

Purpose

The purpose of this lab is to teach us the basic properties of passive transport, diffusion, osmosis, and differential permeability. The concept of filtration will also be explained, and the impact of tonicity on cells will be examined.

Procedures

2-B: Measurement of diffusion through a liquid

1. Working in groups, fill three Petri dishes with 40 ml. of 25C 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 5C and at 45C
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

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 millimeters 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 MerckIndex. Make a note of this information.
6. Interpret your result with respect to the information obtained from the MerckIndex

2-D: Demonstration of filtration

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 and 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?

2-F: Measurement of osmosis

1. Attach the dialysis bag filled 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 the 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 submerged but not touching the bottom of the beakers, and suspend each by gently applying a ring stand clamp to the glass tubes. Check for solutions 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 the 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 have 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 solution at a low boil for 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.

2-H: The effects of tonicity on red blood cells-Demonstration

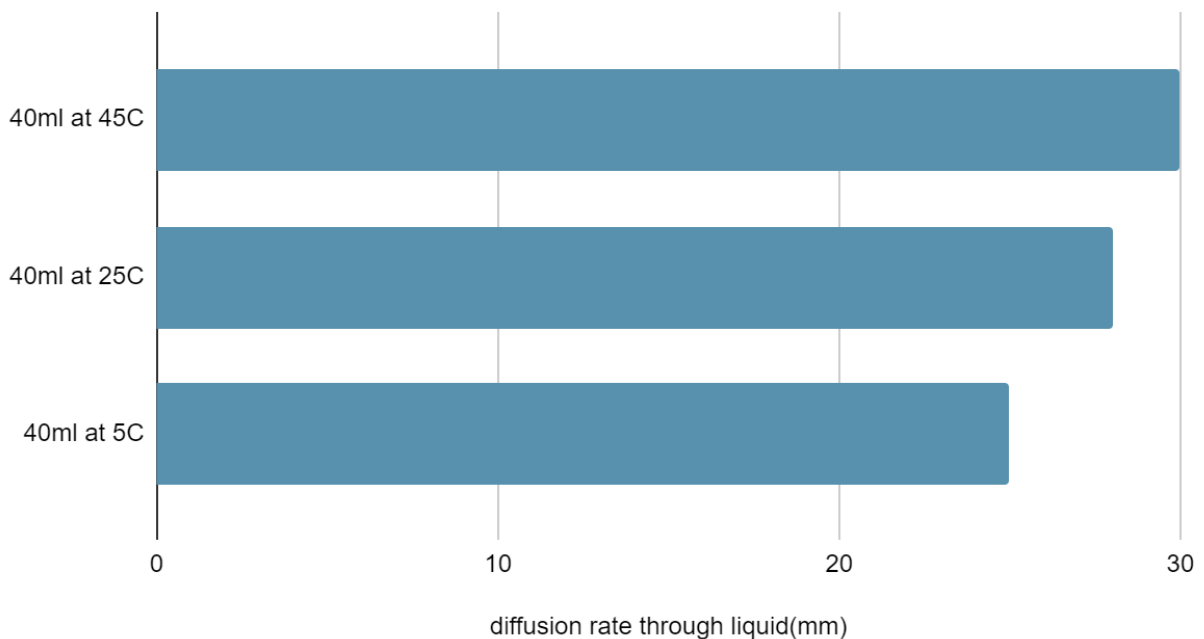
1. One milliliter of each of the following solutions will be in three separate test tubes.
 - a. Distilled water (hypotonic)
 - b. Physiological saline–0.85% NaCl (isotonic)
 - c. Saltwater–2.0% NaCl (hypertonic)
2. A small drop of blood will be added to each tube and the contents thoroughly mixed
3. A wet mount slide will be made of each solution.
4. Examine each slide under the high-dry lens of a compound microscope.
5. Observe the following:
 - a. Hemolysis of cells in the hypotonic solution. (Note the transparent solution.)
 - b. Maintenance of cell size in the isotonic solution.
 - c. Crenation of cells in the hypertonic solution.
6. Make a drawing of each observation and provide an explanation for each.

