Biology
Mr. Croft

# Principles of Diffusion and Osmosis
**40 Points**

***Answer all questions on a separate sheet of paper.***

**Activity One - Learning about Osmosis and Dialysis Membranes**

Procedure:

1. Prepare two pieces of dialysis tubing as follows:
   - Roll the dialysis bag between your thumb and pointer finger under tap water to open the bag.
   - Fold over approximately 1 inch of the end of the dialysis tubing.
   - Carefully tie a knot 1/2 inch from the end of the dialysis tubing.
   - Be sure to keep the tubing moist by rinsing with tap water as needed.
2. Prepare two water baths as follows:
   - Fill two cups with 200 mL of distilled water.
3. Fill the first dialysis bag with violet dye:
   - Roll the untied end of the dialysis bag between your thumb and pointer finger to reopen the end of the bag.
   - Squeeze the transfer pipet bulb and fill it with dye up to a level just below the pipet bulb.
   - Gently insert tip of the transfer pipet deep into the bag.
   - Apply pressure on the bulb to transfer the contents of the pipet to the dialysis bag.
   - Carefully tie a knot at the open end of the bag using the technique in Step 1.
4. Place the dialysis bag in one of the water baths.
5. Fill the second dialysis bag with orange dye. Follow the same procedures as outlined in Steps 3&4. You should now have two filled dialysis bags (one violet, and one orange) in a beaker of water.
5. Wait 15 minutes (work on Activity 2 while you are waiting).
6. One at a time, remove each bag filled with dye and place it on a piece of wax paper.

Questions (3 Points Each):

1. Record your observations for each dialysis bag.
2. Is there any change in the color of the liquid in the bags? in the water?
3. Which of the two dyes has a smaller molecular size? Why? Which has a larger molecular size? Why?

**Activity Two - Effect of Salt on Osmosis**

Procedure:

1. Prepare a fresh dialysis bath as described in Activity One.
2. Add 5 Teaspoons of salt to the dialysis bath. Stir until the salt is dissolved.
3. From Activity One, obtain dialysis bag that still contains dye.
   Hint: This is the high molecular weight dye, and it did not pass through the dialysis membrane.
4. Place the dialysis bag saved from Activity One into the dialysis bath that now contains salt.
5. After 24 hours, observe if there is any change in the dialysis bag.

6. Periodically check to see if any changes in color or volume has occurred.

Questions (3 Points Each):

7. Has the color changed?
8. Does the volume of the bag appear the same or different?
9. Analyze the contents of the bag.

Review Questions (7 Points Each):

10. What molecular weight cut off value would allow both dyes from Activity One to penetrate the membrane?
11. How could the rate of dialysis of the orange dye from Activity One be increased?
12. A protein having a molecular weight of 65,000 has been purified. The protein solution has a high concentration of sodium chloride as a result of the last purification step. Suggest a method for removing the salt from the protein solution.

Information about the dialysis tubing and dyes:

Dialysis membranes are made of purified cellulose containing microscopic pores. The pore size is controlled during manufacturing. The pore size determines the membrane's permeability to solutes of different sizes. Increasing size generally corresponds to increasing molecular weight when the molecules have similar shapes. The dialysis tubing being used in this experiment has a molecular cut off of approximately 10,000. The orange dye has a molecular weight of about 300 and the violet dye has a molecular weight in excess of 100,000.