

DIFFUSION AND OSMOSIS

NAME _____ DATE _____

INTRODUCTION

The life of a cell is dependent on efficiently moving material into and out of the cell across the cell membrane. Raw materials — such as oxygen and sugars — needed for the production of energy and other cellular products must enter the cell. Whereas waste products — such as carbon dioxide and ammonia — which are generated during energy production and cellular digestion must be removed from the cell. Most of these materials move passively — costing the cell no energy — through the process of **diffusion**.

Diffusion is the movement of molecules from an area of higher concentration of those molecules to an area of lower concentration. This difference in concentration is called a **concentration gradient**. Diffusion happens because of the kinetic energy of the particles. A good metaphor for this molecular motion is what happens if you were to open a bottle of hydrogen sulphide gas (H_2S smells like rotten eggs) in one corner of a room. It would not be long before someone in the opposite corner of the room would smell the odour of rotten eggs. The molecules appear to be moving from an area of higher concentration of H_2S gas (the bottle) to an area of lower concentration of H_2S gas (the opposite corner of the room). Eventually a dynamic equilibrium is reached; the concentration of H_2S gas will be approximately equal throughout the room and no **net** movement of H_2S will occur from one area to the other. The rate of this diffusion will depend on 1) the concentration gradient, 2) temperature and 3) surface area to volume ratio.

Since all life takes place in water — either external waters or internal waters — we must also address the special case of the movement of water across cellular membranes. The diffusion of water through a selectively permeable membrane is referred to as **osmosis**. Water moves from a region of higher concentration of water to a region of lower water concentration. This is often also stated as movement from a region of higher **water potential** to a region of lower water potential. Distilled water (pure water) has the highest concentration of water or the highest water potential.

However, diffusion and osmosis do not entirely explain the movement of ions and molecules into and out of the cell. Some molecules are valuable enough to the cell to expend energy transporting them across the cell membrane. This **active transport** uses energy from ATP to move substances through the cell membrane. Active transport usually moves substances against a concentration gradient, from regions of low concentration of that substance into regions of higher concentration.

A. DIFFUSION

In this experiment you will measure the diffusion of small molecules through dialysis tubing, an example of a selectively permeable membrane. Small dissolved molecules (solutes) and water molecules can move freely through a selectively permeable membrane, but larger molecules will pass through more slowly, or perhaps not at all. The size of the minute pores in the dialysis tubing determines which substances can pass through the membrane. We will explore the process of diffusion through a semi-permeable membrane in this activity.

PROCEDURE

1. Obtain a 30cm piece of 2.5cm dialysis tubing that has been soaking in water. Tie off one end of the tubing to form a bag. To open the other end, rub the end between your fingers until the edges separate.
2. Obtain a sample of the 15% glucose/1% starch solution. Test the solution for the presence of **glucose** using Benedict's solution.
 - a. Put 30 drops of solution in a test tube.
 - b. Add 10 drops of Benedict's Solution to the test tube.
 - c. Use the test tube clamp to place the test tube in the hot water bath for 3 minute.
 - d. Let the test tube cool.
 - e. Observe the colour of the solution and record. (yellow is the presences of glucose)
3. Record the results in Table 1
4. Obtain a second sample of the 15% glucose/1% starch solution. Test the solution for the presence of **starch** using Lugol's iodine (IKI).
 - a. Put 30 drops of solution in a test tube
 - b. Add 5 drops of Lugol's iodine solution to the test tube
 - c. Observe the colour of the solution and record (black is the presence of starch)
5. Record the results in Table 1.
6. Place 15mL of the 15% glucose/1% starch solution in the bag (**bag**). Tie off the other end of the bag, leaving sufficient space for expansion of the contents in the bag. About 2 cm. Record the colour of the solution in Table 2.
7. Fill a 250mL beaker two-thirds full of water. Add approximately 4mL of Lugol's solution to the water and record the colour of the solution in Table 2 (**beaker**). This also served as a test of the water for the presence of **starch**. Record the results in Table 2.
8. Take a sample of the water and Lugol's solution from the beaker. Test the solution for the presence of **glucose**. Record the results in Table 2.
9. Immerse the dialysis bag in the beaker of IKI solution.
10. Allow the set up to stand overnight.
11. Test the liquid in the beaker and in the bag for the presence of **glucose and iodine**. Record the results in Table 2.
12. Complete the Summary Questions for this section of the lab.

