Laboratory 2 – Molecular Activity and Membrane Transport

<u>Purpose:</u> The purpose of molecular activity and membrane transport is to examine the elementary properties of passive transport, diffusion, osmosis, and differential permeability. It a way to understand the fundamentals of filtration and the effects of tonicity on different cells.

Procedures:

2-B: Measurement of diffusion through a liquid Procedure

- 1. Working in groups, fill three Petri dishes with 40 ml. of 25°C water.
- 2. Drop one crystal of potassium permanganate into each dish. Be sure to use the same amount of potassium permanganate for each dish. Record the time.
- 3. Measure, in millimeters, and record the largest diameter of the colored spot after 5 minutes.
- 4. Repeat steps 1-3 for water at 5°C and at 45°C.
- 5. Construct a graph of ranges and means for each temperature.
- 6. Based on your knowledge of diffusion, what is an explanation for these results?

2-C: Measurement of diffusion through agar Procedure

- 1. Petri dishes have been filled with agar. Two holes have been made in the agar. Into one hole, place two drops of methylene blue. Into the other hole, place two drops of potassium permanganate. Record the time and immediate diameter of each spot. This will be your time zero Measurement.
- 2. Measure the diameter of each spot in <u>millimeters</u> once every minute for fifteen minutes. Calculate the averages from the data collected by all groups doing this exercise. Summarize these data.
- 3. Construct a graph of average diffusion diameter versus time for both chemicals.
- 4. Determine the diffusion rate of each chemical. Which has the fastest diffusion rate, methylene blue or potassium permanganate? Record these results.
- 5. Look up the molecular formula and structure of methylene blue and potassium permanganate in a Merck Index. Make note of this information.
- 6. Interpret your result with respect to the information obtained from the Merck Index.

2-D: Demonstration of filtration Procedure

- 1. Fold three filter papers into cones and insert them into three separate glass funnels. Wet the papers to make them stick to the glass.
- 2. Prepare three 100-milliliter solutions of charcoal and water. Make one thick, one medium thickness, and one thin. Record the mass of the charcoal used in each preparation.

NOTE: if your "thin" solution continually runs through the filter, making it impossible to count drops, it is too thin; you will need to make all your solutions proportionally thicker.

- 3. Pour 50 ml of each solution, one at a time, into a funnel.
- 4. Immediately count the number of drops produced per minute.

NOTE: it may be easier to count the drops for 15 seconds then multiply by four to obtain drops per minute.

- 5. Count the number of drops per minute when the funnel is half-filled.
- 6. Count the number of drops per minute when the funnel is nearly empty.
- 7. Did the charcoal pass into the filtrate? Which solution had the fastest rate of filtration? What is the driving force behind filtration? What other factors influence the rate of filtration? Do your results illustrate these influencing factors?
- 8. Repeat these procedures with the remaining 50 ml. of solution.

2-F: Measurement of osmosis Procedure

- 1. Attach dialysis bags filled as much as possible with sucrose solutions securely to the bottom of two open, thin glass tubes. One bag should be filled with a 25% sucrose solution and the other should be filled with a 50% sucrose solution. Make sure ends of the tubes are immersed in the solutions. NOTE: reliable results depend on your ability to tightly seal the dialysis bags.
- 2. Insert both bags into separate beakers of distilled water making sure the dialysis bags are fully submersed but not touching the bottom of the beakers, and suspend each by gently applying a ring stand clamp to the glass tubes. Check for solution leaking out of the bags.
- 3. Allow five minutes for the systems to equilibrate. Then, mark the fluid levels of each glass tube with a felt pen. Record the time.
- 4. Record the fluid level of the glass tubes in millimeters every 10 minutes for 50 minutes.
- 5. If the fluid level rises to the top of the glass tube sooner than 50 minutes, record the time it took to get there, measure the length in millimeters from the equilibration line to the top of glass tube. Divide that length by the number of minutes to get your rate in mm/min.
- 6. Determine the rate of osmosis for each system. Which system had the fastest osmotic rate, the 25% or 50% sucrose solution? Explain these results.

2-G: Measurement of differential permeability of sugar and starch.

- 1. Fill a dialysis bag with a 1% starch -10% glucose solution. Reliable results depend on your ability to tightly seal the dialysis bag.
- 2. Tie the bag to a glass rod and suspend it in a beaker of distilled water.

NOTE: Test the water from the bottom of the beaker to ensure that it is free of starch and/or sugar.

3. After 15 minutes has passed check the water again for starch and sugar in the following way:

Test for starch:

a. Add 10 drops of Lugol's solution to 5 ml of water obtained from the beaker.

Reddish color = No starch

Navy blue color = Starch present

Test for sugar:

a. Add 3 ml of Benedict's solution to 5 ml of water obtained from the beaker. Simmer the solutionat a low boilfor 5 minutes.

