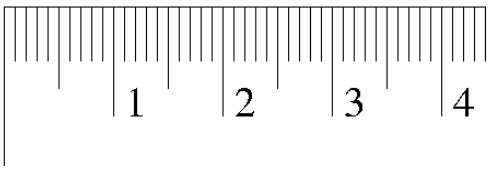
2.5 Diameter of Field: determining the size of a single cell

A. Measuring the diameter of field

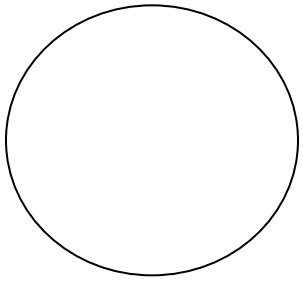
The field is the circle of light that you observe when you look into the microscope. The diameter of this field changes as you increase magnification. There is an inverse relationship between magnification and diameter of field.

1. Place a thin, clear, metric ruler on the stage. Hold it in place with the stage clip.

2. Use the **scan objective** to focus and observe the millimeter marks on the ruler.



The space from one hatch mark to the next is a millimeter. Numbers indicate centimeters.



Draw the area of the ruler that you see in your field of view.

Total Mag. _____

a. Diameter of field using scan objective lens _____ mm. **Include half spaces.**

= _____ micrometers (μm)

b. Diameter of field using low power objective lens = 1.8 mm _____ μm

c. Diameter of field using high power objective lens = 0.5 mm _____ μm

Question: Why is the diameter of the field of view considered to have an inverse relationship with magnification?

Answer:

B. Human epithelial cell

Question: What domain and kingdom do humans belong to?

Domain _____ Kingdom _____

1. Obtain a flat toothpick and obtain a sample of your cheek epithelial cells from the inside lining of the oral cavity.

2. Smear the cells on a clean glass slide and dispose of toothpick in biohazard trash.

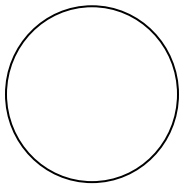
3. Obtain a small bottle of methylene blue dye. Make sure that the dropper does not actually touch the slide (*do not contaminate the dropper bottle with your cheek cells*). Let a small drop of methylene blue dye fall onto the slide.

4. Place a clear cover slip on top of the specimen.

5. Bring the epithelial cells into focus using the **scan** objective and the coarse adjustment knob. Zoom in on a few cells by using the **low power objective lens.**

6. Confirm with your instructor that you have viewed a cheek epithelial cell.

7. Switch the objective lens to **high power**. Sketch one cell



Total Mag. _____

8. Label the **cell membrane**, **nucleus**, and **cytoplasm** of the cell

9. What is the genetic material found in the nucleus of this eukaryotic cell? _____

C. Plant Cells - Onion

Question: What domain and kingdom do onion cells belong to?

Domain _____ Kingdom _____

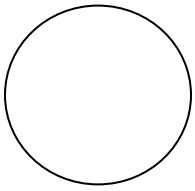
1. Remove the thin, transparent epidermis (skin) from an onion leaf. Alternately, you may view a prepared slide of onion root tip. Do not discard commercially prepared slides.

2. Place on a clean slide and add a drop of methylene blue. Do not contaminate the dropper (do not touch the onion skin with the dropper). Cover with a clear slip.

3. Observe with the scanning objective lens using the coarse adjustment knob first, then the fine adjustment knob.

4. Observe using the low power objective lens. Make sure you see the rectangular shaped onion cells. Confirm with the instructor, if necessary. Sketch the onion cells. Be sure to indicate the total magnification used in your drawing,

Sketch onion cells



Total Mag. ____

2.6 Observation of microorganisms (Kingdom: Protista)

Members of the kingdom Protista are single celled eukaryotic organisms. They have a nucleus and a complex internal cellular structure. The microorganisms you will view today are commonly found in pond water. The kingdom Protista is divided in phyla based in part on the different organelles that enable motility. Flagella are long whip like tails. Cilia, much shorter though more numerous than flagella, beat like little oars. A pseudopod is a cytoplasmic extension that acts as a "false foot". In addition to many Protists you may also see bacteria and multicellular animals called rotifers.

Question: What domain do members of the kingdom, Protista, belong to?

Domain _____ Kingdom: Protista

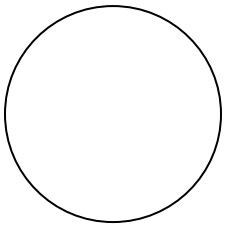
1. Prepare a wet mount of microorganisms by adding 1 drop of the culture to a clean slide. Cover with cover slip. Do NOT add methylene blue to these living organisms.

2. Observe under scan, then low power. Do you see any structures inside the organism? Try to observe using high power. Confirm the observation with your instructor.

3. Use the following website, the student identification key or a pond guidebook to identify 2 different organisms found in your pond water sample.

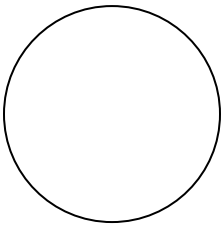
   http://www.microscopy-uk.org.uk/index.html?http://www.microscopy-uk.org.uk/pond/

4. Sketch the observed microorganisms →



Name of organism _____

Total Mag. _____



Name of organism _____

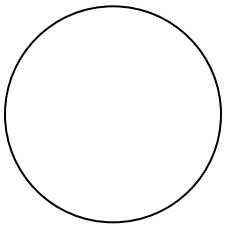
Total Mag. _____

D. Vinegar Eels

Vinegar Eels, *Tubatris aceti*, are small animals found in the nematode phylum.
Question: What domain and kingdom do Vinegar Eels belong to?

Domain _____ Kingdom _____

Sketch vinegar eels



Total Mag. _____

Click the scan objective lens into place, wrap the cord securely, and return the microscope to the cabinet

Laboratory Clean up

Dispose of slides in the sharps container.
The letter e slide is not discarded
Clean the lab bench area replacing items as directed by the instructor
Do not leave any items in the sink or on the lab bench
Wash your hands before leaving the laboratory

Laboratory Exercise 2
NOTES

Lab Exercise 3

THE SCIENTIFIC METHOD: SOLUBILITY OF GAS IN WATER

Purpose

- To engage in the scientific method
- To test a hypothesis concerning the effect of temperature on the solubility of carbon dioxide in water
- To gain practice in the analysis and graphing of data
- To formulate conclusions about changing levels of atmospheric CO_2 over time

3.1 Introduction

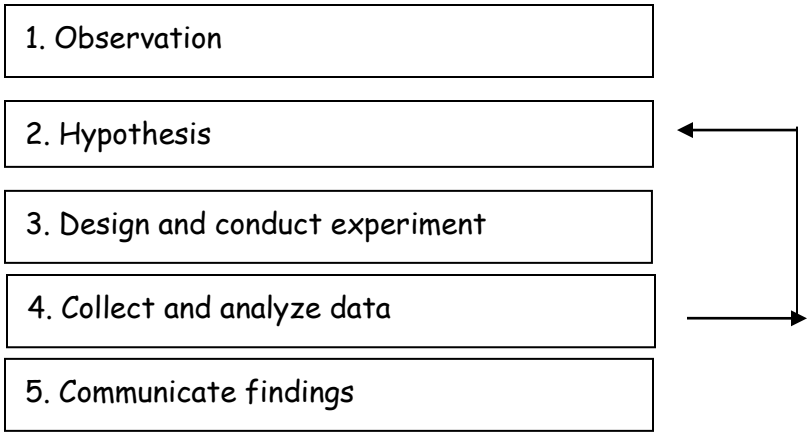
Science is a dynamic, methodical, approach to acquiring knowledge about the natural world. One element of the scientific method is a **hypothesis,** which provides a possible explanation for a particular observation. The hypothesis reflects the state of knowledge before the experiment has been performed. A hypothesis does not need to be correct, but it must be testable. A hypothesis has the potential to be falsified if data suggests another explanation for the observations. Some hypotheses are confirmed by numerous experiments conducted by many scientists and these, such as gravity, and a round earth, become **theories**.

A **theory** is a coherent set of hypotheses which have been confirmed through repeated experimental tests. Theories are not easily discarded because a great deal of evidence supports them. For example, if you fall off a building you do not have a possibility of floating because "gravity is just a theory". One theory discarded in the face of new evidence was the earth-centered view of the planetary orbits. The earth-centered view held that the sun and planets orbit the earth. This was falsified by the Copernican system which places the sun at the center of planetary orbits.

The scientific method attempts to eliminate two general types of experimental error. **Random error** occurs due to the imperfection of instruments that measure data. Another source of error in experimentation is **bias**. A preference, or bias, for a certain outcome. may lead us to unconsciously ignore data that does not fit our hypothesis.

It is important to add proper **controls** and standards to eliminate bias. All data must be treated equally. Data that does not "fit" may in fact indicate a new and important finding. Scientists present findings in journals and at meetings so that the scientific community can evaluate the merits of the work. It is important that other scientists be able to repeat the experiment and yield the same results.

The steps of the scientific method



3.2 Experimental design

The experiment is designed so that only one variable is tested at a time. The aspect that varies between groups is called the **experimental** (independent) **variable**. There can only be one experimental variable per experiment.

The group that receives the experimental treatment is the **experimental group**. For example, if the effect of aspirin on heart disease is being investigated, the experimental group is the group of individuals taking aspirin. There may be more than one experimental group within an experiment. For example, one group may take a pill containing no aspirin, another group consumes one aspirin a day, while a third group may take 2 aspirins a day.

The **control group** (control treatment) provides the baseline to which the experimental group will be compared. The control group is often a group that receives water or a placebo, or a group that receives the standard treatment. For example, in the aspirin study, the control group would receive a pill that looks the same as aspirin, but contains no active ingredient (a placebo).

Controlled variables are quantities that a scientist wants to remain constant between the experimental and control groups. The controlled variables insure that only one experimental variable is tested per experiment. There are usually a number of controlled variables in a single experiment. If you are examining the effect of aspirin on heart disease, controlled variables might include that all participants are adults, are the same sex, have no other health complications, similar levels of blood cholesterol, and non-smokers. Controlling these variables insures that only the experimental variable is investigated and the reliability of the data obtained is increased.

The **dependent variable** changes in response to experimental variable. Simply put, the dependent variable is what is **measured** to assess the experimental outcome.

Results must be counted or measured in some way so that discrete information can be obtained. In the aspirin example, the number of people who develop heart disease is counted as well as the age at which signs of heart disease are apparent.

The experiment should be conducted a number of times to insure that the results are real. In addition, within an experiment there should be an appropriate number of **replications**. An experiment designed to investigate the effects of fertilizer on plants would use many plants in each group.

3.3 Solubility of gas in water

Scientists hypothesize that approximately 250 million years ago, during the Permian period, the world's oceans became depleted of oxygen. A chain of events then led to the Permian mass extinction, a time during which most living species became extinct.

A. View the NOVA video and complete the worksheet
http://www.youtube.com/watch?v=y6ig6zKiNTc

1. How many years ago did the Permian mass extinction occur?

2. What % of species became extinct in the Permian mass extinction?

3. Mammal like reptiles and exotic ocean animals were present during the Permian period. What types of life were NOT on Earth 250 million years ago?

4. How many major extinctions have occurred on Earth?

5. What type of gas did the volcanoes in the Siberian Traps release?

6. Water can hold a type of gas critical to living organisms (fish and other aquatic animals require it to survive). What is this gas?

7. What type of gas do deadly bacteria in the lower layers of some lakes produce?

8. What does this gas smell like?

9. Develop a flowchart of the events that led to the Permian extinction
   a. Anaerobic bacteria thrive in the oceans and produce hydrogen sulfide
   b. Atmospheric carbon dioxide levels increase
   c. Atmosphere warms
   d. Dissolved oxygen levels in the oceans drop
   e. Hydrogen sulfide accumulates in the oceans and atmosphere
   f. Most aquatic life that depends on oxygen dies
   g. 95 percent of Earth's life is killed by hydrogen sulfide
   h. Oceans warm
   i. Volcanoes erupt

Concept question: Explain (briefly) how volcanic eruptions can change the atmospheric and ocean environments

B. Experiment

Atmospheric gases, such as oxygen and carbon dioxide, are soluble in water. How much of a particular gas dissolves in water depends on the temperature of the water and on the pressure of that gas above the water.

The gas in carbonated water (seltzer) is carbon dioxide. The high pressure inside the bottle causes more carbon dioxide to dissolve in the water than would dissolve at typical ground-level atmospheric pressures.

Materials

Carbonated water at room temperature
Carbonated water at 4 ^{o}C
3 glass beakers
40 ^{o}C water bath
Ice
Thermometer

State the hypothesis prior to beginning the experiment. The hypothesis indicates at which temperatures you believe CO_2 gas will be more or less soluble in water.

Hypothesis

Procedure

Read through the entire procedure before beginning the experiment

1. Obtain 3 - 250 ml beakers. Place one beaker on a bed of ice.
2. Pour 100 ml of ice-cold carbonated water into the beaker on ice.
3. Pour 100 ml room temperature carbonated water into the other 2 beakers.
4. Immediately place a beaker in the 40^{o}C water bath. Bring the beaker on ice and the beaker at room temperature with you so that you can observe them simultaneously.
5. Record observations in the data table.
6. After 5 minutes, determine the temperature of each water environment by placing the thermometer in the carbonated water.

Data Table

Temperature of carbonated water ^{o}C	Observations

Analysis and conclusion

Questions Refer to sections 3.1 and 3.2

1. The aspect that varies between groups in the experiment is called the **experimental** (independent) **variable**. Identify the experimental variable in the experiment.

2. **Controlled variables** are extraneous factors that are kept constant to minimize their effect on the outcome of the experiment. Identify 3 controlled variables

3. The **control group** provides the baseline to which the experimental groups will be compared. Identify the control treatment in the experiment.