Results

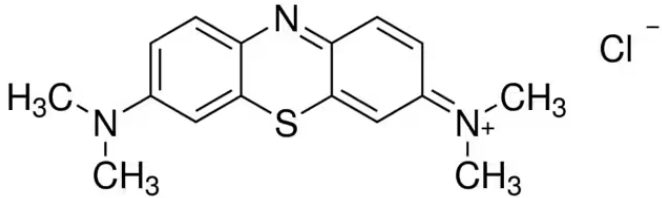
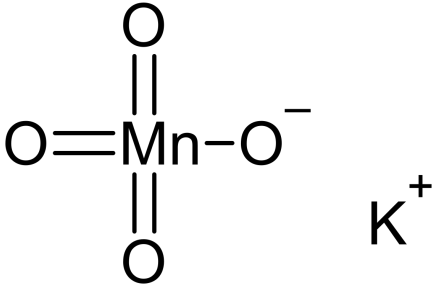
2-B: Measurement of diffusion through a liquid

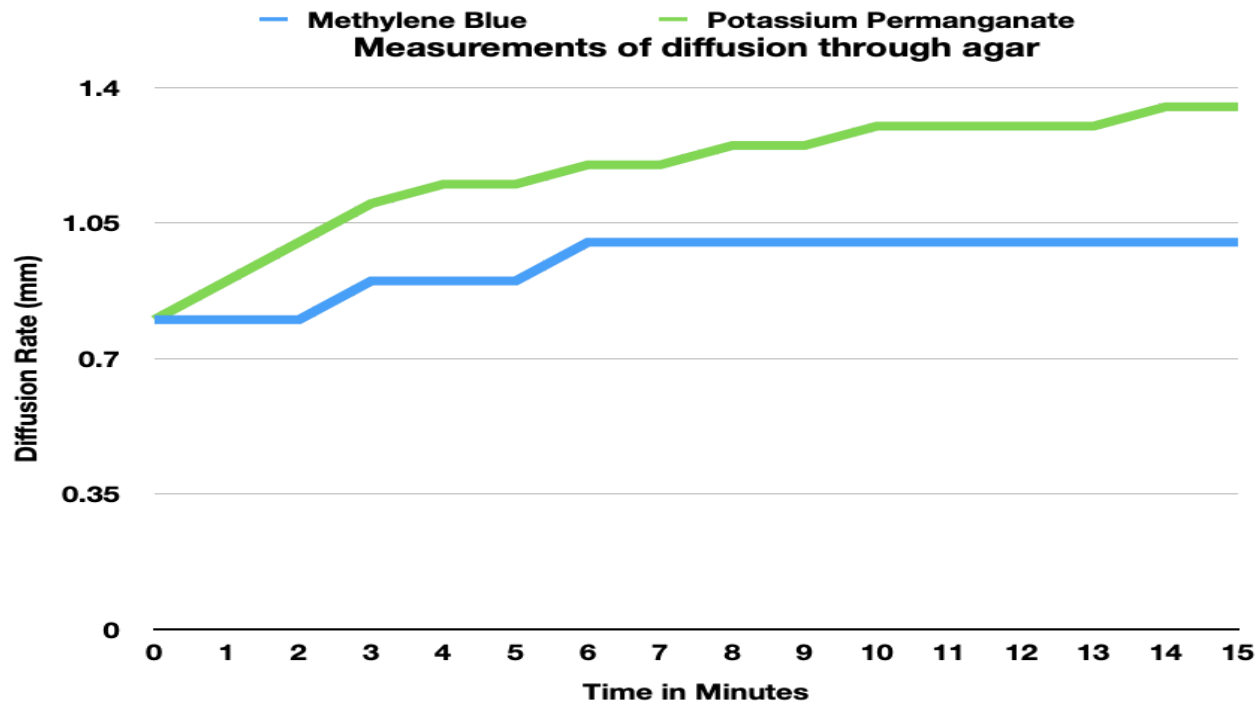
(Room temp) 40 ml at 25°C= 28mm (hot)40 ml at 45°C= 30mm (cold)40 ml at 5°C=25mm

Measurement of diffusion through a liquid after 5 min



2-C: Measurement of diffusion through agarMolecular weight: Methylene Blue: **319.85g/mol** Potassium Permanganate: **158.03g/mol**

Time (Minutes)	Methylene blue	Potassium Permanganate	Formula and Structure for Methylene Blue (C ₁₆ H ₁₈ ClN ₃ S)
0	0.8mm	0.8mm	 Formula and Structure for Potassium Permanganate(KMnO ₄) 
1	0.8mm	0.9mm	
2	0.8mm	1.0mm	
3	0.9mm	1.1mm	
4	0.9mm	1.15mm	
5	0.9mm	1.15mm	
6	1.0mm	1.2mm	
7	1.0mm	1.2mm	
8	1.0mm	1.25mm	
9	1.0mm	1.25mm	
10	1.0mm	1.3mm	
11	1.0mm	1.3mm	
12	1.0mm	1.3mm	
13	1.0mm	1.3mm	
14	1.0mm	1.35mm	
15	1.0mm	1.35mm	
Avrg:	.94mm	1.18mm	