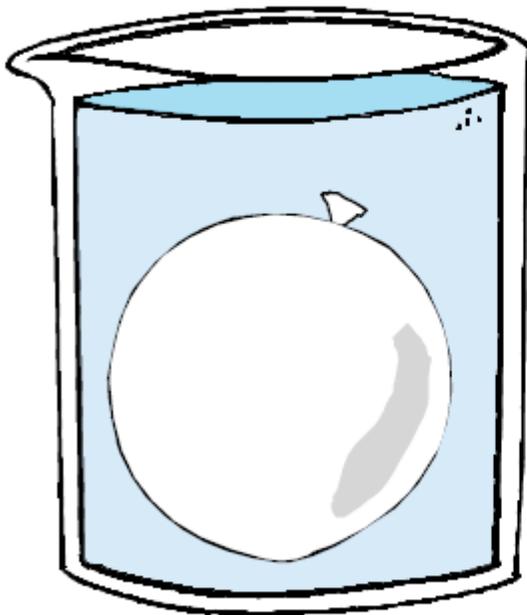
TABLE 1

	Colour of Solution	
	Initial	Final
15% glucose 1% starch		
H ₂ O & IKI		

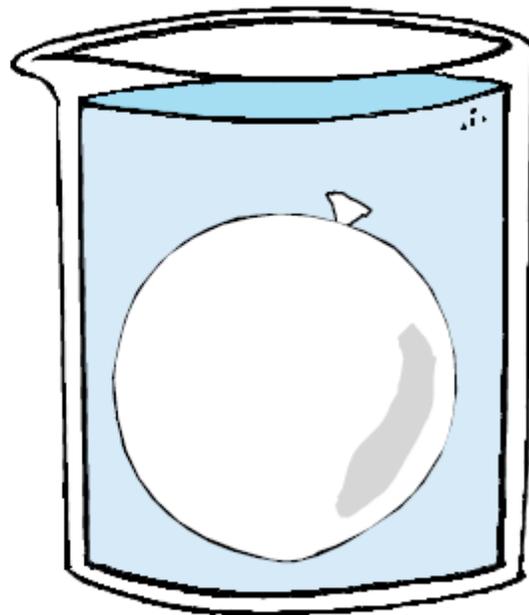
TABLE 2

	Initial Contents	Colour of Solution		Presence of Glucose		Presence of Starch	
		Initial	Final	Initial	Final	Initial	Final
Bag	15% glucose 1% starch						
Beaker	H ₂ O & IKI						

Use the diagram below to illustrate the contents of the bag and the beaker as well as the movement of molecules during the experiment



INITIAL STATE



FINAL STATE

B. OSMOSIS

Using the principles you learned in the first exercise, we will now investigate the movement of water in and out of an actual cell rather than merely a model. Water moves from areas of higher concentration of water to a region of lower water concentration. Water is at its highest concentration as distilled water. The concentration of water decreases as solutes are dissolved in the water.

If a model cell (like our dialysis tubing) is filled with a salt solution and placed in a beaker of distilled water, water will diffuse into the cell until a dynamic equilibrium is reached. At that point, no **net** movement of water will occur between the cell and the beaker. If a model cell is filled with distilled water and placed in a beaker of salt solution, water will diffuse out of the cell until a dynamic equilibrium is reached. Once again, at that point, no **net** movement of water will occur between the cell and the beaker.

Let us now use these principles to investigate the movement in and out of plant cells using potato.

PROCEDURE

1. Prepare 1 litre of each of the following sucrose solutions:

0.0M 0.2M 0.4M 0.6M 0.8M 1.0M

2. Pour 100mL of each solution into a labelled 250mL beaker.
3. Slice a potato into discs that are approximately 5cm thick. Use a cork borer to cut four potato cores for each sugar solution beaker. Do not include any skin on any of the potato cores.
4. Determine the mass of the four potato cores together. If there is any wait time for a balance, keep your potato cores in a covered beaker until it is your turn. Record the mass of each set of potato cores in Table 3.
5. Put a set of four cores into the appropriate beaker of sucrose solution. Cover each beaker with plastic wrap to prevent evaporation. Let stand overnight.
6. Remove the cores from each beaker, blot them gently on a paper towel, and determine the mass of each set of four cores. Record the final mass in Table 3. Calculate the percent change in mass for each sucrose solution and record in Table 3.