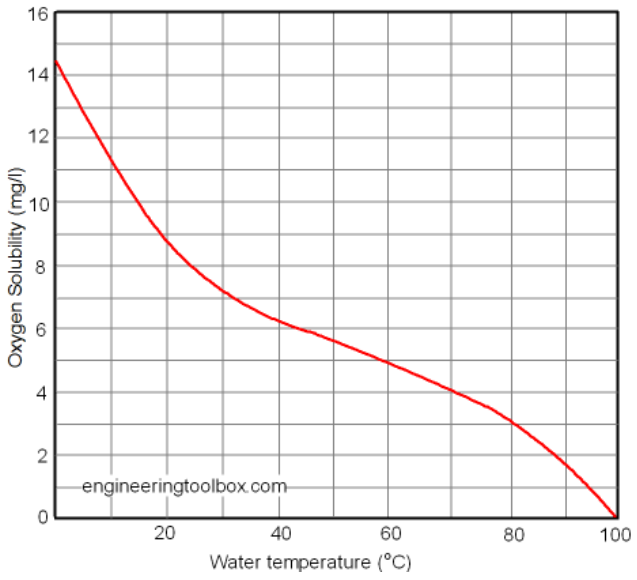
4. The **dependent variable** changes with respect to the experimental (independent) variable. The dependent variable is what is measured in the experiment. Identify the dependent variable.

3.4 Graphing Data

1. Examine the graph below:

   a. As temperature increases, the solubility of oxygen in water _____.

   b. Water in a particular stream varies from 8 - 13mg/liter. What temperature range does this reflect?

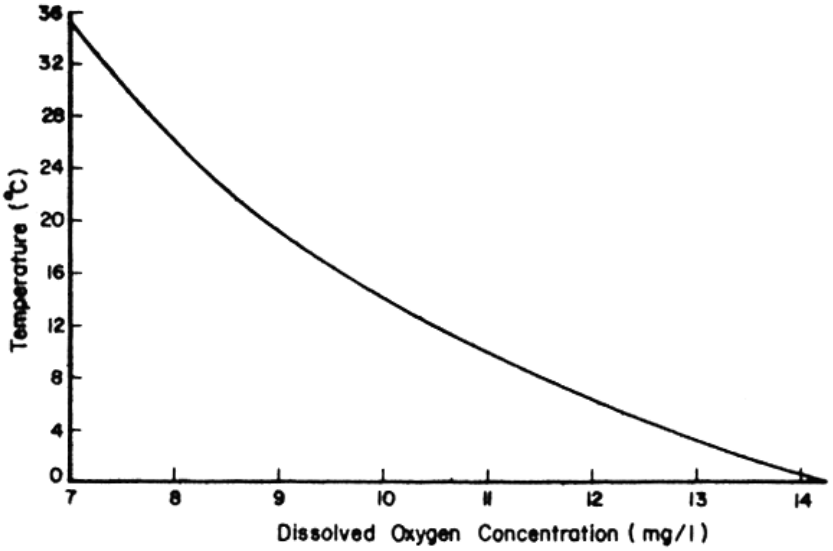


2. Fish require oxygen to live. As water passes through gills, dissolved oxygen (DO) is transferred to blood. Dissolved oxygen in water is affected by:

   Photosynthesis: During light hours, aquatic plants produce oxygen.
   Mixing: Waves and waterfalls aerate water and increase oxygen concentration.
   Decomposition: As organic material decays, bacteria consume oxygen.
   Salinity: As water becomes more salty, its ability to hold oxygen decreases.

   Examine the graph below:

   a. At which temperature is dissolved oxygen highest?___ ^{o}C



   b. A trout requires more DO when the water temperature is 24 ^{o}C (75 degrees F) as compared to when the water temperature is 4 ^{o}C · (to support an increase in metabolic activities). How do the DO levels compare at these two temperatures?

   c. A power plant discharges warm water into a river. Why would this have a detrimental effect on fish that live in the river? Be specific in your answer and relate it to this lab.

3. High levels of CO_2 have been implicated in global warming and as a causative factor in mass extinctions.

    a. Construct a line graph of the 1970–2018 data **by decade** from 1970 and ending with to 2010.

    b. State a conclusion as to the change in CO_2 levels over time.

Conclusion:

Carbon dioxide measurements from the Mauna Loa observatory in Hawaii
(values rounded to nearest tenth)

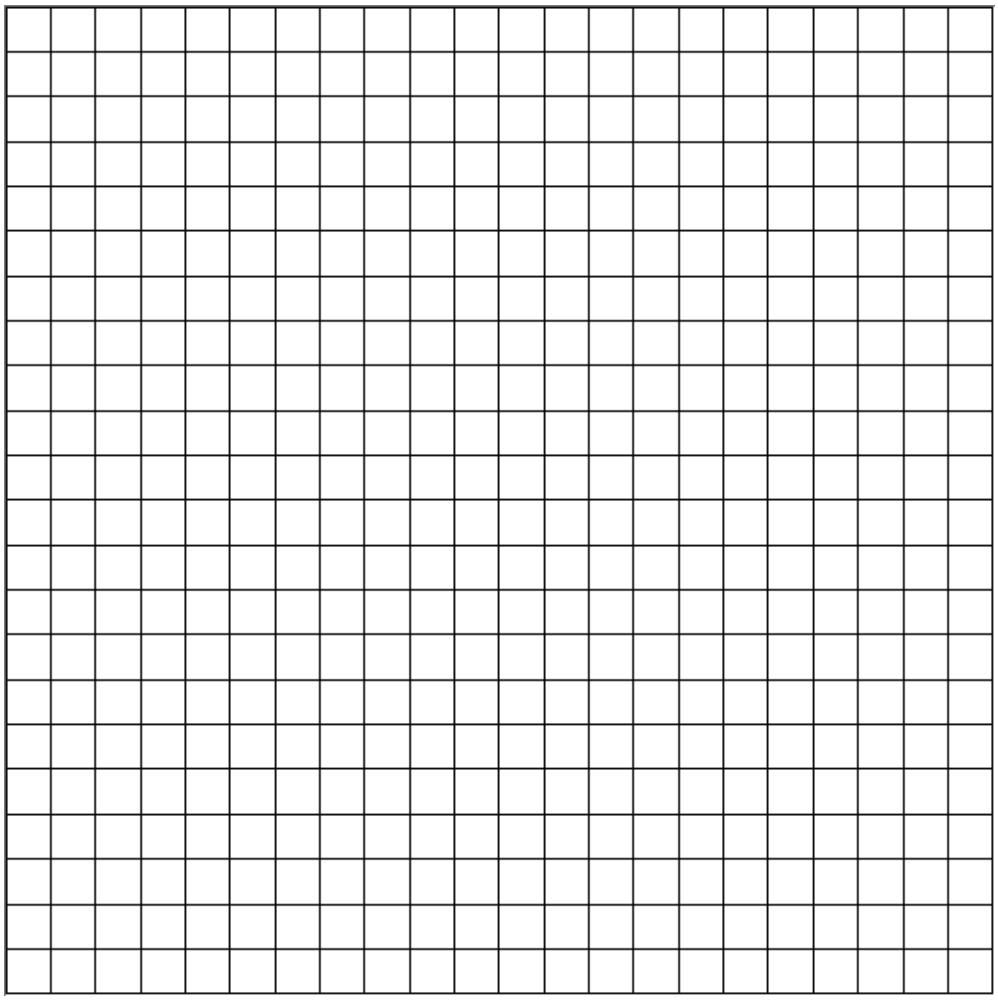
Year	CO_2 (ppm)	Year	CO_2 (ppm)	Year	CO_2 (ppm)	Year	CO_2 (ppm)	Year	CO_2 (ppm)
1970	316.9	1980	338.8	1990	354.4	2000	369.6	2010	389.9
1971	326.3	1981	340.1	1991	355.6	2001	371.1	2011	391.7
1972	327.5	1982	341.5	1992	356.5	2002	373.3	2012	393.9
1973	329.7	1983	343.1	1993	357.1	2003	375.8	2013	396.5
1974	330.2	1984	344.7	1994	358.3	2004	377.5	2014	398.7
1975	331.1	1985	346.12	1995	360.8	2005	379.8	2015	400.8
1976	332.0	1986	347.4	1996	362.6	2006	381.9	2016	404.2
1977	333.8	1987	349.19	1997	363.7	2007	383.8	2017	406.6
1978	335.4	1988	351.6	1998	366.7	2008	385.6	2018	408.5
1979	336.8	1989	353.1	1999	368.4	2009	387.4	2019	

https://www.esrl.noaa.gov/gmd/ccgg/trends/data.html

NAME: _____ SECTION: _____

Title _____

Y axis label: _____



X axis label: _____

Laboratory Exercise 3
NOTES

Lab Exercise 4

CELL MEMBRANE BIOLOGY

Purpose:

- To understand the relationship between homeostasis, diffusion, and osmosis
- To investigate passive mechanisms of transport across the plasma (cell) membrane
- To examine hypertonic, hypotonic, and isotonic solutions with respect to cellular homeostasis

4.1 Introduction

Homeostasis is defined as the maintenance of a stable internal environment. In order to maintain homeostasis, cells continually transport substances in and out across the cell (plasma) membrane.

The **cell membrane** is the cellular structure that regulates the transport of materials into and out of the cell. The lipid bilayer architecture of the cell membrane allows certain molecules to pass through while keeping others out. The cell membrane is **selectively permeable**.

Enter cell:	Ions	Nucleotides
	Sugars	Oxygen
	Amino acids	Water
	Hormones (some)	Vitamins
Leave cell:	Ions	
	Urea	
	Water	
	Carbon dioxide	
	Secreted proteins	

4.2 Diffusion

Diffusion is defined as the net movement of molecules or ions from a region of high concentration to a region of lower concentration. Diffusion continues until a state of equilibrium is reached, which means that the molecules are randomly distributed

throughout the system. Diffusion is considered a form of **passive transport** because no energy is required in the process. Diffusion can occur in a gas, a liquid, or a solid medium. Diffusion also occurs across the selectively permeable membranes of cells.

All molecules possess **kinetic energy** which provides the force for movement. Molecules are in constant motion and as they move, they collide with each other. The more molecules in an environment, the higher the concentration of molecules, the higher the frequency of molecular collisions and the faster the speed of diffusion.

How might these factors influence the rate of diffusion?

- Temperature
- Concentration gradient
- Medium molecules diffuse in (gas, liquid, solid)
- Molecular weight of the molecule
- Charge
- Solubility

A. Molecular Weight and Diffusion Rate

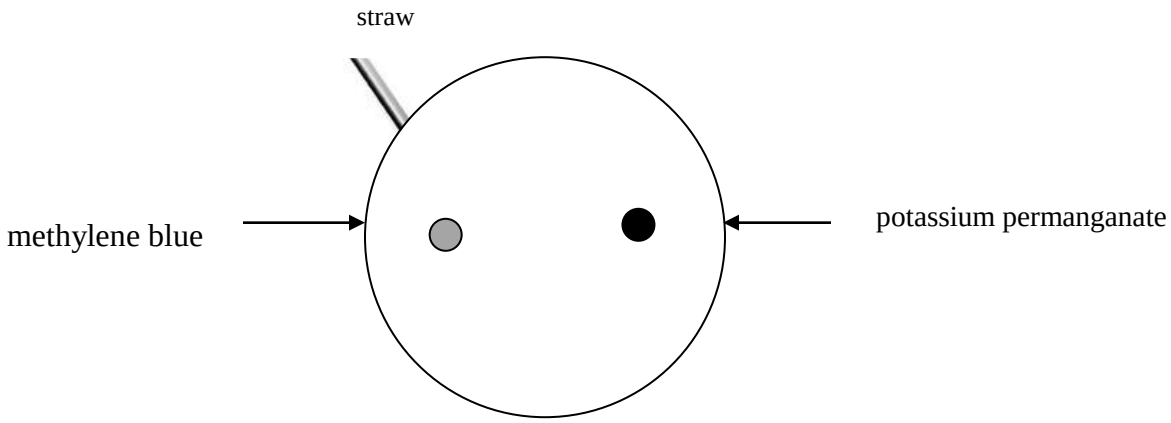
Molecular weight is an indication of the mass and size of a molecule. The purpose of this experiment is to determine the relationship between molecular weight and the rate of diffusion through a semisolid gel. You will investigate two dyes, methylene blue and potassium permanganate.

Molecule	**Molecular weight**	**Color**
Methylene blue	300 grams/mole	blue
Potassium permanganate	150 grams/mole	purple

Materials

Petri dish of agar semi-solid gel
Methylene blue solution
Potassium permanganate solution
Small straws
Small plastic metric ruler

Figure 4.1 Placing a drop of dye into a small well on an agar plate



Procedure

1. Obtain a Petri dish of agar
2. Take the plastic straw and gently stick down into the agar. Lift up withdrawing a small plastic plug of agar. Repeat.
3. Place a **single drop** of each dye into the agar well. (Figure 4.1).
4. After 20 minutes, place a small, clear metric ruler underneath the Petri dish to measure the distance (diameter) that the dye has moved. Enter the data in Table

Table 4.1

	Molecular weight (grams/mole)	Diameter after 20 minutes (millimeters)	Diameter after 40 minutes (millimeters)
Methylene blue			
Potassium permanganate			

Describe the relationship between molecular weight and speed of diffusion

B. Diffusion across a selectively permeable membrane

Cells acquire the molecules and ions they need from their surrounding extracellular fluid. In living cells, the ability of a molecule to cross the cell membrane is influenced by its size, charge, lipid solubility, and other characteristics. Small molecules such as water, oxygen, amino acids, and ions easy cross the membrane by passive transport processes that do not require energy (diffusion and osmosis). Other molecules do not easily fit through the lipid bilayer and the cell must expend energy to bring them across.

You will investigate two molecules, starch and iodine, for their ability to cross a selectively permeable membrane. A **colorimetric test** is employed to assess the movement of these molecules. **Dialysis tubing** is a transparent material with microscopic pores that allow only small molecules to pass. It provides a model of the cell membrane and has many uses in industry and medicine.

Materials

Beaker
Dialysis tubing
Starch solution
Iodine (IKI)

Procedure

1. Obtain a piece of dialysis tubing that has been pre-cut by the instructor. Thoroughly wet the tubing and open the ends. Tie a knot in one end.

2. Add approximately 2ml (~2cm) starch solution to the dialysis bag. Tie a knot at the top of the tubing. Rinse the bag briefly with tap water to remove any traces of starch.

3. Fill the beaker approximately ½ full with tap water.

4. Add iodine (IKI) to the beaker of water until a deep yellow color is obtained.

5. Submerge the dialysis bag in the water and incubate at room temperature until a color change is observed (~15 minutes).

