

Human Physiology Lab (Biol 236L)

Passive and Active Transport

Background:

Substances are routinely transported (received and delivered) across the cell plasma membranes. These substances include compounds, liquids, nutrients, hormones and other signaling molecules, and waste products. Transport into and out of cells can be one of two ways: 1) Passive transport or 2) Active transport. **Passive transport** involves movement of substances that does not require energy (**ATP**). Typically in passive transport solutes move from an area of higher concentration to lower concentration (down or with their concentration gradient). Types of passive transport include simple diffusion, facilitated diffusion, osmosis, and filtration. In **simple diffusion** substances move (e.g. **solutes** like ions, molecules, gases) down their concentration gradient from higher to lower concentration without the aid of cell surface channels (Figure 1). This diffusion can continue until the solute concentration in the extracellular and intracellular space is the same or at **equilibrium**. In **facilitated diffusion** solutes move from higher to lower concentration with the help of cell surface channels. Whereas diffusion and facilitated diffusion involve movement of solutes, **osmosis** involves movement of water (or **solvents**).

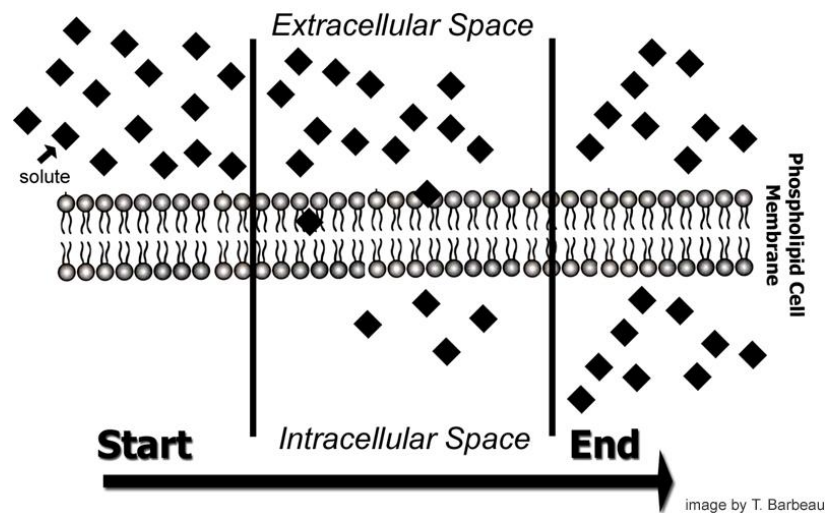


image by T. Barbeau

Figure 1. Example of a solute diffusing across a plasma membrane. At the start of diffusion the extracellular solute concentration is greater than within the intracellular space. After time passes the solute diffuses across the cell membrane until, at the end, solute concentration between extracellular and intracellular space has reached equilibrium.

Osmosis is influenced by the concentration gradient of solutes found on the outside versus the inside of a cell. Water will move, by osmosis, from the side of a cell having a lower solute concentration to the side having a higher solute concentration. (*Think of water as “wanting to dilute more concentrated solutions”.*) Movement of water by osmosis depends on tonicity. **Tonicity** is a measure of the

difference in concentrations (osmotic pressure gradient) of two solutions separated by a semipermeable membrane. You can observe the effects of tonicity on osmosis by examining what happens to red blood cells placed in solutions (Figure 2) that are either **hypotonic** (having lower solute concentration than inside the cell), **hypertonic** (having higher solute concentration than inside the cell), or **isotonic** (having the same solute concentration outside as inside the cell). **Filtration**

