Diffusion through Membranes

Diffusion is a process that allows ions or molecules to move from where they are more concentrated to where they are less concentrated. This process accounts for the movement of many small molecules across a cell membrane. Diffusion allows cells to acquire food and exchange waste products. Oxygen, for instance, might diffuse in pond water for use by fish and other aquatic animals. When animals use oxygen, more oxygen will diffuse to replace it from the neighboring environment. Waste products released by aquatic animals are diluted by diffusion and dispersed throughout the pond.

It is important to consider how the rate of diffusion of particles may be affected or altered.

- Diffusion may be affected by how steep the concentration gradient is. The direction that a diffusing molecule or ion might travel is random. While the particles are diffusing, is there a net movement from where they are concentrated to where they are less concentrated?

- Diffusion may be affected by other different, neighboring particles. For instance, if oxygen diffuses towards a single-celled pond organism at a certain rate, will that rate be altered by the presence of another type of molecule? Would the presence of other molecules block or enhance the diffusion of a molecule? Would the molecule's rate be independent of particles that do not alter the concentration gradient?

One way to measure the rate of diffusion of ions is to monitor their concentration in solution over a period of time. Since ions are electrically charged, water solutions containing ions will conduct electricity. A Conductivity Probe measures the concentration of ions in a solution, but not the concentration of electrically neutral molecules. Salts, such as sodium chloride, produce ions when they dissolve in water. If you place a salt solution inside a selectively permeable membrane such as dialysis tubing, the salt ions can diffuse out of the tubing and into the surrounding water.

OBJECTIVES

In this experiment, you will

- Use a Conductivity Probe to measure the ionic concentration of various solutions.
- Study the effect of temperature on diffusion.
- Study the effect of concentration gradients on the rate of diffusion.
- Determine if the diffusion rate for a molecule is affected by the presence of a second molecule.

MATERIALS

TI-Nspire handheld **or**	ring stand and utility clamp
computer and TI-Nspire software	dialysis tubing, 2.5 cm $\times$ 12 cm
data-collection interface	dropper pipet or Beral pipet
Vernier Conductivity Probe	scissors
three 18 $\times$ 150 mm test tubes with rack	stirring rod
1%, 5%, and 10% salt water	5% sucrose (table sugar) solution
400 mL beaker	dental floss or clamp
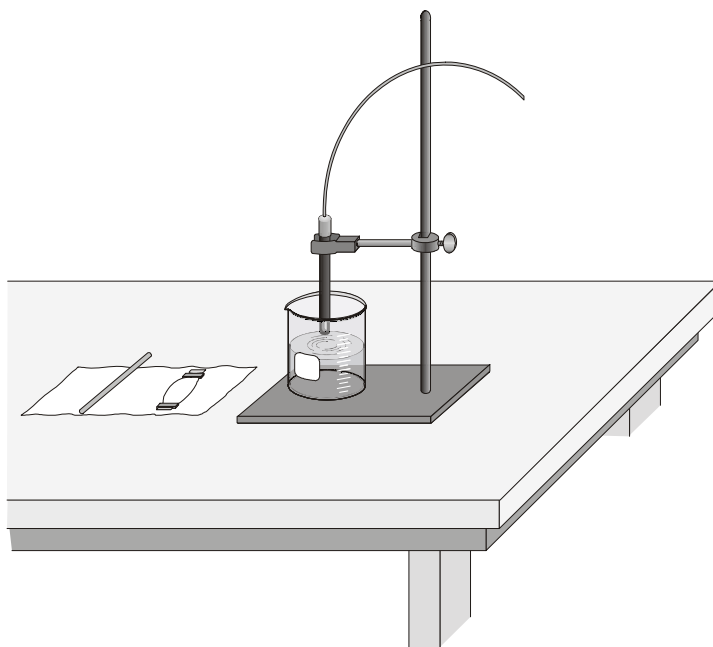
Figure 1

PROCEDURE

1. Obtain and wear goggles.

2. Set up the utility clamp and ring stand as shown in Figure 1.

3. Set the Conductivity Probe to the 0–2000 µS/cm range. Connect the Conductivity Probe to the data-collection interface. Connect the interface to the TI-Nspire handheld or computer.

4. Choose New Experiment from the 🔬 Experiment menu. Choose Collection Setup from the 🔬 Experiment menu. Enter **0.2** as the rate (samples/second) and **60** as the experiment duration in seconds. The number of points collected should be 13. Select OK.

**Part I Concentration gradients**

5. Test whether different concentration gradients affect the rate of diffusion. To do this, three solutions of differing salt concentrations (1%, 5%, and 10%) will be placed in distilled water. Each salt solution will be placed in a dialysis tube and allowed to diffuse into the surrounding water. When salt diffuses, the conductivity of the water in the beaker will increase.

6. In Table 1, predict what you believe will happen in this set of experiments. How will the rate of diffusion change when a 10% salt solution is placed in contact with pure water compared to when a 1% salt solution is placed in contact with pure water?

7. Prepare the dialysis tubing. Obtain a wet dialysis tube and a dialysis tube clamp or a short length of dental floss. Using the clamp or floss, tie one end of the tube closed about 1 cm from the end, as in shown Figure 2.

8. Place a 1% salt solution into a section of dialysis tubing.

   a. Obtain about 15 mL of a 1% salt water solution in a test tube.

   b. Using a funnel or Beral pipet, transfer about 10 mL of the 1% salt water into the dialysis tube, as in Figure 2. **Note:** To open the tube, you may need to rub the tubing between your fingers.

   c. Tie off the top of the dialysis tube with a clamp or a new length of dental floss. Try not to allow any air into the dialysis tube. The tube should be very firm after it is tied or clamped. Trim off any excess dental floss extending more than 1 cm from either knot.

   d. Wash the outside of the tubing with tap water thoroughly, so that there is no salt water adhering to the tubing.

Figure 2

9. Place 300 mL of distilled or deionized water into a 400 mL beaker. Secure the Conductivity Probe with the utility clamp in the water filled beaker as shown in Figure 1.

10. Place the dialysis tube into the water. Be sure the tubing is submerged completely under the water. **Important:** Position the Conductivity Probe and dialysis tubing the same distance apart in each trial.

11. After stirring the solution for 15 seconds, start data collection (). Stir the solution slowly and continuously throughout the one-minute data collection period.

12. Data collection will stop after 60 seconds. Analyze the graph to determine the rate of diffusion for the curve of conductivity *vs.* time:

    a. Examine the graph and identify the most linear region and select the data points in the most linear region.

    b. Choose Curve Fit ►Linear from the Analyze menu.

    c. Record the slope, *m*, as the rate of diffusion in (μS/cm/s) in Table 2.

13. Remove one of the clamps. If the dialysis tubing is tied off with floss, use a pair of scissors and carefully cut one of the dental floss knots and discard the floss. If you accidentally make a cut in the tubing, replace it.

14. Empty all of the liquid out of the dialysis tube. Squeeze the excess liquid out with your fingers.

15. Rinse the Conductivity Probe with distilled water.

16. Click the Store Latest Data Set button () to save data from the first run. Obtain 15 mL of a 5% salt solution in a test tube. Repeat Steps 8–15, substituting this 5% salt solution for the 1% solution.

17. Click the Store Latest Data Set button () to save data from the second run. Obtain 15 mL of a 10% salt solution in a test tube. Repeat Steps 8–15, substituting this 10% salt solution for the 1% solution.
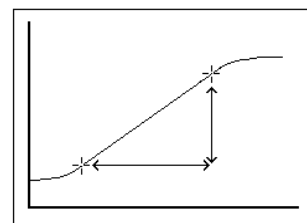
18. Graph all three runs of conductivity data on a single graph.

    a. Click **run3** and select All. All three runs will now be displayed on the same graph.
    b. Examine the graph closely and make a conclusion. Record your conclusion in Table 1.