Questions:

1. Did starch diffuse across the selectively permeable membrane? How do you know?
2. Did iodine diffuse across the selectively permeable membrane? How do you know?

3. Which is the smaller molecule, iodine, or starch?
4. Is diffusion a passive, or an active transport process (choose one)?

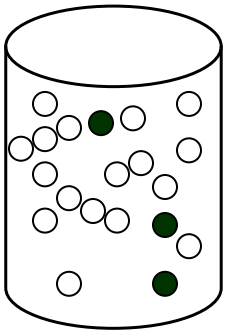
4.3 Osmosis

Osmosis is a special case of diffusion in which water molecules pass through a selectively permeable membrane, but larger molecules do not. Osmosis proceeds from a region of high water concentration, across a semi-permeable membrane, to a region of lower water concentration until equilibrium is reached.

A **solute** is a solid substance, such as salt or sugar that is dissolved in a **solvent**. Water is usually the solvent in living systems.

solute = salt

solvent = water



A typical animal cell contains a salt concentration of 0.9%. A solution of equal solute concentration is referred to as **isotonic**. A cell placed in an isotonic environment will experience movement of water inside and outside the cell, but there will be no change in the biology of the cell.

A **hypertonic** solution contains a high solute concentration with respect to cells. For example, a solution containing 10% salt is hypertonic. When a cell is placed in a hypertonic environment, there is a net movement of water to the outside of the cell (from the higher water environment inside the cell). The cell shrinks in response.

A solution of low solute concentration is referred to as **hypotonic.** A solution containing 0.5% salt is hypotonic with respect to the cell. When a cell is placed in a hypotonic environment, there is a net movement of water into the cell. The cell swells in response.

Hypertonic, hypotonic, and isotonic are relative terms and are used only when comparing two different solutions.

Activity: Osmosis in eggplant and potato cells

Materials

Thin slice of eggplant
Two slices of potato pre-cut
NaCl (table salt)
2 test tubes
10% NaCl solution
1 piece of weigh paper or plastic

Procedure

1. Obtain a thin slice of eggplant. Sprinkle the eggplant with salt. Place on a piece of plastic or weigh paper. Incubate at room temperature for approximately 10 minutes.

2. Obtain two pieces of peeled potato, approximately 2 cm X 0.25cm. Label two test tubes with a wax marker at the 5 cm point

Tube 1: Add distilled water to the 5 cm mark
Tube 2: Add 10% sodium chloride to the 5 cm mark

Add a potato piece to each tube and incubate at room temperature for ~15 minutes Pour off the solution and feel each potato piece. Rinse test-tubes thoroughly with water to remove traces of salt and potato starch

Observations

1. Describe how the eggplant slice looks.

2. In terms of osmosis, why does it appear this way?

3. Were eggplant cells exposed to a hypertonic or a hypotonic environment (choose 1)

4. What is the experimental variable in the potato experiment?

5. Identify 2 controlled variables in the potato experiment

6. Which potato piece is stiff? Explain why with respect to osmosis.

7. What happens to human red blood cells when placed in saline, an isotonic solution?

8. A slug is a garden pest. Why, in terms of osmosis, do some people salt slugs?

Laboratory Exercise 4
NOTES

Lab Exercise 5

BIOMOLECULES

Purpose

- To investigate carbohydrates, lipids, and proteins
- To perform colorimetric tests to identify specific biomolecules
- To analyze data and identify features of the scientific method
- To identify the components of an unknown sample

5.1 Introduction

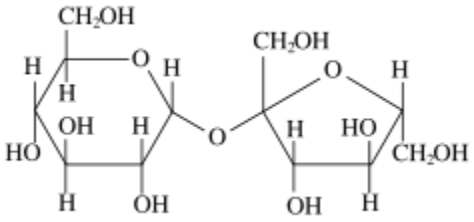
Humans are omnivores in that they consume a variety of food types from several different ecosystem levels. The biomolecules we consume - carbohydrates, lipids, and proteins - provide us with energy and building blocks for our bodies to function. Vitamins and minerals are important dietary components required in small amounts. **Water** is not a nutrient or a source of calories but is critical to life.

5.2 Carbohydrates

Carbohydrates include sugars and starches and are composed of monosaccharide building blocks. **Glucose** is a simple sugar, a monosaccharide. **Fructose** is a monosaccharide found in honey, tree fruits, berries, and many vegetables. It is the sweetest naturally occurring sugar.

Two simple sugars joined together form a **disaccharide**. An example of a disaccharide is sucrose (table sugar) which is formed by glucose + fructose. **Lactose**, also known as milk sugar, is a disaccharide composed of a galactose and a glucose molecule. The enzyme, lactase is required to break down lactose into its two monosaccharide sugars. Lactase is normally secreted by intestinal cells. In many people, the production of lactase diminishes with age and they become lactose intolerant.

Sucrose



Glucose + Fructose

$C_6H_{12}O_6$ $C_6H_{12}O_6$

Starches are polysaccharides which contain many linked sugar molecules.

Benedicts Test for Sugar

Reducing sugars (most 6 carbon sugars) react with a copper containing reagent called Benedict's. Benedict's reagent is blue, but when heated in the presence of a reducing sugar, changes color. Green, yellow (+sugar), orange (++ sugar), or red (+++ sugar).

Materials
Benedict's reagent
Wax pencil
Test-tubes and rack
70^{o} C water bath

Hypothesize about which substance(s) will produce a color change in the Benedict's test.

Procedure

1. Label test-tubes 1 – 6.
2. Draw a line 1 cm from the bottom of each test tube and another line 2 cm from the bottom of each tube.
3. Fill in Table 1 to specify which tube will receive which test substance.
4. Add the appropriate test solution to the level of the 1 cm line.
5. Add Benedict's reagent to the 2 cm line of each tube. Mix gently. Record initial color in Table 5.1. This is the color after Benedict's reagent has been added but before heat.
6. Boil, or heat tubes in the hot water bath for 5 minutes. Record data in Table 5.1

Table 5.1 Benedict's test for sugar

Tube	Contents	Color before heating	Color after heating	Check if sugar resent	Name of sugar (section 5.2)
1	Distilled water				
2	Glucose				
3	Egg white				
4	Milk				
5	Orange Juice				
6	Unknown				

Analysis:

1. The aspect that varies between test-tubes in the experiment is the **experimental** (**independent**) **variable**. Identify the experimental variable in the experiment.

2. The **negative control group** provides the baseline to which the experimental group is compared. The control group is often a "no treatment" condition. Identify the negative control in the experiment. (Choose from column marked Contents in Table 5.1)

3. The **positive control treatment** provides a known effect or response in the experiment. Identify the positive control in the experiment. (Choose from column marked Contents in Table 5.1)

4. **Controlled variables** are extraneous factors that are kept constant to minimize their effect on the outcome of the experiment. The volume of Benedict's reagent added to each tube is a controlled variable. Identify 3 additional controlled variables.

5. The **dependent variable** changes in response to the experimental variable and is used to assess the experimental outcome. Identify the dependent variable.

6. Do your results support your hypothesis? Explain.

Iodine test for starch

Lugol's reagent (IKI) is an amber color which, in the presence of starch, turns dark blue.

Materials
Wax pencil
Test-tubes and rack
Potassium iodide (IKI)

Hypothesize about which substance(s) will produce a color change with the iodine test.

Procedure

1. Label test tubes 1 – 6.
2. Draw a line 1 cm from the bottom of each test tube (use a wax marker).
3. Add the appropriate test solution to the level of the 1 cm line. If using potato or onion piece, or paper, a test tube is not required.
4. Add 3 drops of IKI (iodine). If using potato or onion piece, or paper, the IKI can be dropped directly onto the substance.
5. Record data in Table 5.2

Table 5.2 Iodine test for starch

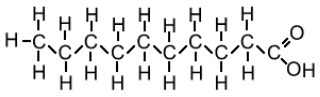
Tube	Contents	Final Color	Check if starch present
1	Distilled water		
2	Glucose		
3	Potato Juice		
4	Milk		
5	Egg White		
6	Unknown		
7	Paper Towel*		
8	Pasta *		

***** do not require a test tube

Analysis: Did your results support your hypothesis? Explain.

5.3 Lipids

Lipids include oils, fats, and waxes. Lipids provide stored energy, are essential components of cell membranes and are the building blocks of certain hormones. Dietary lipids are insoluble in water and cannot be utilized by the body unless broken down into smaller molecules. The body digests lipids by breaking them down into tiny components by **emulsification** with bile salts and by enzymes called lipases. The emulsification of lipids occurs in the small intestine.



Saturated Fatty Acid

Emulsification of lipids

Materials
test tubes and rack
vegetable oil
water

1. Draw a line at the 1 cm mark and the 2 cm mark on 2 test tubes.
2. Add vegetable oil to the 1 cm mark in both test tubes.
3. Add an additional cm of water to each test tube.
4. Add 10 drops of detergent to one of the tubes
5. Cover the tubes with parafilm and shake vigorously about 30 times (instructor will demonstrate).

Analysis:

1. Describe the results (do not include the presence of foam) with respect to lipid emulsification.

2. Identify the **experimental variable** in the lipid experiment.

3. Which tube represents the **control group in** the lipid experiment?

4. List 3 **controlled variables** in this experiment

5. What is the **dependent variable** in the experiment?

5.4 Proteins

Proteins are complex molecules often with a 3 dimensional structure. These biomolecules are composed of amino acids building blocks joined by peptide bonds. Proteins form structural components of cells, insulin, hemoglobin, muscle, enzymes, eye, hair color, and thousands of other essential body components. Proteins can be identified using the Biuret test. In the presence of peptide bonds, Biuret reagent turns from blue to purple.

Materials

Test tubes
Test tube racks
Wax pencil
Biuret reagent

Hypothesize about which substance(s) will produce a color change with the Biuret test

Procedure

1. Label test-tubes 1 – 6.
2. Draw a line 1 cm from the bottom of each test tube and another line 2 cm from the bottom of each tube.
3. Fill in Table 3 to specify which tube will receive which test solution.
4. Add the appropriate test solution to the level of the 1 cm line.
5. Add Biuret reagent to the 2 cm line of each tube. Mix gently.
6. Record data in Table 5.4.

Table 5.4 Biuret test for protein

Tube	Contents	Final Color	Check if protein present
1	Distilled water		
2	Potato Juice		
3	Glucose		
4	Milk		
5	Egg White		
6	Unknown		

What is the composition of your unknown sample? _____

5.5 Summary

1. What are the three main classes of biomolecules tested in this lab? _____
2. Biruet reagent tests for the presence of _____ and if positive turns a ____color.
3. Benedict's reagent tests for the presence of ____ and if positive turns a ____color.
4. Iodine tests for the presences of _____ and if positive turns a _____ color.
5. One carbohydrate category is starch. What is the other?_____
6. Which test requires high heat after the addition of colorimetric reagent? _____
7. What type of biomolecule is egg white? _____
8. List 3 substances that contain carbohydrates tested in the laboratory _____
9. What are the building blocks of proteins? _____
10. Saturated fats and oils are in the biomolecules category of _____
11. What color change, if any, would glucose produce in the iodine test? _____
12. What test solution was the negative control in all experiments in today's lab? ____
13. Identify two biomolecules contained in milk_____/_____
14. What is paper made from and why would paper test positive in the iodine test?

15. Diabetes is a chronic disorder that results in an increased level of sugar (glucose) in the bloodstream. It is caused by inadequate insulin, a hormone produced by the pancreas that allows cells to use and store glucose. One symptom of diabetes is excess glucose in the urine (glycosuria). Which colorimetric test could be used to assay a person's urine and what color change could indicate mild diabetes?

Laboratory Exercise 5
NOTES

Lab Exercise 6

ENZYME ACTION

Purpose

- To examine the function and properties of enzymes in living cells
- To distinguish between substrate, enzyme, and product in a reaction
- To collect and analyze data on tissue-specific catalase activity
- To collect and analyze data on the effect of temperature on catalase action

6.1 Introduction

All living organisms depend on **enzymes** to catalyze (speed up) chemical reactions. Without enzymes, reactions would not occur fast enough to support life. There are many types of enzymes, active in all types of cells. Each reaction uses a specific enzyme.

All enzymes are proteins. The amino acid sequence determines the three dimensional shape of the enzyme. This shape is critical because it allows the enzyme to interact with its substrate molecule, the molecule that the enzyme works on. Enzyme action changes the substrate into a product(s). The enzyme itself remains unchanged in the reaction, it can be reused. Each enzyme works best at a specific temperature and pH. Most enzymes in the human body exhibit maximal activity at 37º C and a pH around 7. Extremes in temperature and pH can change the shape of the enzyme rendering it inactive.

Substrate + Enzyme → Product(s) + Enzyme

<u>Enzyme Properties</u>

1. All enzymes are proteins.
2. Enzymes speed up, or catalyze, chemical reactions, often by many thousand-fold.
3. Enzymes are required in small amounts.
4. Many enzymes require assistants, called cofactors. Cofactors may be metal ions such as iron and zinc, or vitamins.
5. Enzymes can be reused. They are unchanged by the reaction.
6. Enzymes are specific. The substrate is the substance that the enzyme works on. Each substrate is digested by a specific enzyme.
7. Enzymes are affected by pH (acidity) and temperature. The rate of the reaction is dependent on these two variables.
8. Enzymes exposed to extremes in temperature or pH become *denatured* (lose their three dimensional shape and their activity).

6.2 Catalase

Cells produce hydrogen peroxide (H_2O_2) as a toxic by-product of normal cellular reactions. The enzyme catalase quickly breaks down hydrogen peroxide into water and oxygen. In other words, catalase protects cells from the toxic effects of hydrogen peroxide. All aerobic cells produce catalase. One molecule of catalase enzyme may work on 40 million molecules of hydrogen peroxide per second!