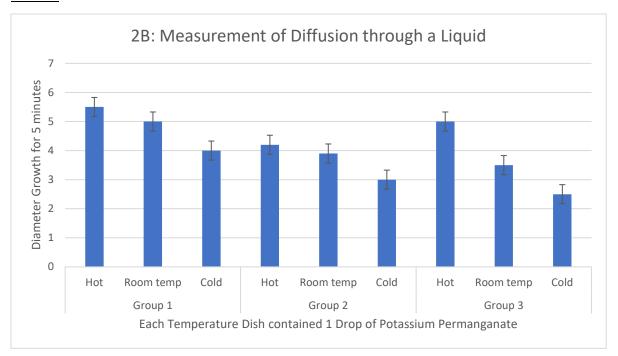
Blue color = No sugar

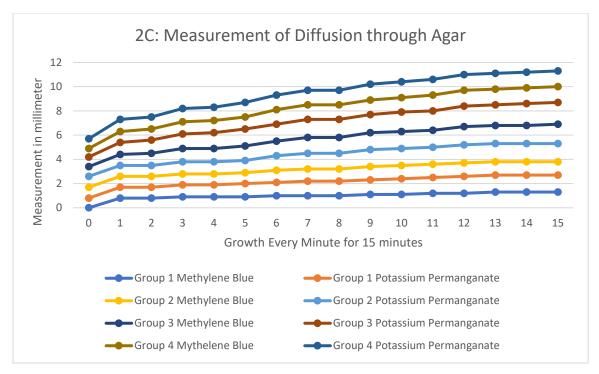
Color change = Sugar present

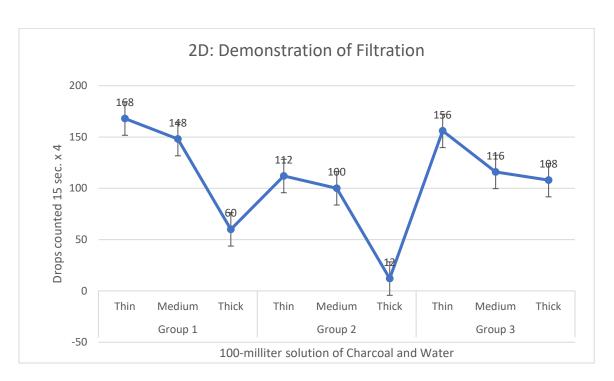
(green = little sugar; yellow = moderate sugar; orange = more sugar; red = lots of sugar)

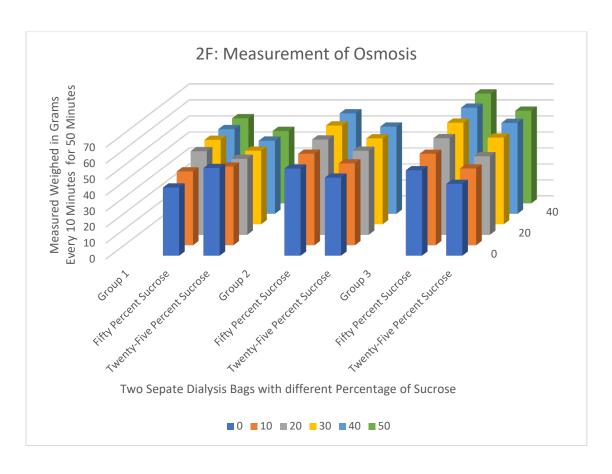
- 4. Test the water in the beaker again at 30, 45 and 60 minutes.
- 5. Record these results. Explain the significance of these findings in relation to the permeability of the dialysis bag.

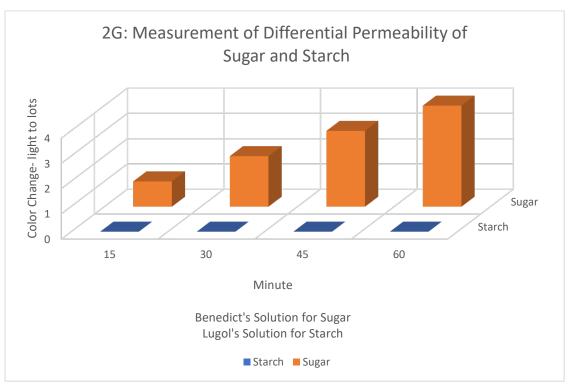
Results:











<u>Discussion:</u> I found all the lab experiments interesting. My two favorite experiment was 2B, the Measurement of diffusion through a liquid, and 2-C, the Measurement of diffusion through agar. Those experiments were unique because I enjoyed watching the substance expand in diameter. I was surprised that the hot water in 2B absorbed quicker than the cold water. However, I was not surprised with the agar experiment because I was able to do that particular experiment in my Biology class a few years ago. But it brought back memories of experimenting with the agar for the first time, which I recall being amazed.

<u>Conclusion</u>: Molecular activity and membrane transport lab activities are a great way to learn, observe, and understand how different materials diffuse through passive and active transportation. This was fun!