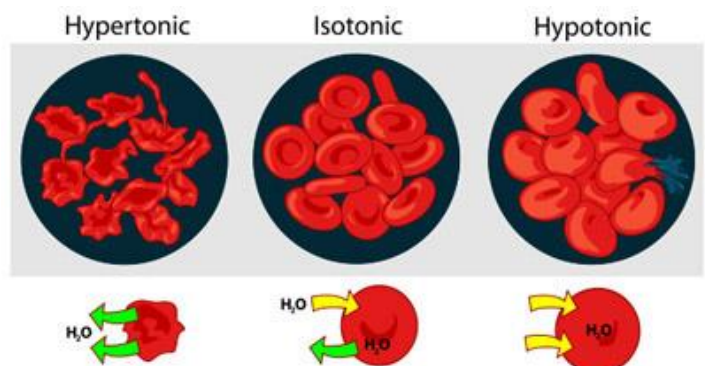


Figure 2. Tonicity and osmosis in red blood cells.

involves the movement of substances across an epithelial membrane, such as that found in capillaries. The substances are moved (or pushed) across the membrane by blood pressure within the capillaries. **Active transport** involves the movement of substances against their concentration gradient, usually with the aid of cell-surface pumps, from areas of lower to areas of higher concentration (Figure 3). This kind of transport is called active because it requires energy input. In the following laboratory you will observe the process of passive transport: osmosis and diffusion across cell membranes using two different experiments.

Example of Active Transport

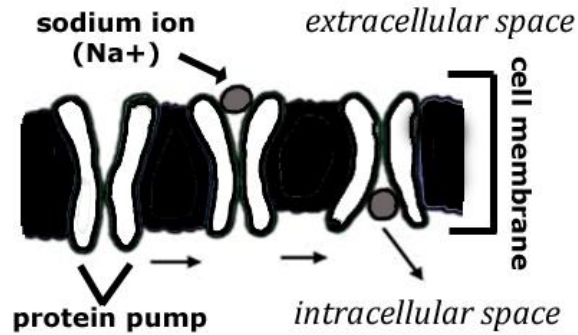


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Figure 3. Active transport of sodium ions across the cell membrane.

Part 1: Osmosis across an egg cell membrane.

To examine the effects of tonicity on osmosis across cell membranes you will examine the movement of fluid across the membrane of an egg that has been de-calcified by decalcification (24 hr soak in vinegar) to remove the calcium egg shell and expose the egg cell membrane. With the membrane exposed we will place the egg cell into hypertonic corn syrup solution or a hypotonic solution of tap water. [***Your instructor will have decalcified the egg ahead of time, taken initial egg volume measurements, and then soaked the egg in corn syrup about 2 hours ahead of lab.*]

Methods:

1. Carefully take a decalcified egg and measure the circumference at its length and width using a tape measure.
2. Gently place the egg into a beaker and add enough corn syrup to cover the egg.
4. Allow the egg to sit in the corn syrup for **60 minutes**.
5. After 60 minutes remove the egg and wipe off the corn syrup. Observe the egg for changes & record these in Table 1. Pour the used corn syrup back into the jar and clean out your beaker.
6. Carefully place egg into a new beaker and cover with water, and let egg sit for 1 hour.
7. After 1 hour check the egg and note any changes, and record these in Table 1.

Table 1: Tonicity and Changes in Osmosis Across an Egg Cell Membrane

	Pre-Soak (original) V_o	Post-Syrup Soak (V_s)	Post-Water Soak (V_w)
Egg Diameter (Length)*			
Egg Diameter (Width)*			
Egg Volume*			

* To calculate egg volume use the following equation:

$$V = KV_c \times [L \times W^2]$$

Where V = egg volume

KV_c = volume coefficient of 0.496

L = length circumference / 2

W = width circumference / 2

Questions:

1. Did the egg volume change after soaking in corn syrup, and if so, how? _____.
2. When the egg was soaking in the corn syrup, which environment was more hypertonic? The inside of the egg or the outside of the egg?
_____.
3. Explain, with respect to osmotic movement of water, why might the egg volume have changed after soaking in corn syrup.
_____.
4. Did the egg volume change after soaking in water, and if so, how? _____.
5. When the egg was soaking in the water, which environment was more hypertonic? The inside of the egg or the outside of the egg?
_____.
6. Explain, with respect to osmotic movement of water, why the egg volume might have changed after soaking in water.
_____.

Part 2: Diffusion across a Semipermeable Dialysis Membrane.