Substrate		Enzyme		Products	
		catalase			
Hydrogen peroxide	----------→		**water +**	**oxygen**	(+ catalase)
$2\ H_2O_2$			$2H_2O$	O_2	

If you have used hydrogen peroxide to clean a cut, you have seen catalase in action. When hydrogen peroxide comes in contact with the cut, it reacts with the catalase enzyme in the damaged cells to produce oxygen foam. When the catalase reaction is conducted in a test-tube, the oxygen gas bubbles. *The height of the foam is an indication of the amount of catalase activity present.*

A. Activity: Investigation of catalase activity in plant and animal tissues

Materials

5 test-tubes
wax pencil
millimeter ruler
4 blended tissue extracts (may include apple, potato, onion, Baker's yeast, beef steak, or/and beef liver)
distilled water
hydrogen peroxide (H_2O_2) solution (3%)

Hypothesis
State a hypothesis concerning the relative abilities of the various tissue types to catalyze the conversion of hydrogen peroxide to water and oxygen.

Procedure

1. Label tubes 1 – 5. Use a wax pencil to draw a line 1 cm from the bottom of each tube. Draw an additional line at the 2 cm mark

2. Add the appropriate extract to each tube to the level of the 1 cm line.

3. Add H_2O_2 to the 2 cm line in each tube. Do not contaminate the dropper. If extract comes into contact with the dropper, discard the dropper.

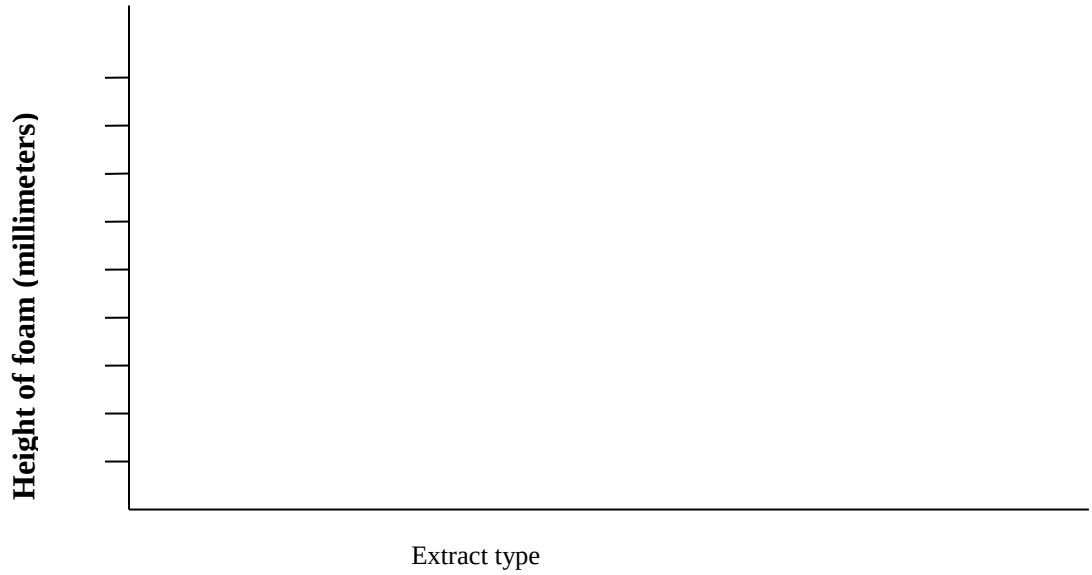
4. Measure the height of the foam (in millimeters) and record the data.

	Extract Type	Foam Height (mm)
Activity A	Distilled water	

5. Wash the test-tubes. Rinse well with water and drain.

Results Prepare a bar graph of catalase activity. Include labels.

Title of Bar Graph _____



Analysis

1. Did the results of the experiment support the hypothesis? Explain.

2. Write the reaction. Use molecular formulas. Identify substrate, enzyme and products.

3. What type of gas is released in the bubbles? _____

4. What happens to catalase itself in the reaction – is it used up? destroyed? unchanged?

5. The experimental variable (independent variable) is the aspect that varies between the experimental groups. What is the experimental variable in this experiment? _____

6. Controlled variables are those conditions that are kept constant between experimental groups. The controlled variables insure that only one experimental variable is tested per experiment. What are 3 controlled variables in this experiment?

7. The dependent variable changes in response to the experimental variable. The dependent variable is what is physically measured to assess the experimental outcome. What is the dependent variable in this experiment? _____.

B. Activity: The effect of temperature on the activity of catalase

Materials

3 clean test-tubes	hydrogen peroxide solution
wax pencil	extract of liver or yeast
millimeter ruler	crushed ice
water bath at 70^{o}C	thermometer

.

Hypothesis

State a hypothesis concerning the relative action of catalase at various temperatures. Refer to section 6.1

Procedure

1. Label tubes 1- 3. Use a wax pencil to draw a line 1cm from the bottom of each tube. Draw another line at the 2 cm mark.

2. Add extract to the 1 cm line. Note that extract type is a *controlled variable* in this experiment. Do not add hydrogen peroxide until AFTER step 3.

3. Incubate the tubes at the following temperatures for 5 minutes
   Tube 1 on ice (record the temperature of the ice)
   Tube 2 at room temperature (record the temperature)
   Tube 3 at 70 ^{o}C

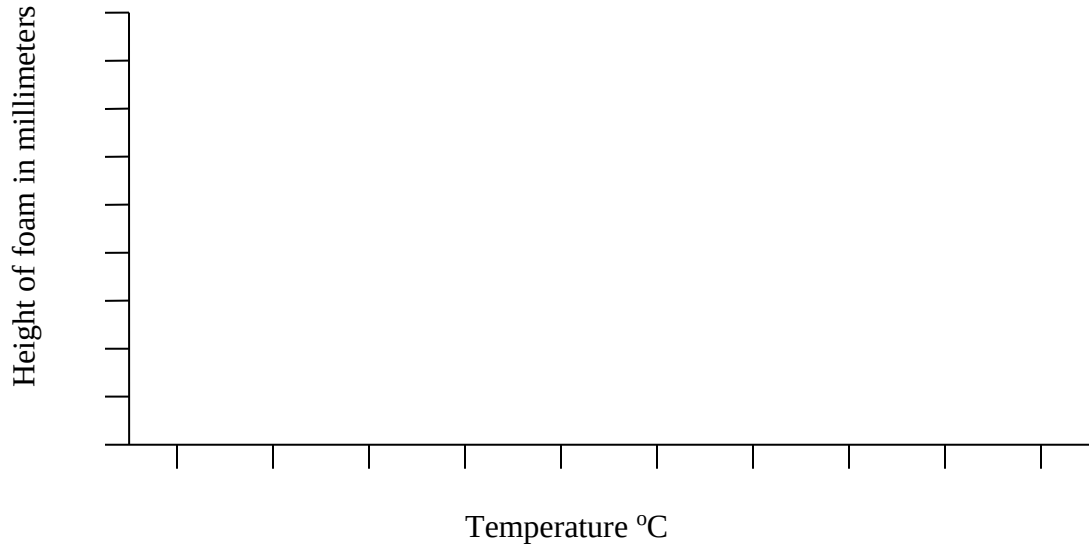
4. After 5 minutes, return the tubes to the lab bench. Add H_2O_2 (hydrogen peroxide) to the 2 cm line. Use ice-cold H_2O_2 for the tube on ice.

5. Measure the height of the foam on each tube. Record.

	Temperature ^{o}C	Height of foam (mm)
Activity B		

6. Wash the test-tubes with water and drain to remove the excess water

Results Prepare a line graph of catalase activity as influenced by temperature.
Title of Line Graph _____



Analysis

1. Explain the results shown in the line graph. Did the results support the hypothesis?

2. Identify the experimental variable in the experiment_____

3. List 2 controlled variables in the experiment _____

4. What is the dependent variable in the experiment? _____

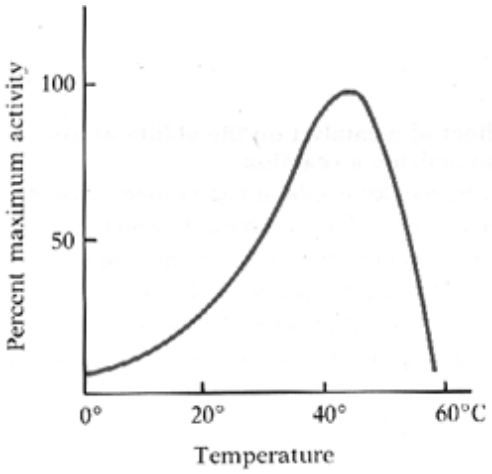
6.3 Analyzing Graphs

1. Figure A

a. What is the preferred temperature for this enzyme? _____ degrees C.

b. What happens to enzyme activity when the temperature exceeds 50ºC? What is the technical term used to describe what happens to enzymes when heated to high temperature? _____.

FIGURE A**.**

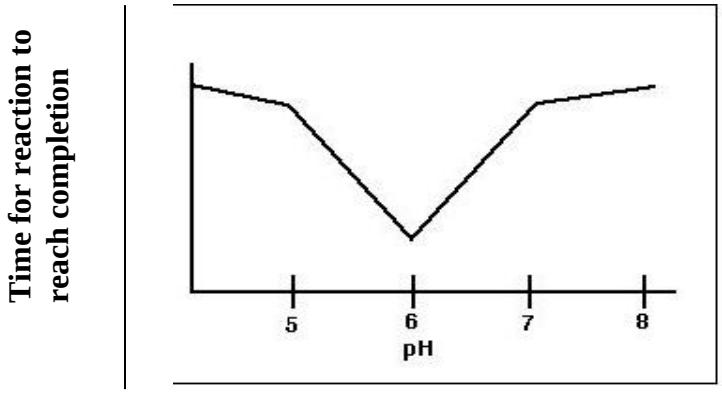


2. Figure B. Amylase is an enzyme in saliva that digests starch. The graph shows the relationship of pH to amylase activity.

   a. What is the ideal pH for the amylase enzyme? _____

   b. What the technical term used to describe what happens to the enzyme at very low (acidic) and very high (basic) pH? _____

FIGURE B



3. Figure C. Relationship of substrate concentration to enzyme reaction rate.

At what substrate concentration does the enzyme reaction reach **half** maximal rate? _____ mM (millimolar)

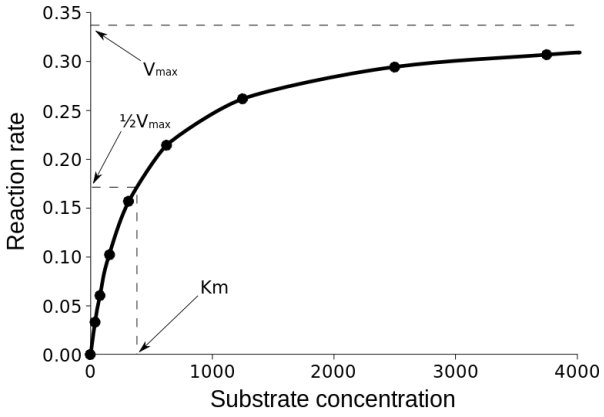


FIGURE C

4. An enzyme reaction is saturated with substrate. How can more product be produced?

   a. add more substrate
   b. add more time
   c. add more enzyme
   d. add more product

Laboratory Exercise 6
NOTES

Lab Exercise 7

PHOTOSYNTHESIS

Purpose

- To recognize how the first photosynthetic organisms changed the atmosphere of the earth.
- To describe the 2 phases of photosynthesis.
- To test a hypothesis concerning the effect of different light conditions on the rate of the light dependent reaction of photosynthesis.
- To collect data and generate a bar graph depicting the results of this experiment.

7.1 Introduction

The early earths' atmosphere consisted of hydrogen gas, nitrogen gas, water vapor, methane, ammonia, carbon monoxide and carbon dioxide. Oxygen was not present until the evolution of the first photosynthetic organisms, the cyanobacteria, about 3.5 billion years ago.

Photosynthesis is the process that transforms light energy from the sun into chemical energy. Photosynthetic organisms include plants, algae, cyanobacteria and Euglena, a photosynthetic protozoa. Photosynthesis occurs inside the specialized organelles known as chloroplasts. Photosynthesis can be summarized by the following equation:

$$6\ CO_2 + 6\ H_20 \rightarrow C_6H_{12}O_6 + 6O_2$$

Photosynthesis is not one reaction but rather a metabolic pathway, a series of reactions that can be divided into 2 separate phases. In the first phase, the light dependent reaction, the pigment chlorophyll absorbs light energy. This energy is used to split water molecules. The breaking of the water molecules releases energy that is stored as ATP and NADPH and oxygen is released as a bi-product. The second phase of photosynthesis is the Calvin Cycle where the energy harvested in the light dependent reaction is used to fixate carbon atoms from carbon dioxide and glucose is formed.

7.2 Experimental Design

In this experiment you will examine how different light conditions effect the rate of the light dependent reaction of photosynthesis. The rate of this reaction can be estimated by measuring oxygen production in disks cut from spinach leaves.

Leaves are riddled with gas-filled intracellular spaces, evident by observing a leaf floating on water. Small circular disks are prepared from spinach leaves. The leaf disks are then placed in a solution of sodium bicarbonate and subjected to a vacuum. The trapped gases will be removed and replaced by the bicarbonate solution causing the disks to sink to the bottom of the flask. The disks will be divided into 3 petri dishes and placed under different light conditions – dark, room light, and under a sunlamp. As the light dependent reaction occurs oxygen will diffuse into the intracellular spaces replacing the sodium bicarbonate solution with this gas. Once enough oxygen has been generated, the leaf disks will regain buoyancy and turn on one edge or float to the surface. The percentage of leaf disks turned or floating on edge after 20 minutes will be used to measure the rate this phase of photosynthesis.

Activity: Effect of different light conditions on the rate of photosynthesis

Materials

Spinach leaves, cork borer, wood or thick cardboard

0.2 % sodium bicarbonate solution

250 ml flask side armed flask with 1-holed rubber stopper

Petri dishes – 2 clear, 1 black per group

Sunlamp

Culture dish

Forceps

7.2 Procedure

1. Pour the sodium bicarbonate solution into the 3 petri dishes, filling 2/3 of the way. Fill the 250 ml side armed flask with approximately 100 ml of sodium bicarbonate solution.
2. Take spinach leaves and cut 60 disks using the core borer. Spinach leaves may be stacked together. Avoid areas that contain large veins.
3. Add spinach disk to the 250 ml side armed flask.
4. Attach the vacuum tubing to the side arm of the flask. Put the rubber stopper firmly over the mouth of the flask. Use tape or your finger to secure the hole in the stopper.
5. Attach flask to vacuum nozzle. Leave the disks under the vacuum for 15 – 20 second increments. Turn off the vacuum, slowly release the tape/finger on the hole in the rubber stopper to check if the disks have sunk to the bottom of the

flask. Repeat this process until all disks have sunk to the bottom of the flask. Do not over aspirate as this will damage the plant tissue.