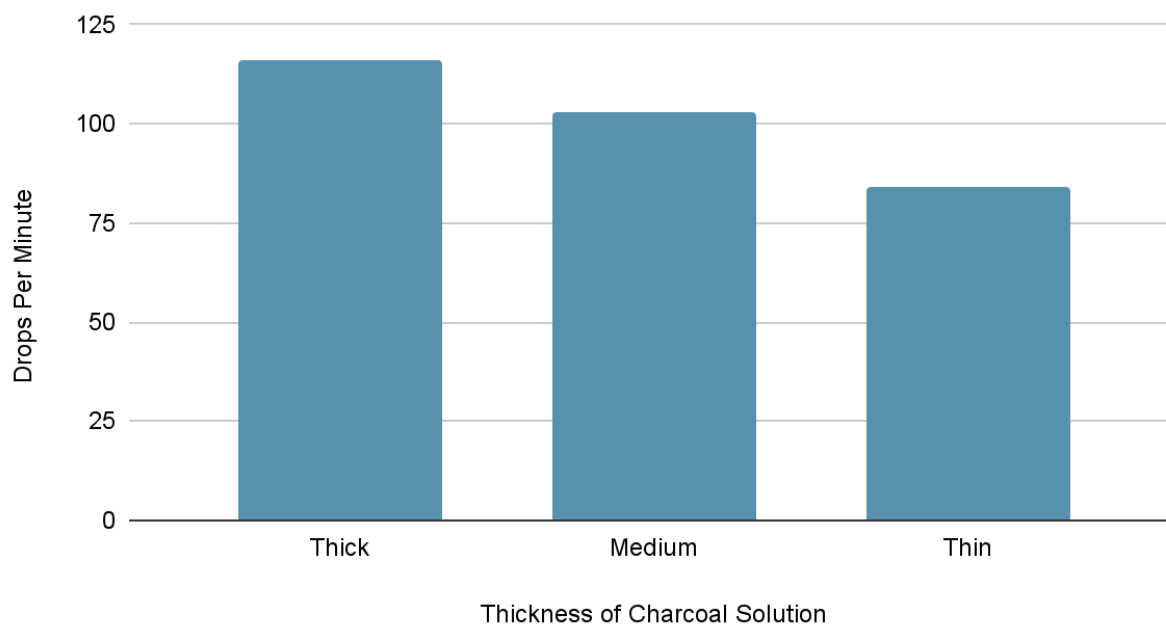


2-D: Demonstration of filtration

Mass of charcoal- Thin Solu: 0.39G Medium Solu: 1.11g Thick Solu: 7.00g

Drops/minute(filled):	Thin Solu: <u>120</u>	Medium Solu: <u>156</u>	Thick Solu: <u>160</u>
Drops/minute(Half):	Thin Solu: <u>80</u>	Medium Solu: <u>96</u>	Thick Solu: <u>116</u>
Drops/minute(near empty):	Thin Solu: <u>52</u>	Medium Solu: <u>56</u>	Thick Solu: <u>72</u>
Average drops per minute:	Thin: <u>84</u>	Medium: <u>103</u>	Thick: <u>116</u>

