Osmosis

In order to survive, all organisms need to move molecules in and out of their cells. Molecules such as gases (e.g., O₂, CO₂), water, food, and wastes pass across the cell membrane. There are two ways that the molecules move through the membrane: *passive transport* and *active transport*. While active transport requires that the cell uses chemical energy to move substances through the cell membrane, passive transport does not require such energy expenditures. Passive transport occurs spontaneously, using heat energy from the cell's environment.

Diffusion is the movement of molecules by passive transport from a region in which they are highly concentrated to a region in which they are less concentrated. Diffusion continues until the molecules are randomly distributed throughout the system. Osmosis, the movement of water across a membrane, is a special case of diffusion. Water molecules are small and can easily pass through the membrane. Other molecules, such as proteins, DNA, RNA, and sugars are too large to diffuse through the cell membrane. The membrane is said to be semipermeable, since it allows some molecules to diffuse though but not others.

If the concentration of water on one side of the membrane is different than on the other side, water will move through the membrane seeking to equalize the concentration of water on both sides. When water concentration outside a cell is greater than inside, the water moves into the cell faster than it leaves, and the cell swells. The cell membrane acts somewhat like a balloon. If too much water enters the cell, the cell can burst, killing the cell. Cells usually have some mechanism for preventing too much water from entering, such as pumping excess water out of the cell or making a tough outer coat that will not rupture. When the concentration of water inside of a cell is greater than outside, water moves out of the cell faster than it enters, and the cell shrinks. If a cell becomes too dehydrated, it may not be able to survive. Under ideal conditions, the water concentration outside is nearly identical to that inside.

In this experiment, you will use a Gas Pressure Sensor to measure the rate of pressure change as water moves in to or out of the cell (dialysis tubes filled with various concentrations of syrup solution). The pressure generated is called osmotic pressure and is in response to the overall movement of molecules, both water and syrup, inside the dialysis cell.

OBJECTIVES

In this experiment, you will

- Use a Gas Pressure Sensor to investigate the relationship between water movement and solute concentration.
- Determine the water potential of potato cells.



Figure 1

MATERIALS

computer Vernier computer interface Logger *Pro* Vernier Gas Pressure Sensor four 16 x 100 mm test tubes 20 cm piece of plastic tubing test tube rack 25 mL graduated cylinder various concentrations of syrup solutions dialysis tubing, 2.5 cm x 15 cm dialysis tubing clamp or floss utility clamp ring stand plastic tubing clamp plastic tubing with Luer connector 20 oz. Styrofoam cup or 600 mL beaker warm water bath plastic syringe

PROCEDURE

- 1. Place four test tubes in a rack and label them 90%, 80%, 70%, and 60%.
- 2. Fill each test tube with 15 mL of the corresponding syrup solution.
- 3. Connect the plastic tubing with the Luer connector to the valve on the Gas Pressure Sensor.
- 4. Connect the Gas Pressure Sensor to the computer interface. Prepare the computer for data collection by opening the file "22 Osmosis" from the *Biology with Vernier* folder of Logger *Pro*.
- 5. Using a ring stand and clamp, mount the pressure sensor above the insulated water container, as in Figure 1. Note: Be careful not to get any solution on the sensor.
- 6. Obtain a piece of wet dialysis tubing and dialysis tubing clamp and clamp off one end as shown in Figure 2.
- 7. Prepare the dialysis tubing.
 - a. Connect the 20 cm segment of plastic tubing to the syringe and draw up the contents of the 90% syrup solution.
 - b. Rub the open end of the dialysis tubing between your thumb and forefinger to create an opening.
 - c. Insert the end of the plastic tube into the dialysis tubing and carefully inject the syrup solution as shown in Figure 3.
 - d. Rinse the syringe and tubing with water and set it aside.
- 8. Complete the experimental setup.
 - a. Slide a plastic tubing clamp on to the plastic tubing connected to the Gas Pressure Sensor.
 - b. Insert the end of the tubing into the top of the dialysis tubing filled with the syrup solution so that it is about 1 cm above the liquid.
 - c. Wrap the dialysis tubing tightly around the plastic tube.
 - d. Slide the clamp over the end of the wrapped dialysis tubing and squeeze the tubing clamp shut as shown in Figure 4, leaving minimal airspace above the liquid.









Figure 4

- 9. Fill the insulated water container with 600 mL of 37° C water and place the clamped dialysis tube in the container. Check that the dialysis tubing holding the solution is completely submerged and without kinks. Wait five minutes.
- 10. After 5 minutes click **Collect** to begin data collection.
- 11. When data collection is finished, carefully release the tubing clamp by pushing up on one side of the clamp while pushing down on the other. **Important:** Do not disconnect the tubing from the sensor prior to releasing the tubing clamp. There will be enough pressure within the dialysis tubing to create a mess.
- 12. Rinse out the dialysis tubing and beaker.
- 13. Determine the rate of change in osmotic pressure.
 - a. Move the mouse pointer to the point where the data values begin to increase. Hold down the left mouse button. Drag the mouse pointer to the end of the data and release the mouse button.
 - b. Click the Linear Fit button, k, to perform a linear regression. A floating box will appear with the formula for a best fit line.
 - c. Record the slope of the line, m, as the rate of change in osmotic pressure for a 90% syrup solution.
 - d. Close the linear regression floating box.
- 14. Store the data by choosing Store Latest Run from the Experiment menu.
- 15. Repeat Steps 6–14 for the remaining syrup solutions.