6. Pour the disks into the culture dish, discard any that may still be floating.
7. Using forceps gently transfer 15 – 20 disks to each of the 3 petri dishes. The black dish represents "the dark" light condition. Place one of the clear dishes under the sunlamp. Place the second clear dish on the bench top away from the sunlamp.
8. Wait 20 minutes, you can answer many of the questions in the analysis section of this lab while you wait.
9. After 20 minutes count the number of disks that have floated or are standing on edge in each petri dish and calculate the percentage floating/standing on edge. Record the data in table 7.3 and generate a bar graph to represent the data.

7.3 Analysis and Results

Data Table – Results of Different Light Conditions on Spinach Leaf Disks

Light Condition	# Disks Floating	% Disks Floating
Dark		
Room light		
Sunlamp		

Graph - Prepare a bar graph of the % disks floating for each light condition. Be sure to give the graph a title and label each axis.

Title: _____

x axis label: _____

Analysis

1. Predict what the results will be if your hypothesis is supported.

2. What is the independent variable in this experiment

3. What is the dependent variable in this experiment.

4. Name one other independent variables that could affect the rate of photosynthesis?

5. What petri dish is serving as the control group?

6. What is the function each of the following components of photosynthesis:

    a. Light -

    b. H_2O -

    c. O_2 -

    d. Chloroplasts -

7. In this experiment the spinach leaves were infiltrated with sodium bicarbonate solution. Why did the disks float again when exposed to light?

Laboratory Exercise 7
NOTES

Lab Exercise 8

HUMAN GENETICS & CYTOGENETICS

Purpose

- To explore the inheritance of single gene traits
- To distinguish between autosomal and sex-linked gene inheritance
- To construct and read karyotypes
- To identify characteristics of chromosomes
- To associate genetic make-up with phenotype

8.1 Human Genetics - Terms and Concepts

Chromosome	One of 46 strands of DNA in a human nucleus.
Gene	A unit of heredity located on a chromosome. Example: eye color gene
Allele	An alternate form of a gene. Example: blue eyes
Homologous Chromosome	Homologous chromosomes contain the same genes at the same location; however, the alleles may differ.
Dominant allele	The dominant allele masks the recessive allele. Example: people with dominant allele, B, make brown eye pigment. Both genotypes BB and Bb give brown eyes. Use an uppercase letter for the dominant allele.
Recessive allele	The recessive allele is masked by the dominant allele. People the genotype bb have blue eyes. Use a lowercase letter for the recessive allele.
Homozygous	The two alleles on homologous chromosomes are the same Example: BB = two brown eye alleles bb = two blue eye alleles
Heterozygous	The alleles on homologous chromosomes are different. Ex: Bb
Genotype	The genetic makeup of an individual. Example: BB, Bb, or bb

Phenotype	The appearance of the individual Ex: brown eyes, blood type A
Punnett Square	Diagram used to illustrate transmission of traits through generations
Autosomes	Chromosome pairs 1 – 22 are not different in males and females
Sex Chromosomes	Sex chromosome pairs are shown last in the karyotype. Females possess 2 large X chromosomes while males possess one X chromosome and a smaller Y chromosome.

8.2 Human Traits Determined by a Single Gene

Each human somatic cell has 46 chromosomes in its nucleus. Each chromosome contains thousands of individual genes. There are thought to be about 20,000 genes in human DNA. Interestingly, the number of genes in human DNA is not appreciably different from the number of genes in chimpanzees or mice.

Most traits are determined by more than one gene. For example, skin color and height are determined by many genes. Some phenotypes however, are determined by a single gene. We will explore some of these single gene traits in the laboratory.

1. Use a lab partner to help you determine your phenotype for the traits listed.

2. Complete **Table 8.1.** Use two alleles per trait for the genotype. *If you exhibit the dominant phenotype, use a dash to represent the second allele.*

Example:

B_ genotype for the phenotype of brown eyes (dash indicates second allele could be B or b which means a genotype of BB or Bb)

bb genotype for the phenotype of blue eyes

Interlocking fingers
Interlock fingers. Observe which thumb is on top (right or left). The tendency to place the left thumb on top is due to a dominant allele (I) and the genotype is I- (either II or I-). The right thumb on top is determined by the ii genotype.

Ear lobes
The dominant allele (E) results in the phenotype of free earlobes. The recessive allele (e) is for attached earlobes.

Widow's peak
Widow's peak occurs when the hairline forms a distinct point in the center of the forehead. Widow's peak is controlled by a dominant allele (W).

Tongue curling
A dominant allele (T) gives the individual the ability to curl the tongue in a U-shape.

Hitch hiker's thumb
A person homozygous recessive for this trait (hh) can bend the last (distal) thumb joint back to about a 90 degree angle. Those with the H allele cannot.

Pigmented iris
A person with the B allele has brown eyes. The recessive allele (b) encodes blue eyes.

PTC tasting
If you can taste PTC, you have the dominant allele (P). Place the PTC paper on your tongue for a few seconds. If you cannot taste anything, you do not possess the dominant allele.

Table 8.1. Human Traits Controlled by a Single Gene

TRAIT	Describe Phenotype	Genotype
Interlocking fingers		
Earlobes		
Widow's peak		
Tongue curling		
Hitch hiker's thumb		
Eye color		
PTC tasting		

8.3 Sex linkage

About 2000 genes reside on the X chromosome. Female humans inherit 2 X chromosomes, one from the mother and one from the father and so, have 2 alleles for each X-linked gene. Males inherit an X chromosome from the mother and a Y from the father and have 1 allele for each X-linked gene. The Y chromosome determines sex in humans and many other animals.

Colorblindness is the inability to perceive differences between some colors. The red-green colorblindness, or deuteranopia, gene is located on the X chromosome. Individuals with this type of colorblindness have difficulty discriminating reds, yellows, and greens. About 10 million US men are colorblind (~7% of the male population).

Go to the following website - http://colorvisiontesting.com/ishihara.html and complete the online test for colorblindness.

The use of the X to symbolize X-linked genes is useful in pedigree analysis.

Alleles

X^D = normal allele

X^d= colorblind allele

Male genotypes		**Female Genotypes**	
X^DY	normal vision	X^DX^D	normal vision
X^dY	colorblind	X^DX^d	carries colorblind allele, normal vision
		X^dX^d	colorblind

1. Examine the colorblindness chart. Are you colorblind? _____

2. What is your probable genotype? _____

3. A carrier female has children with a male who has normal vision. What is the chance their son will be colorblind? Show the cross and explain your answer. Use the allele symbols X^D and X^d in the Punnett square.

8.4 Cytogenetics - Introduction

The genetic material of humans is contained on 23 pairs of chromosomes in the nucleus of each somatic cell. 22 pairs are **autosomes** and 1 pair is the **sex chromosomes**. Human females inherit 2 X sex chromosomes while males inherit 1 X and 1 Y chromosome.

Cytogeneticists examine chromosome structure, seek out causes of chromosomal abnormalities, and study how chromosome structure relates to an individual's phenotype (appearance and biochemical traits). Abnormalities in chromosomes can result in early embryonic death, congenital defects, development of cancer, and infertility or sterility.

Studies of chromosomes begin with the extraction of chromosomes from cells. Chromosomes are then placed on a glass slide, stained with dye, and examined under a microscope. Each chromosome pair is assigned a number (from 1 to 22, then X and Y) that is based on staining pattern and length. A **karyotype** is a representation of a person's chromosomes. The chromosomes are shown in pairs and arranged in order of decreasing size. Individual **genes** cannot be seen on karyotypes.

What to look for in a human karyotype

- Are there 46 chromosomes?
- Is there one pair of each autosome and 1 pair of sex chromosomes?
- Are there any deletions, rearrangements, or other abnormalities in the chromosomes?

Chromosomal notation: Karyotypes are presented in a standard form. First, the total number of chromosomes is given, followed by a comma, and the sex chromosome constitution.

Normal human female is designated as **46, XX**
Human male with an extra chromosome 15 is designated as **47, XY, 15+**
Human female with an extra X chromosome is designated as **47, XXX**

There are many disorders that can be diagnosed by examining chromosomes. In **Down syndrome**, an extra chromosome 21 is present. In **Turner syndrome**, a sex chromosome is missing so that the individual has 45 total chromosomes. **Fragile X syndrome**, the most common inherited cause of mental retardation, takes its name from the appearance of the X chromosome.

8.5 Cytogenetics – Terms and Concepts

Karyotype	A picture of an individual's chromosomes in which the chromosomes are paired and arranged in order of decreasing size.
Monosomy	The lack of one of a pair of chromosomes. A common monosomy is X chromosome monosomy, known in humans as Turner syndrome and denoted as 45, X.
Trisomy	Condition of having three chromosomes of a particular type**.** A common trisomy in humans is Down syndrome, or trisomy A male with trisomy 21 is denoted as 47, XY, 21+.
Nondisjunction	Failure of chromosome pairs to separate during meiosis I or meiosis II.

8.6 Activity: Reading Karyotypes

1. Examine Karyotype 1 on the lab handout.

Is this a male or female? _____
Designate the karyotype using the symbols in the above examples _____

2. Examine Karyotype 2 on the handout. This newborn child was seen in neonatal ICU. Defects included: cleft lip and palate, nonfunctional eyes, polydactyly of the hands, and heart defects.

Is this a male or female _____
What type of chromosomal abnormality is shown? _____
How many total chromosomes does this individual exhibit? _____
Designate the karyotype using the symbols in the above examples _____
How could this abnormality occur? _____

3. Examine Karyotype 3 on the handout. The mother of this 11-year old patient, stated that her child has always been a "little behind" in subjects requiring language skills. The patients' teachers have commented that he/she is often inattentive and hyperactive. The patient is at the 85^{th} percentile for height / 45^{th} percentile for weight.

Is this a male or female _____
What type of chromosomal abnormality is shown? _____
How many total chromosomes does this individual exhibit? _____
Designate the karyotype using the symbols in the above examples _____

Why must the abnormality be the result of nondisjunction occurring in a sperm cell?_____.

4. Examine Karyotype 4 on the handout. This 10 year old patient demonstrated poor academic performance and often experienced low self-esteem. There are often no physical manifestations seen other than the patient being in the 80% percentile for height with an occipitofrontal head circumference in the 30% percentile.

Is this a male or female _____
What type of chromosomal abnormality is shown? _____
How many total chromosomes does this individual exhibit? _____
Designate the karyotype using the symbols in the above examples _____
How could this abnormality occur? _____.

8.7 Questions for Review

_____1. Is an XY individual male, or female?

_____2. Which chromosomes are the autosomes?

_____3. Which parent (mother or father) determines if a son is colorblind?

_____4. Which chromosomes are the sex chromosomes?

_____6. Genotype of a homozygous recessive individual (any letter)

_____7. Where do girls get their X chromosomes from (which parent, or both?)

_____, _____8. List two genotypes that represent homozygous individuals

_____9. List a heterozygous genotype

_____10. The gene for colorblindness is located on this chromosome

_____11.An allele that is masked by the expression of its homologous allele

_____12. An allele represented by an uppercase letter

Laboratory Exercise 8
NOTES

Lab Exercise 9

USING GENETIC CROSSES TO ANALYZE A STICKLEBACK TRAIT

Purpose

1. Characterize stickleback fish according to the presence or absence of pelvic spines.
2. Use Punnett squares to predict the frequencies of genotypes and phenotypes in the offspring of a genetic cross, based on the genotypes of the parents.
3. Develop hypotheses about whether a phenotype is dominant or recessive.
4. Evaluate the evidence in support of or against the hypothesis

9.1 Introduction

Geneticists breed organisms with different characteristics, or phenotypes, to answer questions about how the phenotypes are inherited. Is one phenotype dominant and one recessive? Is each phenotype controlled by a different version (allele) of a single gene, or are many interacting genes involved? In this activity, you will answer these questions by analyzing the outcome of breeding a stickleback with pelvic spines and one without pelvic spines—the same procedure, called a genetic cross, that Dr. David Kingsley described in the film *Evolving Switches, Evolving Bodies*.

The fish shown in the photo above is a marine three spine stickleback. Like all marine and sea-run stickleback, this fish has a pair of pelvic spines (only one is visible in the photo), which serve as a defense from large predatory fish. In some freshwater populations, such as in Bear Paw Lake, Alaska, stickleback lack pelvic spines. (The scale is in centimeters.) How are these two phenotypes inherited?

Materials

Stickleback trait sheet
F_1 stickleback fish cards
F_2 stickleback fish cards

PROCEDURE

Watch the short film entitled *The Making of the Fittest: Evolving Switches, Evolving Bodies (*https://www.youtube.com/watch?v=Pv4Ca-f4W9Q&feature=youtu.be)

9.2 Stating the Hypothesis: Which Phenotype Is Dominant?

In this activity you will examine the offspring of a genetic cross between a stickleback from the ocean (marine stickleback) and a freshwater stickleback from Bear Paw Lake.

1. Based on what you learned in the film, what are the phenotypes of these two parental stickleback fish?

Indicate your choice with a check mark.
Marine __ pelvic spines present __ pelvic spines absent
Bear Paw Lake __ pelvic spines present __ pelvic spines absent

2. If we start with the simplest assumption that the presence or absence of pelvic spines is controlled by a single gene with two alleles, how would you denote the genotype of the two **homozygous** parents?

Marine _____ Bear Paw Lake _____

3. What is your hypothesis for which phenotype is dominant and which one is recessive?

4. Based on your hypothesis, what would you expect to be the results of the cross between the marine and Bear Paw Lake stickleback parents? Make your prediction using the Punnett square below.

5. What would be the ratio of stickleback with pelvic spines to stickleback without spines in the first filial (F_1) generation?
6. Using the Punnett square below, what do you expect would be the result of crossing two F_1 fish to produce the second filial (F_2) generation?

7. What would be the ratio of stickleback with pelvic spines to stickleback without spines in the F_2 generation?

8. If you had 40 offspring in the F2 generation, approximately how many would you expect to have pelvic spines and how many to lack pelvic spines?