Effects of Solution Density on Filtration Rate



2-F: Measurement of osmosis

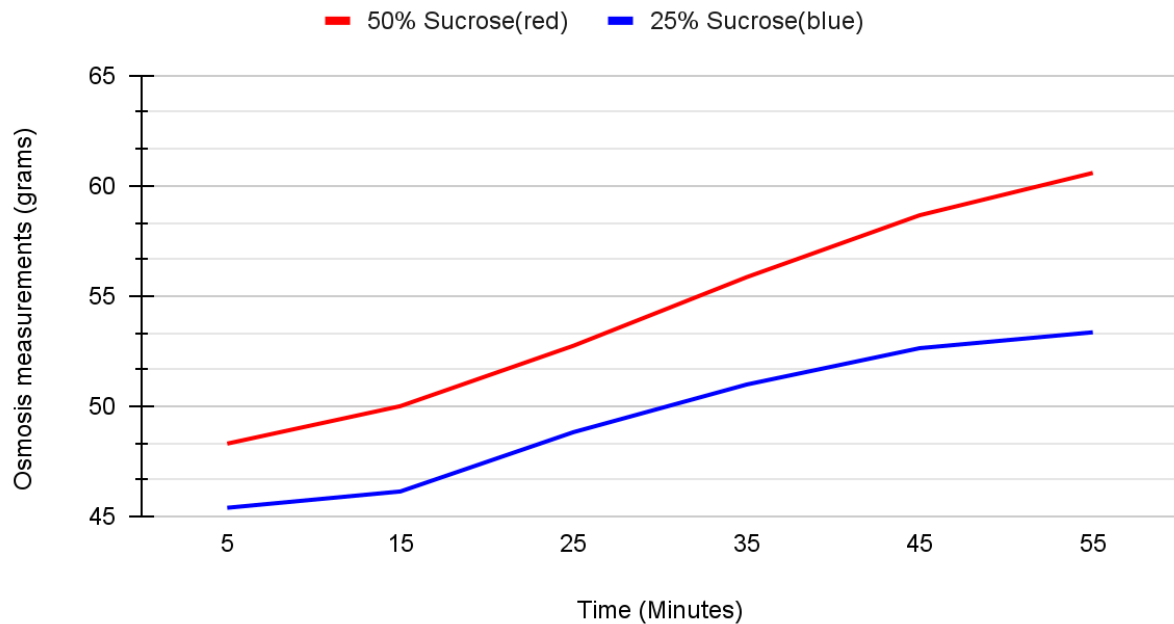
50% Sucrose(red) Initial weight 44.93g

5 minutes	48.30g
15 minutes	50.0g
25 minutes	52.74g
35 minutes	55.85g
45 minutes	58.66g
55 minutes	60.58g

25% Sucrose(blue) Initial weight 42.80g

5 minutes	45.39g
15 minutes	46.13g
25 minutes	48.82g
35 minutes	50.98g
45 minutes	52.63g
55 minutes	53.35g

Measurement of osmosis

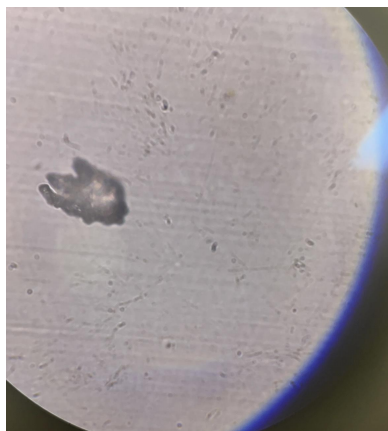


2-G: Measurement of differential permeability of sugar and starch

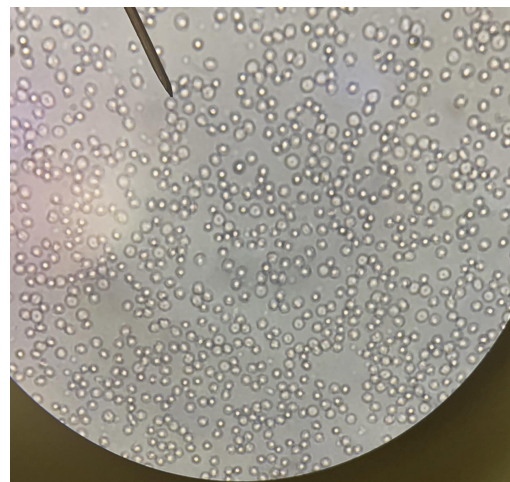
Minutes	Test for starch	Test for Sugar/ Color
15 Min	No starch in water	Blue=No Sugar
30 Min	No starch in water	Yellow=Moderate sugar
45 Min	No starch in water (Reddish)	Orange=More sugar
60 Min	No starch in water (Reddish)	Reddish= Lots of sugar

2-H: The effects of tonicity on red blood cells-Demonstration

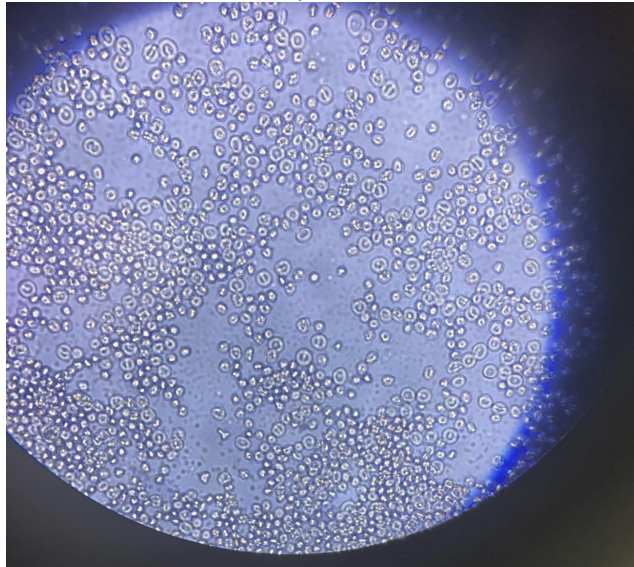
Salt water—2.0% NaCl (hypertonic)



