CHEMICAL PROPERTIES OF CELLS

Introduction:

In this laboratory you will study proteins, carbohydrates and lipids; common organic molecules found in cytoplasm, the fluid part of the cell. Large organic molecules often contain small building block molecules joined in sequence. As we shall see, a protein contains amino acids; starch (a carbohydrate) contains glucose; a fat or oil (lipids) contains glycerol and fatty acids. Various reagents will be used to test for the presence of these molecules. Most often we will be looking for a color change. If a color change is observed, it is said that the test is **positive** because it indicates that a particular molecule is present. If a change in color is not observed, it is said that the test is **negative** because it indicates a particular molecule is not present.

The pH of cytoplasm remains relatively constant because it is buffered. We will study the buffer phenomenon.

Exercise #1 — Benedict's Reagent & Testing for Simple Sugars

Carbohydrates are divided into three classes:

monosaccharides–a single 6-carbon molecule

disaccharides–two single sugar molecules linked together

polysaccharides–three or more sugar molecules linked together

Glucose, a monosaccharide, is only one sugar unit and is a simple sugar. Maltose, a disaccharide, is two sugar units and is also a simple sugar. Starch, a polysaccharide, is a large sugar built from a chain of glucose units.

Benedict's Reagent changes color after heating in the presence of simple sugars, but not large sugars such as starch. The table below summarizes the color changes.

Simple Sugar	Color of Benedict's Reagent
Maltose	Changes from blue to green after heating.
Glucose	Changes from blue to green to red to yellow to orange, depending on the amount of glucose present.

The following experiment will test for the presence of simple sugars in Onion Juice, Potato Juice, Glucose Solution and Maltose Solution.

Name_____

Benedict's Test Report Sheet

Materials Needed:

Test tube rack & 5 clean test tubes
Distilled Water, Onion & Potato Juice, Glucose & Maltose Solution
A dropper bottle of Benedict's Reagent
A beaker & hot plate
A centimeter ruler
A wax pencil and labeling tape

Procedure:

1. Label your test tubes 1, 2, 3, 4, 5 with labeling tape. Directly on the glass, mark with a wax pencil a line 2 cm from the bottom and another at 4 cm.

2. Fill the beaker 1/3 with tap water & heat on the hot plate to boiling before adding the test tubes.

   Tube 1: Fill to the 2 cm mark with Distilled Water and add Benedict's Reagent to the 4 cm mark.

   Tube 2: Fill to the 2 cm mark with Onion Juice and add Benedict's Reagent to the 4 cm mark.

   Tube 3: Fill to the 2 cm mark with Potato Juice and add Benedict's Reagent to the 4 cm mark.

   Tube 4: Fill to the 2 cm mark with Glucose Solution and add Benedict's Reagent to the 4 cm mark.

   Tube 5: Fill to the 2 cm mark with Maltose Solution and add Benedict's Reagent to the 4 cm mark.

3. Place all 5 test tubes in the boiling water for 5 to 10 minutes. Record any color change in the following chart.

Chart 1. Benedict's Test

Solution	Final Color of Reagent	Is a simple sugar present?
Distilled Water	_____	_____
Onion Juice	_____	_____
Potato Juice	_____	_____
Glucose Solution	_____	_____
Maltose Solution	_____	_____

Name_____

Exercise #2 — Iodine & Testing for Starch Report Sheet

Starch, a polysaccharide, is a large sugar built from a chain of glucose units. In the presence of starch, Iodine Reagent (IKI Solution) turns from a yellow-brown color to a distinct blue-black color.

Materials Needed:

Onion & potato
Razor blade
2 microscope slides & 2 coverslips
Distilled Water
Iodine Reagent (IKI)

Procedure:

Crack and peel a piece of onion skin and place it on a microscope slide. Add a drop or two of distilled water and a coverslip. Observe in the microscope at low power (100X) or high power (400X). Remove the microscope slide from the microscope and remove the coverslip. Add a drop or two of Iodine Reagent and the coverslip. Observe in the microscope at low power (100X) or high power (400X). Record any color change of the Iodine Reagent in the chart below.

Slice a very thin piece of potato with a razor blade and place it on a microscope slide. Add a drop or two of distilled water and a coverslip. Observe in the microscope at low power (100X) or high power (400X). Observe the pattern or arrangement that suggests a series of roughly circular compartments, each containing a number of oval-shaped objects. Remove the microscope slide from the microscope and remove the coverslip. Add a drop or two of Iodine Reagent and the coverslip. Observe in the microscope at low power (100X) or high power (400X). Record any color change of the Iodine Reagent in the chart below.

Chart 2. Iodine Test

Substance	Final Color of Iodine Solution	Is starch present?
Onion	_____	_____
Potato	_____	_____

Name_____

Exercise #3 — Evaporation & Testing for Lipids Report Sheet

Lipids are compounds that are insoluble in water. Lipids include fats, oils, waxes, phospholipids and cholesterol. Typically, a fat or oil is composed of one molecule of glycerol and three molecules of fatty acids.