9.3 Obtaining the Data: Do the Results from the Experiment Support Your Hypothesis?

Now you will use the fish cards to see whether the result of the crosses described in Part 1 support your hypothesis. The cards show photographs of stickleback fish that were stained with a
solution that turns bones red, making them easier to see. There should be two sets of cards: the first set (16 cards) represents the first filial (F_1) generation and the second set (40 cards) the second filial (F_2) generation. You will be sorting these cards according to whether the fish have pelvic spines.

1. Sort the F_1 set of cards into two separate piles: fish with pelvic spines and fish without
pelvic spines.
2. Repeat the same procedure with the F_2 set of cards.
3. Count and record the total number of fish with each phenotype in the table below.

Table 1. Results of a Cross Between Marine and Bear Paw Lake Stickleback

Generation	Fish with pelvic spines	Fish without pelvic spines
P	1	1
F_1		
F_2		

4. What is the ratio of fish with pelvic spines to fish without pelvic spines in the F_1 generation? _____

5. What is the ratio of fish with pelvic spines to fish without pelvic spines in the F_2 generation? _____

6. Do these results support the hypothesis that the presence or absence of pelvic spines is controlled by a single gene? Explain using evidence.

7. According to these results, which phenotype is dominant and which is recessive? Explain using evidence.

Laboratory Exercise 9
NOTES

Lab Exercise 10

PROTEIN GEL ELECTROPHORESIS: A SIMULATED TEST FOR DNA FINGERPRINTING

Purpose

- To conduct the Kastle-Meyer test to detect blood residue left at a simulated crime scene.
- To conduct gel electrophoresis to analyze simulated DNA samples for fingerprinting.

INTRODUCTION

In this lab your instructor will explain a crime scenario that has taken place in the MS building. You will be presented with evidence that will be tested for the presence of blood and DNA fingerprinting will be conducted.

SAFETY NOTE: Human blood or blood products are not used in any MCCC laboratories. Sheep blood which is purchased sterile and free of antibiotics and preservatives is used to generate blood stained materials. Dye compounds will be used to mimics the response of DNA fragments subjected to gel electrophoresis.

10.1 Blood detection using the Kastle-Meyer test

The Kastle-Meyer test is a quick inexpensive test used to analyze evidence at a crime scene for the presence of blood. Phenolphthalein reacts with hydrogen peroxide in the presence of hemoglobin to turn from colorless to pink. This reagents provide a presumptive test for blood, as food samples which contain hemoglobin (meat) and certain vegetables will also generate a positive response. An additional benefit of this procedure is that the samples remain intact and can be used in further testing including DNA analysis.

PROCEDURE

1. Swab evidence with a dampened cotton swab
2. Add swab to test tube containing about 1 cm of distilled water, swish to release contents.
3. Add several drops of Kastle/Meyer reagent.
4. Add several drops of hydrogen peroxide.
5. A positive result will give a bright pink color.

Based on the scenario described by your instructor, enter a description of the 2 pieces of evidence and indicate if the samples were positive or negative for blood residue.

TABLE 10.1: Blood Detection Results

Sample	Result: positive or negative
1 –	
2 –	

10.2 DNA Fingerprinting

DNA fingerprinting is routinely used today to establish paternity, in the diagnosis of inherited disorders, and for use in criminal cases. DNA fingerprinting enables forensic investigators to determine whether two DNA samples originate from the same individual. Not all of the DNA present is used in this analysis. Restriction enzymes act as molecular scissors and are used to cleave DNA molecules at specific points. Over 2,500 different restriction enzymes have been identified. These enzymes are produced by bacteria and are used to destroy foreign DNA such as bacteriophages - viruses that infect and replicate within a bacterium.

For example the restriction enzyme EcoR1, isolated from E. coli, cuts DNA at the sequence GAATTC.

The length and the number of the fragments produced depends upon the frequency and the distance between the recognition sites. This distinct pattern is known as restriction fragment length polymorphisms (RFLP's) which are unique to each individual therefore forming a DNA fingerprint.

After DNA samples are cut by restriction enzymes, the fragments are separated using gel electrophoresis. PCR, polymerase chain reaction, can be used to analysis very small or degraded samples. This enzyme amplifies even trace amounts of DNA present. The length of the segments analyzed are much smaller and the repeat sites are called microsatellites.

The phosphate group of the DNA molecule is negatively charged which gives the fragments an overall negative charge. In an electrical current the negatively charged fragments will be attracted to the positive pole. Smaller fragments will migrate faster and further in a given time period. Therefore the fragments are separated by size. After separation radioactive markers are added which are complementary to the separated fragments. Photographic film is placed over the gel and the areas that are exposed to the radioactive markers darken. This generates a series of lines that resemble a bar code. The film then becomes the DNA fingerprint. In this experiment we will be using a mixture of dyes that will simulate the migration of DNA fragments.

10.3 Gel electrophoresis

Obtain simulated DNA samples and perform a gel electrophoresis generating a DNA fingerprint. Use this information to determine the guilt or innocence of the suspect.

Step 1: Setting up the agarose gel

1. Obtain a gel former, comb, and masking tape. The instructor will demonstrate how to build the gel. Place the gel set-up in a safe location. Once the gel has been poured, do not move until solidified.

2. Gently pour the agarose into the gel former until the level of the agarose is about ¾ the length of the teeth of the comb. The tape will prevent the molten gel from spilling out of the apparatus. The agarose is kept in a 60^{o}C water bath to keep it molten. Agarose solidifies at room temperature so **return the flask to the bath immediately after use**.

Step 2: Preparation of samples and gel electrophoresis

1. Obtain two simulated DNA samples A and B. The samples are not DNA but dyes that migrate in a manner similar to restriction fragments.

2. Remove the tape from the gel former. Carefully remove the comb. Your instructor will demonstrate how to load the samples on the gel. Record which sample you load into which lane of the gel.

3. Seal the top of the wells with a drop of molten agarose as demonstrated.

4. Place the gel in the gel box. Samples are closest to the negative (black) electrode. Add 1X electrophoresis buffer to just cover the gel. Place the cover on the apparatus and connect the red and black leads (red is positive, black is negative).

5. Notify your instructor at this point and s/he will turn on the voltage for you. The gel is run at ~90 volts for 20 minutes. Once the voltage is applied, observe that samples migrate towards the positive electrode (red). Do not leave the gel until you are sure that the samples are migrating in the correct direction.

   * Complete Section10.5, Concept Review while the gel is running

6. Turn off the power supply. Unplug the leads. Visualize the bands of dye which represent the DNA fragments.

CLEAN UP:
Rinse gel equipment with water. Put gloves, paper towels in the regular trash. Dispose of electrophoresis buffer as instructed (you may recycle it, check with instructor). Wash hands.

10.4 RESULTS

Sketch the bands on the gel when done electrophoresis. Label lanes (1 & 2) and complete Table 10.2

Title _____

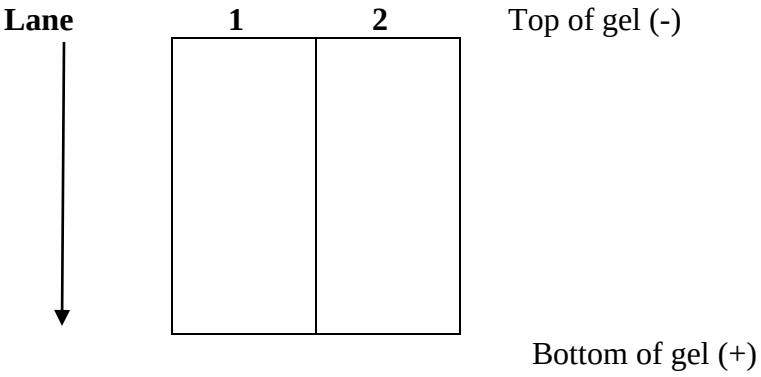


Table 10.2 Electrophoresis of DNA Samples			
Lane	Sample	# bands	Migration (mm)
1			
2			

10.5 Concept Check Questions

1. What is the basis for the separation of DNA fragments using electrophoresis?

2. What is the charge of the samples?

3. What do you think would happen in the molecules had the opposite charge?

4. What would happen if you attached to red and black electrode on the wrong sides?

5. If a DNA molecule has three restriction sites, for restriction enzyme A, how many fragments would be produced.

6. Using restriction enzymes a piece of DNA is cut into 4 pieces, the fragments are separated using gel electrophoresis. Below are the results:

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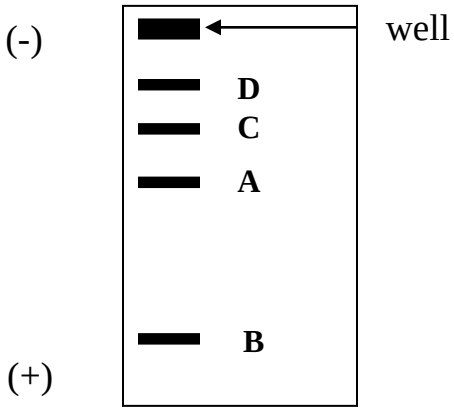


a. Which fragment would be the largest?

b. Which fragment would be the smallest?

7. Below are one strand of two samples of DNA.

Sample #1: **C A G T G A T C T C G A A T T C G C T A G T A A C G T T**

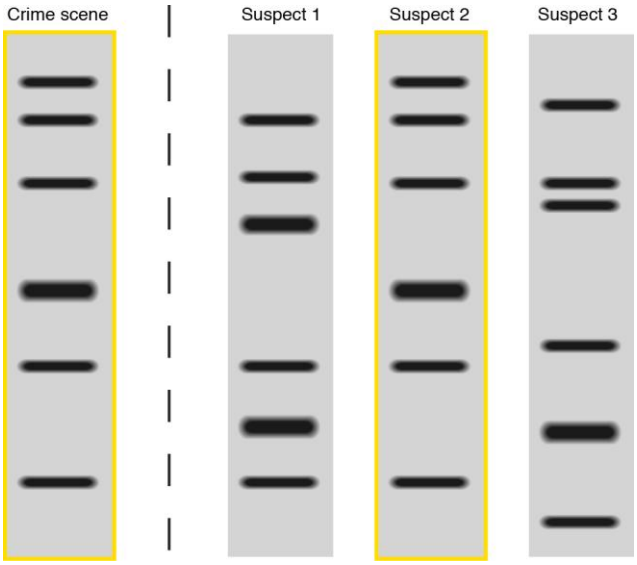
Sample #2: T C A T G A A T T C C T G G A A T C A G C A A A T G C A

Both samples are treated with a restriction enzyme restriction enzyme EcoR1 which cuts DNA at the sequence GAATTC. Indicate the number of fragments that would be generated by each sample.

Sample #1 # of fragments: ____

Sample #2 # of fragments: ____

8. Compare the DNA fingerprint from the crime scene to the 3 suspects. Which suspect committed the crime? _____



Laboratory Exercise 10
NOTES

Lab Exercise 11

ISOLATION OF DNA FROM PLANTS

Purpose

- To extract nuclear DNA from plant cells
- To learn the role of procedures and reagents in the isolation of nuclear DNA

11.1 Introduction

Deoxyribonucleic acid (DNA) is located in the **nucleus** of eukaryotic cells (animals, plants, fungi, and protists). DNA contains information to direct the cell in the manufacture of **proteins**. Proteins control development, organ function, metabolism, enzymatic reactions, photosynthesis, muscle action, brain activity, and many other cellular processes. DNA is often referred to as the "blueprint for life".

DNA is a polymer composed of the **nucleotide bases** guanine (G), adenine (A), thymine (T), and cytosine (C), and two sugar/phosphate backbones. Two DNA strands are twisted to form a **double helix**. The number of nucleotide bases (G, A, T, C) in each human cell is about 3 billion. The 3 billion base pairs in the **human genome** are located on 46 strands of DNA called **chromosomes**. The Human Genome Project has determined the order of the nucleotides on each chromosome. A **gene** is a sequence of nucleotide bases (DNA) that codes for a specific protein. Human DNA contains about 20,000 genes while the cells of the rice plant contain over 40,000 genes.

In the DNA isolation procedure, plant cell walls and cell membranes are broken down by blending or mashing and heating the cells. Detergent in the extraction solution dissolves lipids in the cell membrane causing the cells to **lyse**. When cells undergo lysis, the cellular components, including the DNA, are released. The technique of **filtration** uses a medium, in this case cheesecloth, to separate solids from liquids. The resultant material is referred to as **filtrate**. When cold ethanol is added to the filtrate, DNA **precipitates** at the water/ethanol **interface**. Although an individual DNA molecule is not visible with the naked eye, DNA isolated from large quantities of cells can be observed.

11.2 Procedure: Isolation of DNA from plants

Wheat germ is obtained from wheat seeds. The germ is the embryo. A seed consists of the embryo surrounded by a seed coat and contains nutrition for the seed. When wheat seeds are milled into white flour, the germ and bran (fiber) are removed. Whole wheat flour contains all parts of the wheat seed. Strawberry seeds also contain large amounts of DNA, a commonly cultivated strawberry, *Fragaria ananassa*, is octoploid – contains eight sets of chromosomes in each cell.

Materials

Raw, untoasted, wheat germ	250 ml beaker (2)
Strawberries	Ice in tray
Spatula	50 ml beaker
Electronic balance	50 ml graduated cylinder
Mortar and pestle	Small strainer
DNA extraction buffer	Wooden rod
(water, salt, and detergent)	Wax pencil
95% Ethyl alcohol, ice cold	

Lyse plant cells:

1. Make a crease in a piece of weigh paper or weigh boat. Place the weigh paper/boat on the balance and zero the balance. Weigh 1 gram of raw wheat germ. Add the wheat germ to a 250 ml beaker.

2. Weigh 20 grams of strawberry. Mash thoroughly in a mortar and pestle and add to a separate 250 ml beaker

3. Use the graduated cylinder to measure 30 ml DNA extraction buffer that has been pre-heated to 60^{o}C.

4. Swirl mixtures constantly for 5 minutes.

5. **Questions**

a. What occurs to cell membranes when exposed to detergent in the DNA extraction?

b. Describe cell lysis.

c. How is the germ of the wheat plant similar to a strawberry seed?