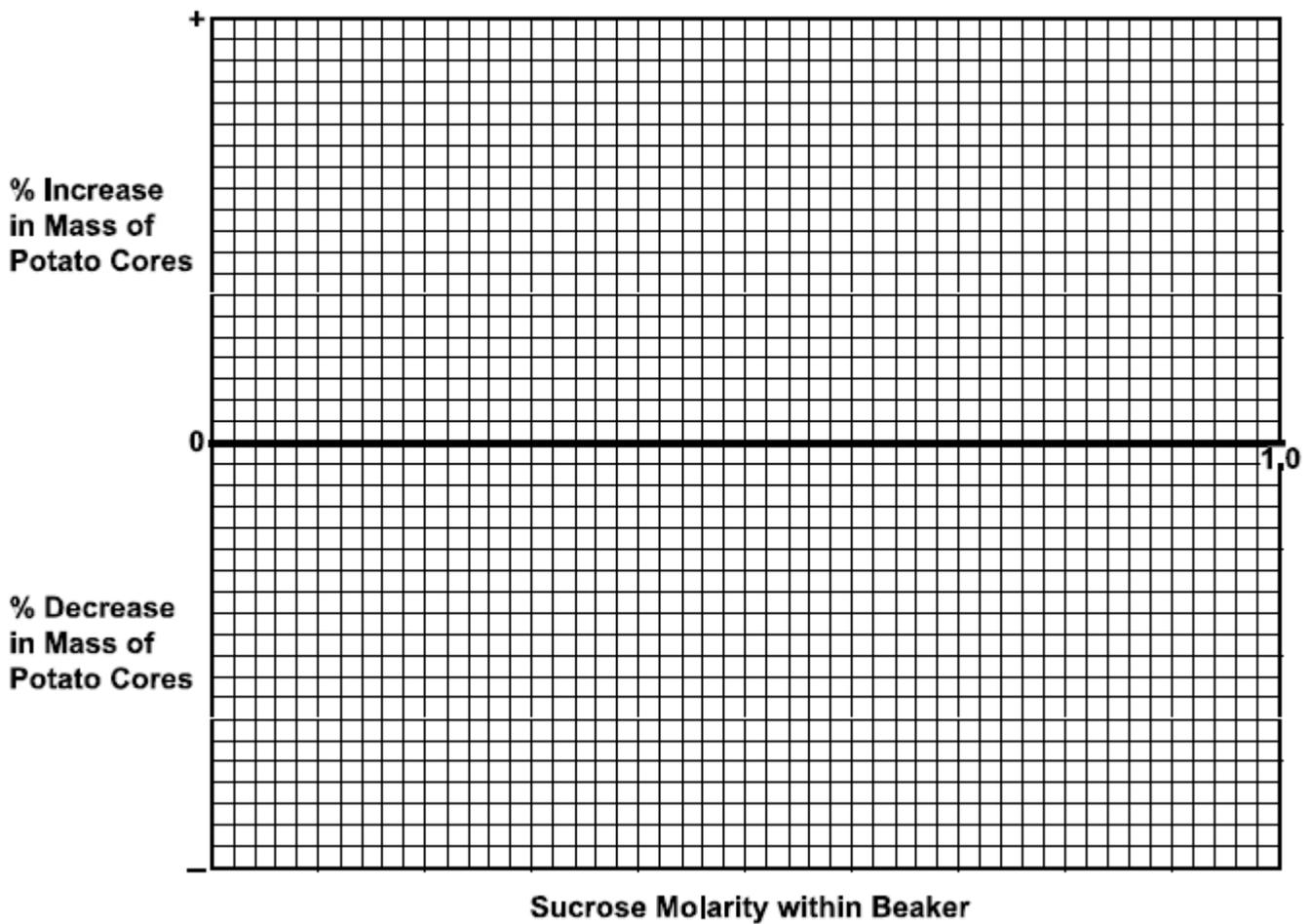
$$\text{Percent Change in Mass} = \frac{\text{Final Mass} - \text{Initial Mass}}{\text{Initial Mass}} \times 100$$

7. Average the class data.
8. Graph both your individual results and the class average for the percentage change in mass.
9. Complete the Summary Questions for this section of the lab.

TABLE 1 # POTATO CORE

Contents in Beaker	Initial Mass	Final Mass	Mass Difference	Percent Change In Mass
0.0 M water				
0.2M sucrose				
0.4M sucrose				
0.6M sucrose				
0.8M sucrose				
1.0M sucrose				

Graph your data



Explain your results.

