Measuring Osmotic Potential

INTRODUCTION

All cells require essential materials to ensure their survival. Chemical, physical, and biological processes are used to move these materials inside of cells. Similar processes move waste materials outside of cells. These processes can be passive, occurring as a result of basic physical laws and requiring no outside energy from the cell or they can be active, requiring energy expenditure. Since all molecules possess kinetic energy (energy of motion), they are constantly moving enabling the passive transfer of certain substances into and out of cells.

Diffusion is the passive movement of molecules from an area of **higher concentration** to areas of **lower concentration**. In a closed environment, molecules will disperse until they reach a state a dynamic equilibrium. As the molecules approach equilibrium, the net or overall rate of diffusion begins to slow and occurs equally in all directions. In an open environment, there are no "walls" to confine the molecules so the molecules will always appear to move away from the immediate source. For instance, odors in a confined room persist much longer, whereas odors outside will dissipate more quickly. Molecular weight indirectly affects the rate of diffusion. When a concentration gradient (difference in concentration) exists, the net effect of this random molecular movement is that the molecules eventually become evenly distributed throughout the environment. There are many examples of diffusion in non-living systems – for example, the ability to smell a friend’s cologne shortly after he or she has entered the room. It can also be seen if you drip food coloring or ink into a clear glass of water. Water will let other molecules move among the water molecules so freely that the water carries or transports them.

The diffusion of particles into and out of cells is regulated by the **plasma membrane**, which constitutes a physical barrier. In general, molecules diffuse passively through the plasma membrane if they can dissolve in the lipid portion of the membrane (as in the case of CO_2 and O_2). The diffusion of solutes (particles dissolved in water) through a **semi-permeable membrane** is called **simple diffusion**. The diffusion of water through a semi-permeable membrane is called **osmosis**. Both simple diffusion and osmosis involve the movement of a substance from an area of its higher concentration to one of lower concentration – down the **concentration gradient**. Certain molecules, for example glucose, and ions move through the membrane by a passive transport process called **facilitated diffusion**. The transported substance either binds to protein carriers in the membrane and is ferried across or moves through water-filled protein channels. As with simple diffusion, the substance moves down the concentration gradient.

Osmosis occurs where a semi-permeable membrane (a membrane through which water can pass but some other particles cannot) separates two bodies of fluid. Plant and animal cell membranes are semi-permeable. As long as the number of particles or the concentration of ions in both of the cell compartments remains equal, osmosis helps to maintain that equalized state in each compartment. If particles are added to one compartment only, then the concentration is increased. The process of osmosis must be tightly controlled by cells, otherwise they will die. For example, if you place a red blood cell in pure (distilled) water, it will quickly take up water until it bursts. That is why plasma, the liquid portion of our blood is made of water with proteins and salts dissolved in it, preventing the unnecessary gain of water by our blood cells. In plants, osmosis is just as important. Plants with too little water will wilt. This happens when water moves out of the cells by osmosis. Without this water, there is little pressure inside the cells and the plant can no longer support itself against the pull of gravity. However, after watering the plant, the cells become re-inflated with water and the plant stands upright.

Because many solutes cannot easily pass through these membranes, water shifts from the compartment of low solute concentration to the compartment of high solute concentration. Osmosis is the simple (passive) diffusion of water across a semi-permeable membrane. Water flows from an area of high water concentration (**high water potential**) to areas of lower water concentration (**low water potential**). Water will always move from an area of higher water potential (higher free energy; more water molecules) to an area of lower water potential (lower free energy; fewer water molecules). Water potential, measures the tendency of water to leave one place in favor of another place. You can picture the water diffusing 'down' a **water potential gradient**.

When two solutions have the same concentration of solutes, they are said to be **isotonic** to each other. If the two solutions are separated by a selectively permeable membrane, water will move between the two solutions by osmosis, but there will be no net change in the amount of water in either solution. If two solutions differ in the concentration of solutes that each has, the one with more solute is **hypertonic** to the one with less solute. The solution that has less solute is **hypotonic** to the one with more solute. These words are used to compare solutions. Now consider two solutions separated by a selectively permeable membrane. The solution that is hypertonic to the other must have more solute and therefore less water. The water potential of the hypertonic solution is less than the water potential of the hypotonic solution, so the net movement of water will be from the hypotonic solution into the hypertonic solution.