Filter plant cell extract:

1. Obtain a small strainer and 2 clean 50 ml beakers. . Carefully strain the wheat germ extract until 10 ml have been obtained. Repeat for the strawberry extract using a separate beaker.

2. Discard the solid plant remnants (regular trash).

3. Questions

    a. List 3 molecules that have passed through the cheesecloth into the filtrate

    b. On what molecular basis does filtration select - charge, solubility, size, density?

    c. Where is the plant DNA now located?

Precipitate DNA:

1. Use a graduated cylinder to measure 2X volume (20 ml) ice-cold ethanol. **Slowly** pour the ethanol down the side of each beaker so that the ethanol is layered over the filtrate. DO NOT MIX.

2. Let the preparation sit undisturbed for up to 5 minutes. Observe the interface.

3. Sketch one of the beakers. Label: ethanol, filtrate, DNA at interface.

Sketch:

Spool DNA

1. Spool DNA from one of the beakers onto a wooden rod. Obtain as much DNA as possible on the rod. Lift the DNA out of the solution and blot the excess alcohol on a paper towel.

2. Describe the appearance of the DNA in terms of color and substance

3. What is the role of ethanol in the DNA extraction procedure?

4. Use a wooden rod to spool the DNA as demonstrated by the instructor.

Record Observations (the appearance of DNA and other observations during spooling)

11.3 Questions

1. What is the function of DNA?

2. Where is DNA located in eukaryotic cells?
3. What are the 4 building blocks of DNA?
4. How many chromosomes DNA are located in a human cell nucleus?
5. Genes carry the code for the cell to make specific proteins. About how many genes are contained in the human genome?
6. What is the embryo of the wheat plant referred to as?
7. What occurs at each of the following steps?
    a. Lysis

    b. Filtration

    c. Precipitation

8. What is the role of each of the following reagents or steps in the DNA extraction procedure? Use the appropriate technical terms.
    a. Detergent
    b. Ice-cold ethanol

9. The strawberry is an octoploid plant (8 sets of chromosomes in each cell nucleus). Humans are diploid (2 sets of chromosomes). How many sets of chromosomes are in wheat, a hexaploid plant?

Laboratory Exercise 11
NOTES

Lab Exercise 12

ANIMAL TISSUES

Purpose

- To compare the four principal types of animal tissue and identify tissue types in an organ specimen.
- To understand the biology and cells of blood.
- To explore the evolution of mammalian red blood cells
- To perform a simulation of blood typing.

12.1 Introduction to Tissue Types

The cells of multicellular organisms - plants, fungi, and animals are organized into multicellular tissues. A tissue is a group of similar cells working together to perform a common function. An organ consists of multiple tissue types. An organ system is composed of multiple organs working together. Animals contain **4 principal tissue types**:

Muscle tissue can contract which leads to movement. Voluntary muscles are attached to the skeleton and are under conscious control. Involuntary muscles include heart muscle and muscles of the stomach and other organs. Muscles are responsible for movements such as running and facial expression, and for less obvious functions such as blood vessel diameter. Muscle tissue composes the extrinsic eye muscles that move the eye. These muscles are located external to the eyeball.

Nervous tissue conducts electrical impulses within the brain, spinal cord, and nerves. Nervous tissue is composed of neurons, cells that have the ability to receive, process, and send impulses to all regions of the body. The optic nerve is located in the back of the eyeball and carries impulses of vision from the retina to the visual cortex of the brain. The retina contains receptors that convert light waves to electrical impulses. A damaged optic nerve or retina can result in blindness.

Epithelial tissue lines surfaces of the body and composes glands. Epithelial tissue lines the inside of the mouth and other entrances into the body. The lining of the stomach, blood vessels, and heart composed of epithelial tissue. Epithelial tissue covers the cornea of the eye.

Connective tissue includes **blood**, fat, bone, cartilage, ligaments, tendons, the dermis of skin, and other structures. Adipose tissue (fat) is found external to the eyeball to cushion the eye. The sclera (white) of the eye is composed of fibrous connective tissue.

12.2 Organ Specimen

Sheep Eye

Wear gloves and goggles. Place a preserved sheep eye on a dissecting tray. You will also need scissors and a dissecting needle

A. Outer Eye For each structure, indicate the tissue type

Outer eye Structure	Function	Tissue type
Optic nerve	Sends impulses of vision to the brain.	_____
Extrinsic eye muscles	Move eyeball up and down, right to left	_____
Fat	On outside of the eye to provide cushioning	_____
Cornea	Transparent covering over front of eyeball	_____
Sclera	The "white" of eye. Fibrous membrane surrounding posterior 5/6ths of outer eyeball	_____

B. Inner Eye Cut eyeball into anterior and posterior portions. Make the circular cut a few millimeters posterior to the edge of cornea. Once you have the eye in two pieces, observe the following structures. Relate each structure to its function in vision. Describe in your own words in space provided.

Vitreous humor - gel-like substance fills posterior portion of inner eyeball. Maintains inner eyeball shape **Description**:

Lens - clear, oval structure behind pupil. The shape of the lens changes to focus light waves onto the retina for sharp vision. **Description**:

Pupil - round opening that allows light to enter eye. The diameter of the pupil is controlled to let more, or less, light enter the eye **Description:**

Iris - responsible for eye color. Contains muscles that control the diameter of the pupil

Description:

Retina - Cream-colored membrane stretched over inner back of the eyeball. Contains photoreceptors for vision. Composed of nervous tissue

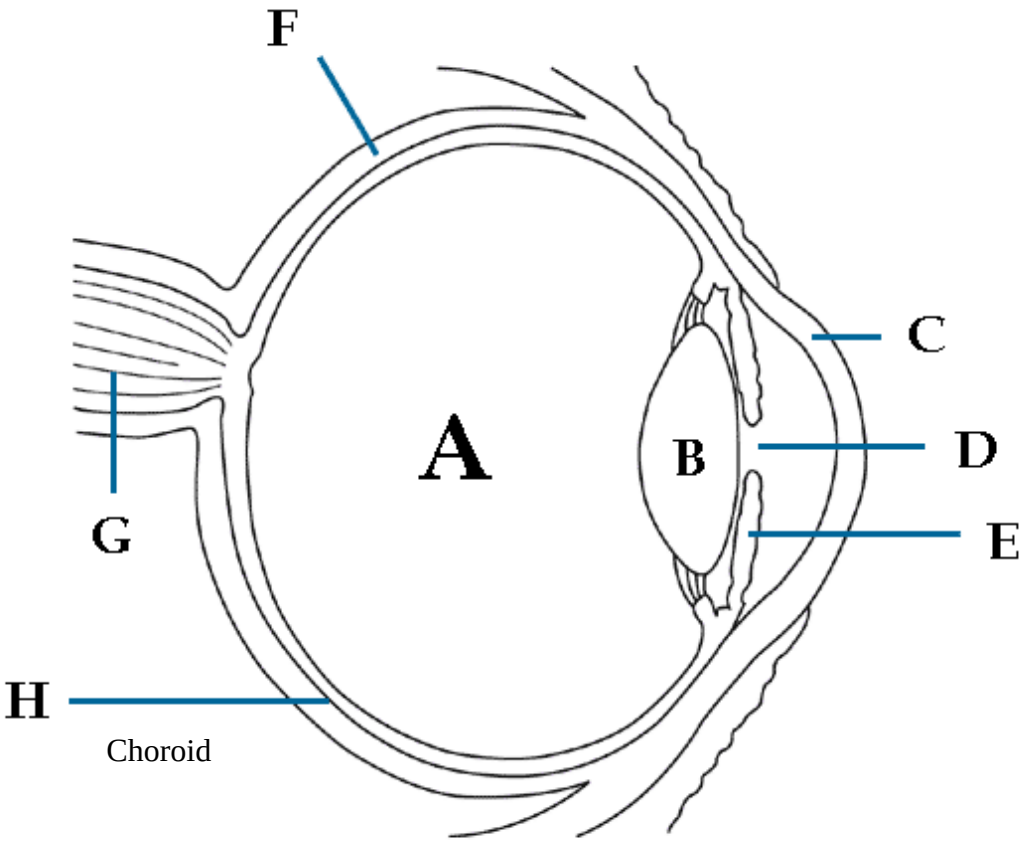
Description:

Choroid – Absorbs excess light

Description:

* Discard eye in bucket, wash utensils and rinse the tray with water. Wash hands

Label diagram of eye -
Lateral view of human eyeball. Fill in A – H.



http://commons.wikimedia.org/wiki/File:Eye_Diagram_without_text.gif

Questions for Review

Match the structure with the function

_____ opening that allows light to enter eye	A. Lens
_____ responsible for eye color	B. Choroid
_____ contains photoreceptors for vision	C. Retina
_____ protects the eye by absorbing excess light	D. Iris
_____ white layer that gives the eyeball its shape and protects it	E. Sclera
_____ maintains shape of the inner eyeball	F. Vitreous humor
_____ focuses light waves onto the retina	G. Pupil

12.3 Blood - Introduction

Blood is **a fluid connective tissue** that is composed of plasma and formed elements (blood cells). The fluid portion of blood is the **plasma** which is composed of mostly water plus blood proteins, ions, and salts. The remaining portion of the blood is composed of the **formed elements**, the cells of blood. Blood cells include red blood cells (erythrocytes), white blood cells (leukocytes) and platelets (thrombocytes).

Red blood cells (rbc) transport oxygen to tissues of the body. Hemoglobin molecules bind oxygen allowing each red blood cell to bind billions of oxygen molecules. There are over 5 million red blood cells in a drop of blood.

12.4 Evolution of Mammalian Red Blood Cells

Mammals evolved during the Triassic period around 250 MYA. At this time the earths' oxygen levels were about 50% less than that of today and even lower than that during the Jurassic period which gave rise to the emergence of birds. Under these environmental conditions, natural selection favored the loss of the nuclei in mammalian red blood cells. The absence of a nucleus allows for a greater volume of hemoglobin to be present and increases the rate of oxygen saturation. This also makes the red blood cells more flexible and able to pass through narrow capillary beds.

Mammalian lungs consist of alveoli, tiny air sacs that allow for gas exchange. Gas exchange in birds occurs thru flow tubes called air capillaries which increases the efficiency of respiration delivering more oxygen per breathe. Therefore was no selective pressure to eliminate the nuclei from birds' red blood cells. This is however not without a cost as toxins are also more rapidly transferred. Coal miners used to take caged canaries down into the mines. If toxic gases such as carbon monoxide were present the canary

would die giving the miners time to escape. The blood cells of other types of organisms' reptiles and amphibians are also nucleated.

Using the key determine which of the slides is human, rabbit (mammalian), frog, bird, or fish.

SLIDE A _____

SLIDE B _____

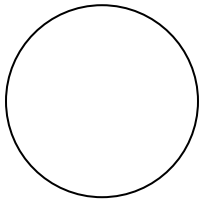
SLIDE C _____

SLIDE D _____

SLIDE E _____

Select a mammalian blood slide:

Sketch a red blood cell and a white blood cell (label the nucleus)



_____ total mag

12.5 Blood Typing

Terms and concepts

Self antigen - A substance on red blood cells that marks the cells as "self". Antigens are usually proteins or polysaccharides.

Foreign antigen - A substance that triggers an immune response by an organism because it is recognized as "non-self" or foreign. Foreign antigens can enter the body by inhalation (breathing), ingestion (eating), or injection (blood).

Antibodies - Produced by white blood cells (B lymphocytes) to react with foreign antigens and ultimately destroy them.

Antisera - Blood serum that contains antibodies to an antigen

Agglutination - A clumping reaction that occurs in blood when antibodies react to foreign blood cell antigens.

Transfusion - The process of giving blood from one individual to another. Only those with the same blood cell antigens are a match.

Red blood cells contain cell surface **antigens** that mark the cell as "self". These antigens are inherited from parent to offspring. Cells that are marked as self will not be targeted by the immune system. Cells that contain foreign antigens (not self) will be targeted by the immune system for destruction.

- If your blood cells exhibit the "A" antigen than you have **type A** blood.
- If your cells exhibit the "B" antigen then you have **type B** blood.
- People with **type AB** blood express both antigens
- Those with **type O** blood do not express either antigen. Type O blood is not recognized as foreign by anyone's immune system because there are no antigens to detect on the red blood cells.

The **Rh factor** is a separate blood cell antigen. If your cells exhibit the Rh antigen, you are Rh positive (Rh+). Cells that lack the Rh antigen are Rh-. Rh- blood will not be recognized as foreign in an Rh+ person.

Fill in Table 12.2

Table 12.2 Blood typing

BLOOD TYPE	**ANTIGEN(S)**	**Transfuse with**
Type A	A	Type A or Type O only
Type B		
Type AB		
Type O	none	
Rh positive(Rh+)	Rh	Rh + or Rh- blood
Rh negative(Rh-)		

Questions:

1. A and B are self markers known as _____

2. Antibodies are molecules produced by B lymphocytes (wbc) that attack _____

3. A person is blood type B-. Why can't this person receive B+ blood?

4. A universal donor is someone that can donate to people of all other blood types without fear of an antigen/ antibody reaction (agglutination). Which blood type is the universal donor (do not list the Rh factor)?

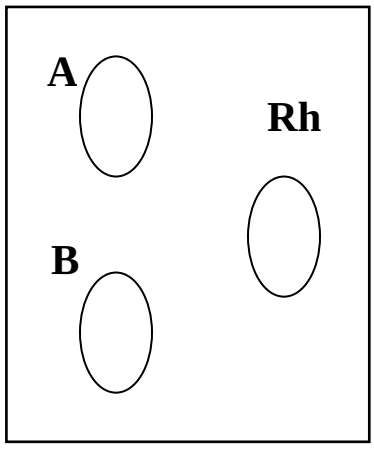
5. A universal recipient is someone who can receive blood from a person with any blood type. Which blood type is the universal recipient (do not include the Rh factor)?