C. PLANT CELL PLASMOLYSIS

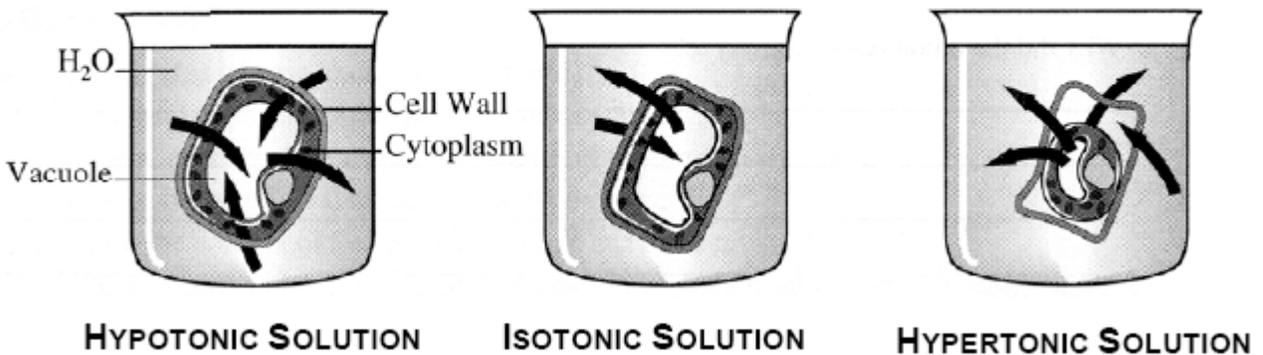
Cells lose or gain water due to the difference in solute concentrations between the cytoplasm (the intracellular fluid) and the solution surrounding the cell (the extracellular fluid). The movement of water in and out of a cell is governed by the laws of diffusion: water flows from a region of higher water concentration to a region of lower concentration. When a cell is in a **hypertonic** solution, it will experience a net loss of water. A hypertonic solution contains a higher concentration of solutes than the cell and therefore a lower concentration of water. Consequently, water will flow **out of** the cell from the region of higher water concentration to the region of lower concentration.

When a cell is in a **hypotonic** solution, it will experience a net gain of water. A hypotonic solution contains a lower concentration of solutes than the cell and therefore a higher concentration of water. Consequently, water will flow **into** the cell from the region of higher water concentration to the region of lower concentration.

When a cell is in an **isotonic** solution, it will experience neither a net gain or loss of water. An isotonic solution contains an equal concentration of solutes as the cell and therefore an equal concentration of water.

Consequently, water will flow equally **into and out of** the cell. **Plasmolysis** is the shrinking of the cytoplasm of a plant cell in response to diffusion of water out of the cell and into a hypertonic solution surrounding the cell as shown in Figure 2. During plasmolysis the cell membrane pulls away from the cell wall. In this lab exercise, you will examine this process by observing the effects of a highly concentrated salt solution on plant cells

Figure 2.



CONCLUSIONS

A. DIFFUSION

Analyse your data and write an analysis of your conclusion. This should be a minimum of two paragraphs.

Items to consider.

- a) What experimental evidence supports the movement of particles?
- b) concentration differences and membrane pore size,
- c) other molecule can we assume also moved across the membrane?
- d) compare the size of each of the following molecules with the membrane pore size,
- e) experimental error and
- f) summarize the process of diffusion

In many animals, glucose, rather than starch, is transported by the blood through the body to all cells. In the digestive system, starches are digested by amylase to yield glucose. Based on the findings of this lab, explain why the digestion of starch to glucose is necessary.

B. OSMOSIS

Analyse your data and write an analysis of your conclusion. This should be a minimum of two paragraphs. Items to consider.

- a) What experimental evidence supports the movement of particles?
- b) concentration differences and membrane pore size,
- c) other molecule can we assume also moved across the membrane?
- d) compare the size of each of the following molecules with the membrane pore size,
- e) experimental error and
- f) summarize the process of osmosis

Why did you calculate percent (%) change in mass of the potato cores rather than use the change in mass directly?

What is plasmolysis?

.Draw a molecular diagram of the cell membrane. Illustrate the following processes and the cellular structures that are involved:

a. diffusion of water molecules across the cell membrane

b. diffusion of polar molecules (i.e., sodium ion) across the cell membrane

c. diffusion of non-polar molecules (i.e., lipid hormone) across the cell membrane