DATA

Table 1						
Syrup Concentration	Rate of pressure change (kPa/min)					
90%						
80%						
70%						
60%						

Table 2					
Syrup solution concentration	Class average rate of pressure change (kPa/min)				
90%					
80%					
70%					
60%					

PROCESSING THE DATA

1. On Page 2 of the experiment file, create a graph with the rate of pressure change (slope) on the y-axis and syrup concentration on the x-axis.

QUESTIONS

- 1. Which solutions, if any, produced a positive slope? Was water moving in or out of the cell (dialysis tubing) under these circumstances? Explain.
- 2. Which solutions, if any, produced a negative slope? Was water moving in or out of the cell under these circumstances? Explain.
- 3. Does syrup move in or out of the cell? Explain.
- 4. Examine the graph of the rate of pressure change *vs*. the syrup concentration. Describe any pattern in the data.
- 5. Use the graph to estimate the concentration of syrup that would yield no change in pressure. Why is this biologically significant?
- 6. When wilted plants are watered, they tend to become rigid. Explain how this might happen.
- 7. Explain the strengths and weakness of this dialysis model with respect to an animal and plant cell.
- 8. Discuss and explain potential reasons to account for variation in class average pressure change at specific syrup concentrations.
- 9. This exercise was carried out at initial temperatures of 37°C. Predict the effect of increased and decreased initial temperature on rates of pressure change for each of these different solution concentrations.
- 10. Predict the rates of pressure change if the dialysis tubing is placed in an insulated cup holding 1000 mL of 37°C water. Explain your reasoning.

11. Predict the rate of pressure change if the dialyisis tubing were filled with 10 mL of 80% syrup solution and 5 mL of 1.0 M sodium chloride solution. Explain your reasoning.

EXTENSION – WATER POTENTIAL

Water potential is a term used when predicting the movement of water into or out of plant cells. Water always moves from an area of higher water potential to an area of lower water potential. The symbol for water potential is the Greek letter Psi, Ψ . Water potential consists of a physical pressure component called pressure potential Ψ_p , and the effects of solutes called solute potential, Ψ_s .

Ψ	=	Ψ_{p}	+	$\Psi_{\rm S}$
Water	=	Pressure	+	Solute
potential		potential		potential

Distilled water in an open beaker has a water potential of zero. The addition of solute decreases water potential while the addition of pressure increases water potential. A water potential value can be positive, negative, or zero. Water potential is usually measured in bars, a metric measure of pressure. (1 kPa = .1 bar)

In this experiment, you will measure the percent change in mass of potato cores after they have soaked in various concentrations of sugar solutions for a 24 hour period. You will use this data to calculate the water potential of the potato cells.

MATERIALS

computer	250 mL beaker
Logger Pro	plastic wrap
four potato cores	balance
0 M, 0.33 M, 0.67 M, or 1.0 M sugar	paper towels
solution	

PROCEDURE

You will be assigned one or more of the sugar solutions in which to soak your potato cores.

- 1. Pour 100 mL of the assigned sugar solution into a 250 mL beaker.
- 2. Measure and record the mass of the four potato cores together.
- 3. Put the four cores into the beaker of sugar solution.
- 4. Cover the beaker with plastic wrap and allow it to stand for a 24-hour period.
- 5. Remove the cores from the beaker, blot with a paper towel, and determine the mass of the four cores together after soaking.

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- 6. Calculate the percent change in mass and record your data for the sugar concentration tested in Table 2 as well as on the class data sheet.
- 7. Repeat Steps 1–6 for any additional assigned sugar solutions.

DATA

Table 2							
Sugar solution concentration	Initial mass (g)	Final mass (g)	Percent change in mass				
0.00 M							
0.33 M							
0.67 M							
1.0 M							

Table 3 Class data of percent change in mass										
Sugar solution concentration	Group 1	Group 2	Group 3	Group 4	Group 5	Group 6	Group 7	Group 8	Total	Class average
0.00 M										
0.33 M										
0.67 M										
1.00 M										

Table 4				
Sugar molar concentration	=			
Solute potential	=			
Water potential	=			

PROCESSING THE DATA

- 1. On Page 3 of the experiment file, create a graph with the percent change in mass on the yaxis and concentration of sugar on the x-axis.
- 2. Perform a linear regression to determine the molar concentration of sugar solution at which the mass of the potato cores does not change.
 - a. Move the mouse pointer to the first data point. Press the left mouse button. Drag the pointer to the last data point.
 - b. Click the Linear Fit button, 🛵, to perform a linear regression. A floating box will appear with the formula for a best fit line.
 - c. Choose Interpolate from the Analyze menu. Move the mouse pointer along the regression line to the point where the line crosses the x axis. This point represents the molar concentration of sugar with a water potential equal to the potato core water potential. Record this concentration in Table 4.
- 3. Use the sugar molar concentration from the previous step to calculate the solute potential of the sugar solution.
 - a. Calculate the solute potential of the sugar solution using the equation

$$\Psi_s = -iCRT$$

where i = ionization constant (1.0 for sugar since it doesn't ionize in water)

C = sugar molar concentration (determined from the graph)

R = pressure constant (R = 0.0831 liter bars/mol-K)

T = temperature (K)

- b. Record this value with units in Table 4.
- 4. Calculate the water potential of the solution using the equation

$$\Psi = \Psi_p + \Psi_s$$

The pressure potential of the solution in this case is zero because the solution is at equilibrium. Record the water potential value with units in Table 4.

QUESTIONS

- 1. What may happen to an animal cell if water moves into it? How does this differ from what would happen in a plant cell?
- 2. What factors affect water potential?
- 3. If a plant cell has a higher water potential than its surrounding environment and the pressure is equal to zero, will water move in or out of the cell? Explain why.