ACTIVITY: Blood Typing Simulation

Note: No actual blood or blood products are used in the blood typing exercise.
Scenario, you are a technician at busy city hospital. A patient is admitted who has been in a serious car accident and has suffered severe blood loss. You need to blood type your patient and 3 potential blood donors to see which donor has a blood type acceptable to be used for a transfusion. Need to practice? Play the "Blood Typing Game."
https://www.nobelprize.org/educational/medicine/bloodtypinggame/gamev2/index.html

1. For each "individual" tested, you will need:
    - A blood typing tray
    - Bottles of Type A antisera, Type B antisera, and Rh antisera
    - Clean toothpicks to mix the reactions
    - Samples of " blood"

2. Add two drops of "blood" to each of the 3 wells on the tray



3. Add two drops of the appropriate antisera and stir with a toothpick (use a fresh toothpick each time)

4. Examine the tray to observe agglutination. Once you can ascertain the blood type of the individual, wash the tray. Fill in table 14.4

5. Rinse trays well and return to instructor

Table 12.3

Blood Typing Results	Agglutinate with anti-A?	Agglutinate with anti-B?	Agglutinate with anti-Rh?	Blood Type
Patient _____				
Donor 1 _____				
Donor 2 _____				
Donor 3 _____				

Question:

Which donor(s) have a blood type that can be safely transfused to the patient _____

Lab Exercise 12
NOTES

Lab Exercise 13

MICROBIOLOGY, FOOD MICROBIOLOGY AND DISEASE TRANSMISSION

Purpose

- To classify individual bacteria by shape
- To obtain data concerning the relative abundance of bacteria in common environments
- To describe bacterial colony shapes and growth patterns
- To utilize the process of fermentation to make yogurt
- To engage in an epidemiological investigation of disease transmission

13.1 Introduction - Microbiology & Food Microbiology (weeks 1 & 2)

Bacteria are single celled prokaryotic organisms and are the most numerous and diverse organisms on Earth. Bacteria are found in soil, air, water, the human body, and almost every other location imaginable. Most bacteria are not pathogenic. For example, *E. coli* are normally found in the human intestine and yogurt contains live *Acidophilus* cultures. That bacteria are found at extreme temperatures and pH attests to their being the most successful group of living things on Earth.

Fungi are eukaryotic organisms most of which are harmless. Fungal spores can be airborne and will grow into fuzzy mats when provided with a nutrient source.

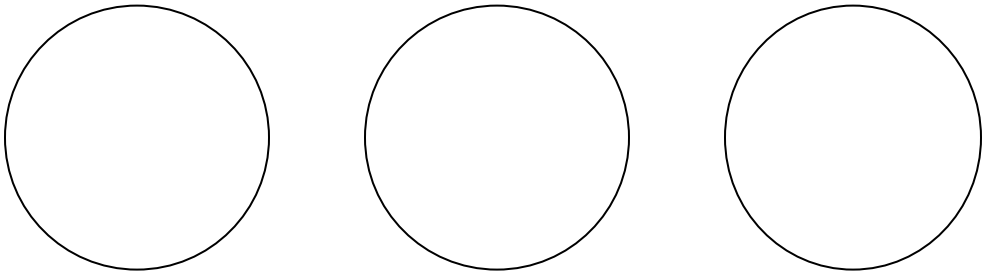
Laboratory Safety

- wipe lab bench with disinfectant before and after the lab
- wash hands with soap and water before leaving the lab
- wear disposable lab gloves and safety glasses

13.2 Classifying Individual Bacterial Cells by Shape

Individual bacterial cells can be classified by three basic shapes. Spirilla are corkscrew shaped, cocci are round, and bacilli are rod-like. Examine the prepared slides set and sketch the appearance of each bacterial type. Sketch one or two individual cells:

Spirillum | Coccus | Bacillus



Total Mag. _____ Total Mag. _____ Total Mag. ____

13.3 Testing Environments for the Presence of Bacteria/Fungi

Materials:

agar plates
wax pencil or marker
sterile water
sterile swabs
moist kitchen sponge
hand soap

Bacteria and also fungi (includes molds) inhabit many different environments. In the following procedure, you will test unwashed and washed hands, a kitchen sponge, and a fomite (inanimate object that may harbor bacteria) for the presence of bacteria and fungi. Fungi can be distinguished from bacteria by a fuzzy appearance.

Hypothesize about which environment (s) will exhibit the greatest number of bacteria and which might contain fungi. What is the basis for your hypothesis?

Hypothesis

Procedure:

1. Obtain 2 agar plates. Hold the lid on and draw a line with a wax pencil on the bottom of the plates to divide the plates into two equal halves. The bottom of the plate contains the agar.

2. In small letters label the areas “unwashed”, “washed”, “sponge”, “fomite”. Write your initials on the plates. Label the bottom of the dish, not the lid.

3. Select one person to be the test subject. Lightly press three fingers on the agar on the side of plate marked “unwashed”. Do not break the agar. Close the dish. Wash hands with soap, dry, and repeat procedure on the side of the plate marked “washed”.

4. Obtain a small piece of the moist sponge. Open the plate and lightly press the sponge onto the agar. Return the sponge. Close the dish.

5. Dip a sterile cotton swab into the sterile water. Select a fomite to swab: cell phone, door knob, drinking fountain, etc. Lightly streak the swab on the region of the plate marked “fomite”.

6. Tape the two plates together. Place plates at room temperature and observe during the next laboratory.

13.4 Food Microbiology

Microorganisms have been used for centuries for food preservation and to improve or change its taste. Evidence exists that yogurt, which is milk fermented by bacteria, has been around for over 4000 years. Today, many of the foods we eat are the result of microorganisms acting on foods for a specific and desired effect. Examples of other fermented foods include wine/beer, sausage, sauerkraut/kimchee, bread, and cheese. The industrial use of microorganisms in food production began in earnest in the late 19th century, when pure cultures of bacteria were grown specifically for that purpose.

Bacteria and Health Hazards

While there are a wide number of foods that are produced with microorganisms, the introduction of harmful organisms to food presents a serious health hazard. Even bacteria that normally colonize the human digestive system (such as *E. coli*) can cause severe illness or even death if ingested. The most commonly encountered microbial pathogens are *Listeria monocytogenes, Staphylococcus aureus, Bacillus cereus, Bacillus anthracis, Clostridium botulinum, Clostridium perfringens,* members of the *Salmonella* and *Campylobacter* genus, *Escherichia coli (*especially type O157:H7), hepatitis and Rota viruses, prions, different species of tapeworms and roundworms, and protozoa. Food-borne illness can be limited to a single person who ingests the contaminated food, or can lead to a more widespread outbreak of the same illness when multiple people consumed the same contaminated product. Many outbreaks are local in nature, such as when food

from a restaurant is not held at the proper temperature, allowing bacteria to grow to disease-causing levels. Increasingly there have been food-borne illness outbreaks that affect many geographical areas due to improper food handling. On May 23, 2019 5 lb bags of Baker's Corner All purpose flour was recalled due to potential *E. Coli* contamination. The CDC reported 17 infected individuals from 8 states. https://www.cdc.gov/ecoli/2019/flour-05-19/index.html

Bacteria and Fermentation

Fermentation is important for the production of a variety of dairy products. Cheeses are typically made using lactic acid-producing bacteria that aid the coagulation of the milk protein casein in curd. The curd is then further treated with a different bacteria (depending on the desired final cheese) to produce distinctive tastes and aromas. The characteristic holes in Swiss cheese are produced by specific bacteria that generate carbon dioxide which create gas bubbles in the cheese.

Yogurt is made by the fermentation of lactose (milk sugar) by bacteria. Lactose is a disaccharide that consists of two simple sugars. During the making of yogurt, the lactose is broken down by the enzyme lactase into glucose and galactose. These sugars are then fermented generating lactic acid and acetaldehyde. These two products lower the pH of the milk giving it a sour tart taste. The lowered pH also effects the milk proteins (caseins) causing coagulation, precipitating the proteins into a solid curd that forms the yogurt. The left over watery liquid is the whey. There are a variety of bacteria that may be utilized in this process, the two most commonly used to are *Lactobacillus bulgaris and Streptococcus thermophilus*

In this experiment we will start with milk. To increase the shelf-life milk that is sold commercially is pasteurized. Pasteurization does not sterilize the milk, but will kill most of the microorganisms present. To manufacture yogurt these organisms must be destroyed so that they will not compete with the added organisms essential for the fermentation process. Commercially available yogurt containing live bacterial cultures will be used to inoculate the milk after heating.

13.5 ACTIVITY: Yogurt Preparation

1. Place beaker on hot plate. Add 300 mL of milk.

2. Measure and record the pH of the milk using pH indicator paper.

3. Insert thermometer into milk and gently heat while stirring. Bring the milk to 85 °C.

4. Remove milk from the hot plate and allow to cool to 43 °C.

5. Measure and record the pH of the cooled milk using pH indicator paper.

6. Measure 50 mL plain yogurt. (This yogurt must be marked as containing live yogurt cultures).

7. Slowly add the yogurt to the in the cooled milk, be sure to thoroughly mix the ingredients to create a homogenous solution. Adding about 30 ml (1/8 cup) nonfat dry milk at this time will increase the nutritional content of the yogurt. The yogurt will also thicken more easily.

8. Measure and record the pH of the mixture using the pH indicator paper.

9. Place the mixture into a glass jar.

10. Incubate the mixture in an incubator or water bath at 30 - 32 °C for 5 - 6 hours

11. Place the yogurt in a refrigerator for 24 hours. Measure and record the final pH using pH indicator paper.

TABLE 14.1: Yogurt pH Results	
Sample	pH
1 – before heating	
2 – after cooling	
3 – after yogurt	
4 – after incubation	

Questions for Review -

1. What is the function of heating the milk in step 3.

2. What is the purpose of adding the yogurt to the cooled milk in step 7?

3. What attributes does lactic acid confer to yogurt?

4. How does the consistency of the milk change during the production of yogurt? What facilitates this change?

5. What are the breakdown products of lactose?

6. Go to the CDC website (http://www.cdc.gov/foodsafety/outbreaks/) and describe one of the Selected Outbreak Investigations that occurred over the last 2 years.

*** STOP YOU WILL COMPLETE THE SECTION ON DISEASE TRANSMISSION NEXT LAB**

Due next lab – Homework assignment on Page 100

13.6 Disease Transmission (week 2)

Transmission of an infectious disease can occur via diverse pathways as each infectious agent has evolved to exploit a particular organism or cell type. Most microorganisms do not cause disease. In order to indicate that a disease is infectious, it must be demonstrated that the disease-causing pathogen is present in ill people but not in healthy people and that healthy people who contract the pathogen develop disease. The severity of the disease depends on the ability of the pathogen to damage the host as well as the ability of the host to resist the pathogen.

Respiratory diseases such as influenza are contracted by exposure to aerosolized droplets spread by sneezing and coughing. Gastrointestinal diseases, many of which cause diarrhea, are contracted by exposure to contaminated food or water, while sexually transmitted diseases are acquired through contact with body fluids. Some pathogens can persist on an inanimate object (common cold) and others can be transmitted via skin to skin contact (warts). Malaria, Lyme disease, West Nile, and Chagas disease are transmitted by insect vectors. Diagnostic methods used to identify disease include clinical presentation (ex. warts), microscopy, biochemical test (ex. ELISA), growth culture, and molecular diagnostics.

Epidemiologists are researchers interested in how an disease is transmitted, the source of the disease, numbers of people infected, geographical distribution, risk factors, and mortality rates. Epidemiologists also study chronic disease, such as heart disease and diabetes, illness due to chemicals and other pollutants, genetic disease, injuries, mental illness, and the risks and benefits of drugs. Epidemiology relies on biology, medicine, and biostatistics in its evidence-based approach. The American Red Cross tests all donated blood for Chagas disease, Hepatitis B, Hepatitis C, HIV, HTLV, syphilis, and West Nile virus.

13. 7 ACTIVITY: Laboratory simulation of viral infection

The test performed in the lab is a simulation. No virus, living materials, or biohazardous reagents are used. The series of test tubes are each filled with a fluid that represents body fluid one might exchange with another individual.

One tube of fluid is “positive”. You cannot identify this cup by visual inspection. You will perform an epidemiological study to determine the source of the positive fluid.

PROCEDURE

1. Your instructor will give you a cup of fluid. Record the ID (letter) of the cup.
2. During your interaction, each of you will exchange fluid by pouring some of your fluid into your partners cup, and having your partner pour some of their fluid into your cup.
3. You will be handed a behavior cards.
   a) **Step 1:** Monogamous trade with person to their right.
   b) **Step 2:** Promiscuous people stand up go to one side of the room and everyone trade with each other.
   c) **Step 3:** Cheaters stand up, go next to promiscuous, and trade with either a cheater or promiscuous.
   d) **Step 4:** One night stands stand up and trade with one other one night stand or promiscuous then sit down.
   e) **Step 5:** Promiscuous trade with each other.
   f) **Step 6:** Cheaters trade with same person from before.
   g) **Step 7**: Everyone sit down.
   h) **Step 8**: Monogamous trade once more with partner.
4. The instructor will perform add pH indicator to your tube, tubes that turn pink indicate infection.
5. Trace back the path the transmission of the disease.

13.8 Bacterial and Fungal Growth Results & Homework

Colony Morphology

A bacterial colony grows from a single bacterial cell. The colony consists of millions of bacterial cells and is visible to the human eye. The student will observe that each colony has a characteristic shape, size, texture, and color.

Procedure:
Obtain agar plates prepared in section 14.3. Leave the covers on. Observe the colonies by eye and under the dissecting microscope. Complete data table

Question: If a single bacterial cell is microscopic, how is it that these bacteria be observed with the naked eye?

Question: How does the appearance of a mold colony differ from that of bacteria?

Table 13.2

Environment	Describe Bacterial Growth	Fungi/ yes or no
Unwashed hands		
Washed hands		
Fomite		
Sponge		

Question
Did the results of this experiment support your hypothesis? **Explain** in detail.

Laboratory Exercise 13
NOTES

Homework – To be handed in Micro Lab Week #2

Some species of bacteria cause disease in humans, other animals, plants, and fungi. Perform an Internet search using the scientific name of each of the bacteria below to learn the common name of the disease. Include a brief description of the **disease not the organisms** in the space provided under each name.

Name:_____ Section:_____

Genus and species

Treponema pallidum **Name of Disease** _____
Description of disease:

Helicobacter pylori **Name of Disease** _____
Description of disease:

Borrelia burgdorferi **Name of Disease** _____
Description of disease:

Clostridium botulinum **Name of Disease** _____
Description of disease:

Yersinia pestis **Name of Disease** _____
Description of disease: