Diffusion and Osmosis

Pre-Lab Questions:

These questions are designed to help you understand kinetic energy, osmosis, and diffusion and to prepare for your investigations.

- What is kinetic energy, and how does it differ from potential energy?
- What environmental factors affect kinetic energy and diffusion?
- How do these factors alter diffusion rates?
- Why are gradients important in diffusion and osmosis?
- What is the explanation for the fact that most cells are small and have cell membranes with many convolutions?
- Will water move into or out of a plant cell if the cell has a higher water potential than the surrounding environment?
- What would happen if you applied saltwater to a plant?
- How does a plant cell control its internal (turgor) pressure?

Procedure 1: Surface Area and Cell Size

Cell size and shape are important factors in determining the rate of diffusion. Think about cells with specialized functions, such as the epithelial cells that line the small intestine or plant root hairs.

- What is the shape of these cells?
- What size are the cells?
- How do small intestinal epithelial and root hair cells function in nutrient procurement?

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Step 1:

1. Place .5 ml phenolphthalein in two test tubes.
2. Add .5 ml of 0.1 M HCl to one test tube,
3. Swirl to mix the solutions, and observe the color.
4. Using the same procedure, add 0.5 ml of 0.1M NaOH to the other test tube.
5. Record your observations.

Table 1: Color of 0.5 ml phenolphthalein when exposed to HCl and NaOH

Substance added to 0.5 ml of phenolphthalein	Observations of reaction
0.5 ml HCl	
0.5 ml NaOH	

- Which solution is an acid?
- Which solution is a base?
- What color is the dye in the base? In the acid?
- What color is the dye when mixed with the base?

Step 2:

1. Using a plastic knife and a metric ruler, cut a phenolphthalein agar block into a 2-cm cube.
2. Pour 100 mL of 0.1 M hydrochloric acid (or vinegar) into a 150-mL beaker.
3. Using a plastic spoon, carefully place the agar cube in the beaker of acid.
4. Gently agitate the solution and turn the cube with a spoon occasionally while soaking.
5. After 10 minutes, gently remove the agar cube using the plastic spoon. Blot the cube dry using a paper towel.
6. Using a plastic knife, cut the cube in half and measure the depth to which acid penetrated the cube.
7. Record observations and results.

Table 2: The diffusion rate of a 2cm^3 cube of phenolphthalein when exposed to acid for 10 min.

Cube	Surface Area (cm^3)	Volume (cm^3)	Surface Area-to-Volume Ratio	Diffusion Depth (mm)	Diffusion Rate (mm/min)
2cm					

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- Why are most cells so small?
- Why aren't cells larger?
- How does the rate of diffusion influence the ability of a cell to obtain needed nutrients?
- Predict how the surface area-to-volume ratio might affect the rate of diffusion into a cell.
- Many cells or organelles that play a key role in nutrient absorption or energy transfer have highly "convoluted" membranes with many folds. How does this affect the surface area of the cell or organelle and the rate of diffusion?

Design a controlled experiment to investigate the effects of surface area and cell volume on the rate of diffusion in agar model cells.

Step 3:

1. Using a dull knife or a thin strip of hard plastic, cut another block of agar that is not $2cm^3$.
2. Pour 100 mL of 0.1 M hydrochloric acid (or vinegar) into a 150-mL beaker.
3. Using a plastic spoon, carefully place the agar cube in the beaker of acid.
4. Gently agitate the solution and turn the cube with a spoon occasionally while soaking.
5. After 10 minutes, gently remove the agar cube using the plastic spoon. Blot the cube dry using a paper towel.
6. Using a plastic knife, cut the cube in half and measure the depth to which acid penetrated the cube.
7. Record observations and results.
8. Record the results of the rest of the class and process the data.

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Table 3: Diffusion rate of acid across phenolphthalein cubes of differing dimensions.

Cube From Group	Surface Area (cm^3)	Volume (cm^3)	Surface Area-to-Volume Ratio	Diffusion Depth (mm)	Diffusion Rate (mm/min)
1					
2					
3					
4					
5					
6					
7					
8					

- What statement can you make about the diffusion rate for the class data?

- What does this show you about the relationship of diffusion and cells?

Procedure 2: Modeling Diffusion and Osmosis

You are in the hospital and need intravenous fluids. You read the label on the IV bag, which lists all of the solutes in the water.

- Why is it important for an IV solution to have salts in it?

- What would happen if you were given pure water in an IV?

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- How would you determine the best concentration of solutes to give a patient in need of fluids *before* you introduced the fluids into the patient's body?

In this experiment, you will create models of living cells using dialysis tubing. Like cell membranes, dialysis tubing is made from a material that is selectively permeable to water and some solutes. You will fill your model cells with different solutions and determine the rate of diffusion.

- How can you use the mass of the filled cell models to determine the rate and direction of diffusion?

- What would be an appropriate control for the procedure you just described?

- Suppose you could test other things besides the mass of the dialysis tubes. How could you determine the rates and directions of diffusion of water, sucrose, NaCl, glucose, and ovalbumin?

- Will protein diffuse? Will it affect the rate of diffusion of other molecules?

Step 1:

Follow Table 4 below to create five model cells filled with 10 ml of various substances. One solution from each pair will be in the model cell of dialysis tubing, and the other will be outside the cell in the cup. Your fifth model cell will have water inside and outside; this is your control. Before starting, use your knowledge about solute gradients to predict whether the water will diffuse into or out of the cell. Make sure you label the cups to indicate what solution is inside the cell and inside the cup.

1. Make dialysis tubing cells by tying a knot at one end of your dialysis tubing.
2. Fill each "cell" with 10 mL of the appropriate solution from Table 4.
3. Knot the other end, leaving enough space for water to diffuse into the cell, but making sure there is no air in the cell.
4. Weigh each cell, record the initial weight, and then place it into a cup filled with the second solution for that pair.
5. Weigh the cell after 30 minutes and record the final weight.
6. Calculate the percent change in weight using the following formula: (final – initial)/initial X 100. Record your results.

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Table 4: Percent change of cell model mass when exposed to various solutes in external environment

Cell #	**Model Cell** (inside dialysis tubing)	**Surrounding Environment** (plastic cup)	**Net Diffusion Prediction** (into or out of cell)	**Mass of Cell Model at Start**	**Mass of Cell Model at End**	**Percent Change in Mass**
1	1 M NaCl	Water				
2	Water	1M Glucose				
3	1 M Sucrose	Water				
4	Albumin	1M Sucrose				
5	Water	Water				

- Which pair(s) that you tested did not have a change in weight? How can you explain this?
- If you compared 1 M solutions, was a 1 M NaCl solution more or less hypertonic than a 1 M sucrose solution? What is your evidence?
- Does the protein solution have a high molarity? What is evidence for your conclusion?
- How could you test for the diffusion of glucose?
- Based on what you learned from your experiment, how could you determine the solute concentration inside a living cell?

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Step 2:

An absent minded teacher (Mr. Franco) forgot to make a key for the colored sucrose solutions needed for lab. Design a lab to help him figure out the concentration of each solution. He knows he has prepared 1.0M, 0.8M, 0.6M, 0.4M, and 0.2M solutions. Tell him what color represents each solution. Remember, Mr. Franco is cheap so you can only use 10ml of each of his colored solutions to run your experiment. He has provided you with Table 5 to help organize your data. Good Luck!!!

Table 5: Sucrose concentration in various cell models to determine unknown solution

Solution Color	Initial Mass (of cell)	Final Mass (of cell)	Percent Change in Mass	Sucrose Concentration in Dialysis Bag

Procedure 3: Observing Osmosis in Living Cells

The interactions between selectively permeable membranes, water, and solutes are important in cellular and organismal functions. For example, water and nutrients move from plant roots to the leaves and shoots because of differences in water potentials. Based upon what you know and what you have learned about osmosis, diffusion, and water potential in the course of your investigations, think about these questions.

- What would happen if you applied saltwater to the roots of a plant? Why?
- What are two different ways a plant could control turgor pressure (internal water pressure/potential) within its cells?
- Will water move into or out of a plant cell if the cell has a higher water potential than it's surrounding environment?

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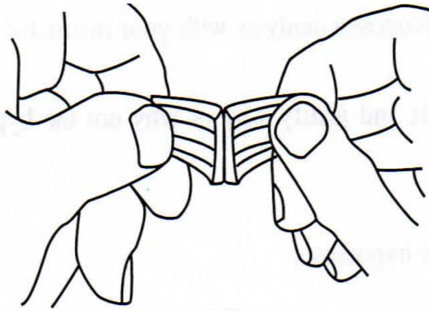
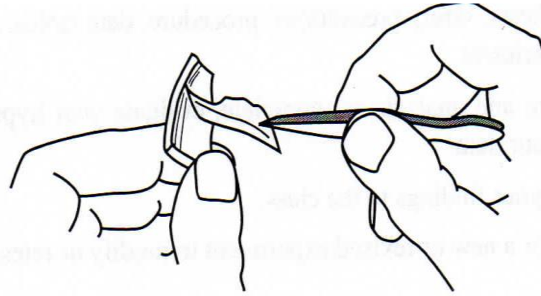
Step 1:

Start by looking at an onion epidermis sample placed on a wet mount. The sample cells are then observed with and without treatment of sodium chloride solution.

1. Place a drop of water on a clean microscope slide.
2. Using forceps, peel the thin purple epidermis off of the concave side of the onion bulb's leaf scale.

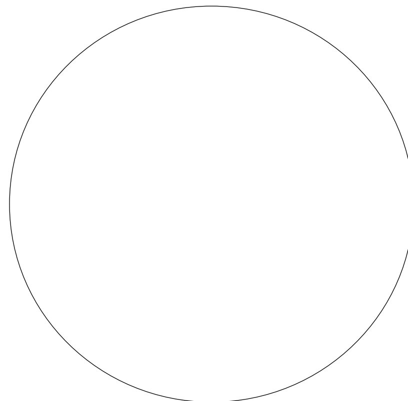


Break Layer



Pull thin layer of epidermis from onion.

3. Place the epidermis sample on the wet mount slide.
4. Place a cover slip on the sample.
5. Place the slide on the microscope stage and focus the specimen.
6. Observe the purple epidermal cells, the cell wall, the cell nucleus, the central vacuole, and the cell membrane.
7. Diagram and label structures in your observations.



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8. Add a drop of the sodium chloride solution on the microscope slide next to the edge of the cover slip.
9. Place the edge of a paper towel on the opposite side of the cover slip as the sodium chloride drop to draw the solution across the slide.
10. Diagram and label structures in your observations.

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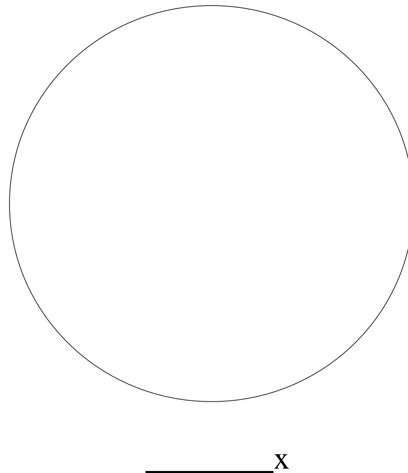

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11. Observe the cells' organelles.
    - How are they the same and how are they different than when treated with water?
    - Based on experimental results, when the 10% sodium chloride solution was added to the slide, was the water potential higher outside or inside the epidermal cells?

Step 2:

Consider the following questions while reflecting upon your knowledge of water potential.

- How can you measure plant pieces to determine the rate of osmosis?
- How would you calculate the water potential in the cells?
- Which colored sucrose solution would have a water potential equal to that of the plant cells? How would you find out?
- Will the water potential in different plants be the same?
- What would your results be if the plant material were placed in a dry area for several days before your experiment?

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- When plants are in the ground, do they swell with water when it rains? If not, how do you explain that, and if so, what would be the advantage or disadvantage?

Design an experiment to use the 1.0M, 0.8M, 0.6M, 0.4M, 0.2M sucrose solutions to determine the water potential of the plant tissues. (You might want to review the information on water potential described in Understanding Water Potential.)

1. Select a plant tissue for your investigation.
2. Decide on the mass of the material you will be testing (stay consistent).
3. Set up 6 experimental cups filled with 100ml of distilled water, 1.0M sucrose, 0.8M sucrose, 0.6M sucrose, 0.4M sucrose, 0.2M sucrose.
4. Let your specimens sit in the solutions for 24 hours.
5. Use table 6 to record your data.
6. **Graph your group's results to calculate the water potential for your specimen.**
7. Share your specimen's water potential with the other groups.
8. Collect data from the other groups.
9. **Graph the water potentials for each specimen. Putting them in order from highest to lowest.**

- What tissue probably has the most solutes?

- What tissue probably has the least solutes?

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Table 6: Change of mass in tissue specimens for various solute concentrations over a 24 hr period

Specimen for Group #	Sucrose Concentration (M) outside specimen (± .05 M)	Mass of Tissue Initial in g (± .1 g)	Mass of Tissue Final in g (± .1 g)	Change in Mass of Tissue (g)	Water Potential Of Tissue (bars)
1					
2					
3					
4					

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Table 6: Change of mass in tissue specimens for various solute concentrations over a 24 hr period(cont)

Specimen for Group #	Solute Concentration in Cell M (± .05 M)	Mass of Cell Initial in g (± .1 g)	Mass of Cell Final in g (± .1 g)	Change in Mass (g)	Water Potential (bars)
5					
6					
7					
8					