Materials Needed:

Brown bag paper or paper towel
Distilled Water
Vegetable Oil

Procedure:

Place a couple of drops of water and on another area of the paper or towel, place a couple of drops of vegetable oil. Wait 15 to 30 minutes and then record your results in the chart below.

Chart 3. Lipid Test

Substance	Results
Water	_____
Vegetable Oil	_____

Name_____

Exercise #4 — Emulsification of Lipids Report Sheet

Emulsification is the dispersal of droplets of one liquid into another liquid. An emulsifier, such as detergent or bile salts, contains molecules with polar and nonpolar ends.When the nonpolar ends of an emulsifier are attached to the nonpolar part of a lipid, the polar ends of the emulsifier are exposed. Since the polar ends of the emulsifier are soluble in water, the lipid becomes dispersed.

Materials Needed:

Test tube rack & 2 clean test tubes
2 microscope slides & 2 coverslips
Distilled Water, Vegetable Oil, Bile Salts
Two plastic pipets
Centimeter ruler, wax pencil & labeling tape

Procedure;

1. Label your test tubes with labeling tape: 1 and 2.
2. With a wax pencil, mark test tube 1 (directly on the glass) with a line at 3 cm and 4 cm.
3. Mark test tube 2 (directly on the glass) with a line at 3 cm, 4 cm and 5 cm.
4. Fill test tube 1 to the first mark with distilled water and to the second mark with vegetable oil. Shake and observe.
5. Fill test tube 2 to the first mark with distilled water, to the second mark with vegetable oil and to the third mark with bile salts. Shake and observe.
6. Allow both test tube solutions to settle for 5 minutes.
7. Using different droppers take a sample of the solution below the oil layer in each test tube, place the drop on a microscope slide and add a coverslip. Observe each drop in the microscope at low power (100X) or high power (400X).

Discussion:

1. In test tube 1, how did the oil interact with the water before and after shaking?

2. In test tube 2, how did the oil interact with the water before and after shaking?

3. What microscopic differences did you notice in the solutions below the oil layer in each test tube?

Exercise #5 — pH & Buffer

pH refers the hydrogen ion (H^+) concentration of a solution. The pH scale ranges from 0 to 14 and indicates the concentration of hydrogen ions (H^+) and hydroxide ions (OH^-). A pH of 0 to 6.9 indicates the solution is acidic and has more hydrogen ions (H^+) than hydroxide ions (OH^-).

A pH of 7.1 to 14 indicates that the solution is basic and has more hydroxide ions (OH^-) than hydrogen ions (H^+).

A pH of 7 indicates the solution is neutral and the concentration of hydrogen ions (H^+) is equal to the concentration of hydroxide ions (OH^-). A buffer is a substance that keeps the pH of a solution constant by removing either excess hydrogen ions or hydroxide ions.

A common example of monitoring pH is seen in our fish aquariums and swimming pools. When we test the pH of our aquariums, we are checking to make sure that the environment is healthy for the fish. Conversely, in our swimming pools we are trying to keep the pH at a point that will not allow the growth of microorganisms. Living cells and organisms are buffered to maintain optimum pH conditions. In humans, the optimum pH of the blood is 7.4. Our bodies are so sensitive to changes in the hydrogen ion concentration that a blood pH change from normal (7.4) to 7.2 or 7.6 can result in death.

Materials Needed:

Test tube rack & 3 clean test tubes
Distilled water, pH 7 Buffer Solution & Cytoplasm
A dropper bottle of dilute Hydrochloric Acid (HCl) Solution
pH paper
Wax pencil & labeling tape

Procedure:

1. Label your test tubes with labeling tape or a wax pencil: 1, 2 and 3.

2. Fill each test tube 1/3 full with the solution indicated below.

Test Tube 1	Distilled water
Test Tube 2	pH 7 Buffer Solution
Test Tube 3	Cytoplasm

3. Test the pH of the solution in each test tube with pH paper or litmus paper by taking approximately 5 cm of the paper and dipping the end into the solution. Record your results in the chart below.

   The pH paper container has a color code for determining the pH.

Name_____

pH & Buffer Report Sheet

(Procedure cont.)

4. Add 10 drops of hydrochloric acid (HCl) solution to each test tube. Shake or swirl the test tube.

5. Test the pH of each test tube solution again, as described above. Record your results in the chart below.

Chart 4. pH & Buffer Test

	Test Tube Contents	pH before Acid	pH after Acid
A.	_____	_____	_____
B.	_____	_____	_____
C.	_____	_____	_____

1. In which test tube(s) solution did the pH change to acidic after the addition of hydrochloric acid (HCl) solution?

2. In which test tube(s) solution did the pH not change after the addition of hydrochloric acid (HCl) solution?

3. Why would the pH of a solution not change when hydrochloric acid (HCl) solution is added?