Human Physiology Connection:

The epidermis, or outer layer of the skin, is made up of cells called keratinocytes, which form a very strong intracellular skeleton made up of a protein called keratin. These cells divide rapidly at the bottom of epidermis, pushing the higher cells upward. After migrating about halfway from the bottom of this layer to the top, the cells undergo a programmed death. The nucleus involutes, leaving alternating layers of the cell membrane, made of lipids, and the inside, made largely of water-loving keratin. The outer layer of the epidermis, called the stratum corneum is composed of these alternating bands. When hands are soaked in water, the keratin absorbs it and swells. The inside of the fingers does not swell. As a result, there is relatively too much stratum corneum and it wrinkles. This bunching up occurs on fingers and toes because the epidermis is much thicker on the hands and feet than elsewhere on the body. Soaking in the tub does hydrate the skin, but only briefly. All the added water quickly evaporates, leaving the skin dryer than before. The oils that hold the water in have usually been stripped out by the bath especially if soap and hot water are involved. But if oil is added before the skin dries, much of the absorbed water is retained. Applying a bath oil or heavy lotion directly after a bath or shower is a good method of hydrating the skin.

MATERIALS

1 M NaCl solution
DI water
100 ml graduated cylinder
paper cups
electronic balance
cork borer
knife
root vegetable (sweet potato, white potato, red potato, yucca)
Pasteur pipette
watch glass
1 mL pipette
ruler
scissors
clock

PROCEDURE

Osmotic Potential in Different Plant Root Tissues

Water potential is a measure of the energy state of water. The contribution to water potential by dissolved solutes, termed osmotic potential is always negative in sign. In other words, solutes decrease the water potential. In this lab we will use the **Gravimetric** to determine the water potential of different plant cells (sweet potato, white or red potato, yucca root).

The Gravimetric technique for measuring water potential is simple to perform and doesn't require expensive equipment. Tissue samples are incubated in a series of solutions of known osmotic (water) potential and mass changes in tissues are noted Gravimetric. One distinct advantage of this technique is that it provides a very accurate estimate of water potential. In this method, tissue samples are weighed before and after incubation in a series of solutions of known osmotic (water) potential. Then, the percent change in weight of the tissue is plotted versus solution concentration (or osmotic potential).

The water potential of the tissue is considered to equal the osmotic potential of the incubating solution at which there is no change in tissue weight (***i.e.*, where the curve intercepts the x-axis**).

1. Each group will measure the movement of water into or out of potato samples.

2. Groups must prepare a series of molar concentrations of NaCl of 50 mL volume. Stock solution of 1M is provided.

To calculate the needed amount of solute, use the relationship C1V1= C2V2:

Example: Stock solution is 1M NaCl. Desired concentration is 0.75 M NaCl:

C1= 1M NaCl V1= (X mL); C2= 0.75M NaCl V2= 50 mL

(1M) (X) = (0.75M) (50mL)

X = 37.5 mL of 1M stock
Add 12.5 mL of water for a final volume of 50 mL

Final Concentration	Amount of stock added	Amount of water added
0.0M	0 mL	50 mL
0.25		
0.5		
0.75	37.5 mL	12.5 mL
1.0M	50 mL	0 mL

4. Using the cork borer, make a cylinder from the potato samples. Trim them with a knife until you have three cores that are 1 cm long each. Make sure there is no peel left on the core. Use the knife to make three 1 cm long slices of each potato sample. Make sure that samples are identical in size and shape with no peel.

5. Determine the mass of all three of the cores for the same plant together. You should mass them to the nearest 0.01 g. Record your data in the data table.

6. Place the cores in the correct paper cups – the three cores of each plant together. Set them aside for 45 minutes.

Russett POTATO				
NaCl Molarity	Initial Mass of 3 cores (g)	Final mass of 3 cores (g)	Change in mass (final – initial) (g)	% change in mass
0				
0.25				
0.5				
0.75				
1.0				

TABLE 10-1

Sweet POTATO				
NaCl Molarity	Initial Mass of 3 cores (g)	Final mass of 3 cores (g)	Change in mass (final – initial) (g)	% change in mass
0				
0.25				
0.5				
0.75				
1.0				

TABLE 10-2

Red Potato				
NaCl Molarity	Initial Mass of 3 slices (g)	Final mass of 3 slices (g)	Change in mass (final – initial) (g)	% change in mass
0				
0.25				
0.5				
0.75				
1.0				

TABLE 10-3

Yucca				
NaCl Molarity	Initial Mass of 3 slices (g)	Final mass of 3 slices (g)	Change in mass (final – initial) (g)	% change in mass
0				
0.25				
0.5				
0.75				
1.0				

TABLE 10-4

7. After 45 minutes, remove each set of three cores from their containers. Briefly blot them with a paper towel to remove excess water. Find the mass of each group of three and record the mass in the data table.

8. Make observations of the texture, color and flexibility of the cores. Record these observations.

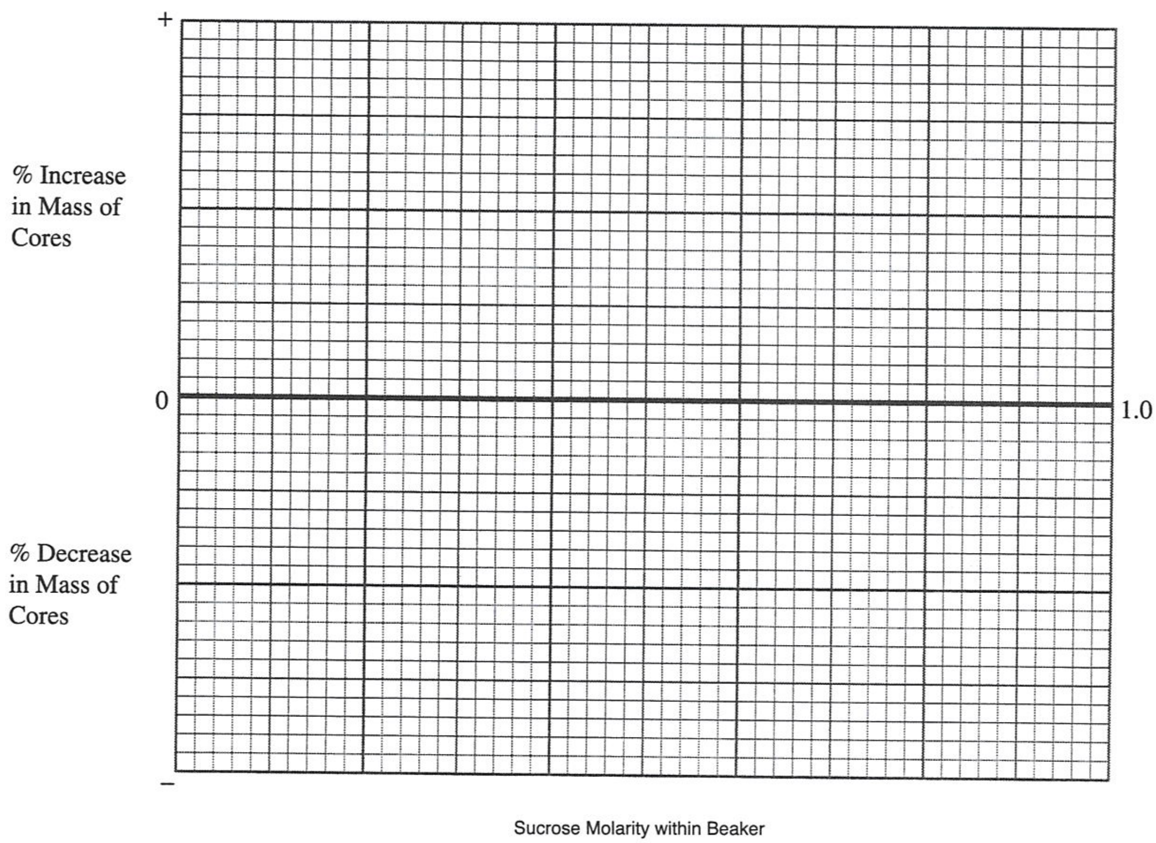
9. Determine the change in mass by subtracting. If the mass increased, place a (+) in the data table next to the amount changed. If the mass decreased, place a (-) in the space.

10. Share the data with your classmates to complete the tables.

11. Plot percent change in mass vs. NaCl concentration (molarity) for each of the four tissue samples. Draw the best fit line for each data set.

12. From the graphs, determine the concentration of the sucrose solution in which there was no net weight gain (*i.e.*, % change = 0). At this point, the water potential of the solution equals the water potential of the cores. An alternate method to determine this point requires performing a regression analysis of the best fit line of your data. The equation for this line is in the form, y = mx + b. Substitute in this equation, y = O, and then solve for x (the point at which the line crosses the x axis and equals the sucrose concentration in which there is no net change in weight of the cores = water potential of the cores).

The overall look of your graph setup should look something like this:



What sucrose concentration balances the osmotic potential of each plant tissue?

FIGURE 10-1

Plant	Sucrose Concentration (M)
Russett	
Sweet	
Red	
Yucca	

TABLE 10-5

Do the different plant tissues show different values in this table?