Cell membranes and blood vessels are **selectively permeable**, meaning that they can alter their pore size to allow movement across the membrane. All cell membranes have the ability to alter their lipid and protein contents to change their pore size. By regulating membrane permeability cells can allow specific molecules or ions to move into or out of the cell. As solutes move across a selectively permeable membrane they move by diffusion from a solution containing more solute to a solution containing less solute.

The membranes used in this experiment are **semi-permeable**, meaning the pore sizes of the membrane are "fixed" and the molecules larger than the pores will not be allowed to pass through the membrane, while those smaller than the pores will pass through easily. This will be demonstrated by the following experiment.

Methods:

1. Work in groups of four. Obtain a piece of pre-cut 10 inch dialysis tubing; tie a tight knot at the bottom of one end. Be careful not to tear the tubing - if the tube leaks, the experiment will not work properly.
2. Add 2 dropperfuls of 15% glucose to the dialysis bag.
3. Add 2 dropperfuls of 1% starch to the same bag. Record the color of the contents in the bag: _____.
4. Fill a beaker $\frac{1}{2}$ full of lukewarm water and add 3 dropperfuls of Lugol's reagent (Iodine) to the beaker. You want the water to look yellowish. If it's not, add more iodine until it is.
Record the color of the contents in the beaker: _____.

5. Place the bag inside the beaker so that the top end of the bag hangs over the edge of the beaker. Secure the open end of the bag with a rubber band so it is held tight to the beaker. When your setup is finished, it should look like Figure 4.

6. Place the beaker on your lab table and allow it to stand undisturbed for **30 minutes**. [During this time you can attend to any other experiments that are running.]

7. Stir a glucose test strip in the beaker water. Consult the color test chart on the glucose test strip container to determine the presence or absence of glucose, and fill in Table 2 below.

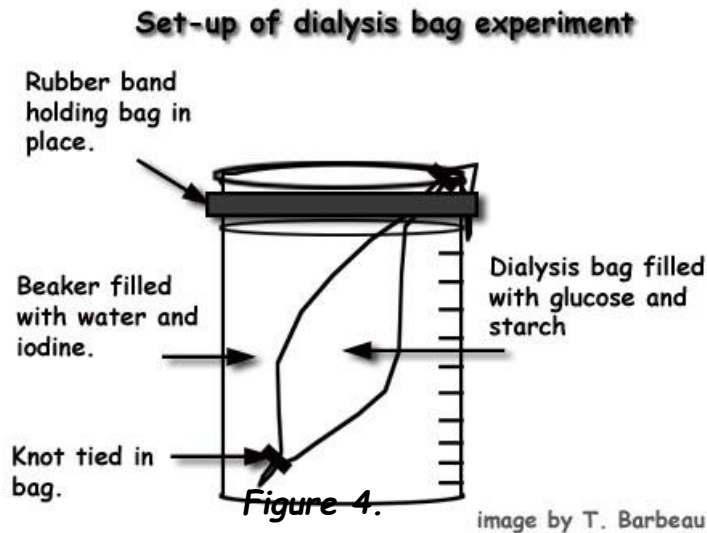


Table 2: Osmosis and Diffusion Across a Semi-Permeable Membrane

Initial Conditions (Start of Experiment)		Final Conditions (After 20 min.)	
Bag Color	Beaker Color	Bag Color	Beaker Color
glucose + or — ? starch + or — ?	glucose + or — ? starch + or — ?	glucose + or — ? starch + or — ?	glucose + or — ? starch + or — ?

Questions:

1. According to the test chart for the glucose strips:

Is glucose detected in the bag? _____.

Is glucose detected in the beaker? _____.

2. Lugol's reagent tests for the presence of _____.

Is it an indicator solution? _____.

3. Which substances entered the bag? _____.

4. Which substances left the bag and entered the beaker? _____.

5. Why did starch not diffuse into the beaker? _____.

6. Is the dialysis tube used in this experiment semipermeable? _____.

How do you know this? _____.