19. (optional) Print a copy of your graph as directed by your instructor.

Part II Effect of other molecules

In this set of experiments you will measure the rate of diffusion of salt while it is in the presence of a non-conducting molecule. Since sugar does not form ions in solution, it should not conduct electricity. Therefore, sugar will be added to the water to determine whether it interferes with the diffusion of salt.

20. In Table 1, predict what you believe will happen in this set of experiments. Will the non-conducting sugar in the water block or reduce the rate of diffusion of salt? Explain your prediction.

Test to determine if water or a sugar solution conducts electricity.

21. Place about 100 mL of distilled or deionized water in a clean 400 mL beaker.

22. Test the conductivity of the water by placing a clean Conductivity Probe into it.

23. Click the Meter View tab (📷). Record the conductivity value in Table 3.

24. Obtain 300 mL of a 5% sugar solution in a clean 400 mL beaker.

25. Test the conductivity of the 5% sugar solution by placing a clean Conductivity Probe into the beaker. Record the conductivity value in Table 3.

Test if 5% sugar interferes with the diffusion of a 5% salt solution.

26. Click the Store Latest Data Set button (📥). Repeat Steps 7–15 with the following changes:

    a. Substitute a 5% salt solution for the 1% solution in Step 8.
    b. Use 300 mL of 5% sugar water, prepared in Step 24, in place of the water in Step 9.
    c. In Step 12c, record the slope, *m*, as the rate of diffusion in (μS/cm/s) in Table 4.

27. Graph the two 5% salt solution runs, one in water and one in a sugar solution, on a single graph.

    a. Click **run4** and select the More option. Select run2 and run4, then select OK. Both runs will be displayed on the same graph.
    b. Examine the graph closely and make a conclusion. Record your conclusion in Table 1.

DATA

Table 1		
	Prediction	Conclusion
Part I		
Part II		

Part I

Table 2 – Summary of Data	
Salt concentration (%)	Rate of diffusion (μS/cm/s)
1	
5	
10	

Part II

Table 3	
Solution	Conductivity (μS/cm)
Distilled water	
Sugar water	

Table 4 – Summary of Data	
Solution	Rate of diffusion (μS/cm/s)
5% salt	
5% salt / 5% sugar	

QUESTIONS

1. What conclusion can you draw from the data in Table 2?

2. How did your conclusion compare to your prediction for Part I? Can you account for any differences?

3. If the rates in any of the three experiments varied in Part I, calculate how much faster each rate was compared to that for the 1% salt solution. For instance, if the rate of the 1% solution was 1 μS/cm/s and the rate of the 10% solution was 5 μS/cm/s, then the rate of diffusion for the 10% solution would be (5/1) five times the rate of the 1% salt solution.

4. Compare the conductivity of pure water with a sugar solution. How do you account for this?

5. What conclusion can you draw from the data in Tables 3 and 4?

EXTENSIONS

1. Make a plot of the rate of diffusion *vs.* the salt concentration. Using your plot, estimate the rate of diffusion of a 3% salt solution.

2. If the results of the experiments in Part I can be extrapolated to diffusion in living systems, how would a single-celled organism respond in an oxygen rich pond compared to an oxygen-poor pond? Explain.

3. If waste products of an aquatic single-celled organism were released into a pond, how would that affect the organism's ability to obtain oxygen from the pond water? Explain how your data from Part II supports your answer.

4. Design an experiment to determine the effect of temperature on the diffusion of salt. Perform the experiment you designed.

5. Ectotherms are organisms whose body temperatures vary with the surrounding environment. On the basis of your data from Extension Question 4, how do you expect the oxygen consumption of ectotherms to vary as the temperature varies? Explain.