Physiological saline—0.85% NaCl (isotonic)



Di Water: Hypotonic(swollen)



Discussion

2-B: Measurement of diffusion through a liquid

The results for diffusion through a liquid line up with our previous knowledge that as temperature increases so does the rate of diffusion and vice versa, as we can see from our results the temperature with the highest rate of diffusion is the Hot temperature(45C) with a measurement of 30mm after five minutes, The medium temperature(25C) had a measurement of 28mm, and finally we see that the slowest is the cold(5C) solution with a measurement of 25mm after five minutes. These results clearly illustrate the relationship between temperature and rate of diffusion, as we can see in the hot temperature we had the fastest rate of diffusion while the medium temperature was perfectly in the middle of the hot and cold temperatures, while the cold temperature went the slowest. I believe this is because as temperature increases so does the energy and movement of the molecules, causing them to diffuse through the liquid at a faster rate, and as the temperature gets colder the molecules are no longer energized and therefore diffuse slower.

2-C: Measurement of diffusion through agar

The results for diffusion through agar show a trend, we see that the methylene blue, while diffusing, was doing so at a much slower rate compared to the potassium permanganate. This is seen as methylene blue had a much more steady rate of diffusion mostly staying the same until at least 2-3 minutes have passed and stagnated towards the end, while potassium permanganate had a faster and more sporadic rate of

diffusion, I believe this trend shows that the higher the molecular weight, the slower the rate of diffusion. This is shown as methylene blue has a molecular weight of 319.85g/mol with an average diffusion rate of .94mm while the potassium permanganate has a molecular weight of 158.03g/mol and an average diffusion rate of 1.18mm, this clearly illustrates that the more the molecular weight, the slower the rate of diffusion and vice versa.

2-D: Demonstration of filtration

No charcoal passed through the filtration apparatus, the solution with the fastest rate of filtration was the thick solution with an average rate of 116 drops per minute. I believe the driving factor behind the rate of filtration is the mass of the charcoal in the solution, to be exact, the higher the initial mass of the solution, the faster the filtration rate at all levels of fluid(Full, Half, and Near Empty). Another possible factor that affects the filtration rate would be the amount of fluid left in the funnel, the less solution in the funnel, the slower the filtration rate, and vice versa. The results do illustrate these factors because the solution with the highest concentration of charcoal went the fastest when it came to the rate of filtration, also as the fluid levels dropped in the funnel so too did the rate of filtration.

2-F: Measurement of osmosis

We observed that the osmotic rate was faster for the 25% Sucrose (blue) and slower for the 50% Sucrose (red). These results, in our opinion, were obtained because, the higher the weight & sucrose percentage, the slower the rate of osmosis and vice versa.

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

The results show a clear trend, Towards the beginning little to no sugar was present in the liquid but as time went on we can see a steady increase of sugar present in the liquid while starch remained undetected the entire time. I believe this shows that the starch molecules were simply too big to pass through the holes present in the dialysis bag while the sugar molecules were small enough to successfully permeate through.

2-H: The effects of tonicity on red blood cells-Demonstration

The slides show what different tonicities will do to a human blood cell if exposed, first we have the Saltwater—2.0% NaCl (hypertonic) solution, while looking at this image we can see that the cell has shriveled up a died due to it being in a hypertonic solution. Second, we have the Physiological saline—0.85% NaCl (isotonic) solution, this image shows that virtually nothing happened to the cells as they remain in their normal-looking state, I believe this is because there is no net movement of water in or out of the cell due to it being an isotonic solution. Finally, we have the Di Water: Hypotonic(swollen) solution, as we can see in the image, the cells have swollen up and absorbed a lot of the water due to it being a hypertonic solution.

Conclusions

2-B: Measurement of diffusion through a liquid

The results clearly illustrated the relationship between temperature and diffusion rate, as we saw from our results as the temperature increased so did the rate of diffusion, this is because as temperature increases so does the energy, and movement of the molecules, causing them to diffuse through the liquid at a faster rate.

2-C: Measurement of diffusion through agar

The results show that there is a relationship between the molecular weight of a solution and its diffusion rate. The smaller the molecular weight, the faster the rate of diffusion, and the bigger the molecular weight, the slower the rate of diffusion. Essentially lighter molecules move faster and therefore diffuse faster, while heavier molecules move slower and therefore diffuse slower.

2-D: Demonstration of filtration

The results for the Demonstration of filtration were straightforward, it showed a pattern that indicated that the driving factor behind the rate of filtration is the mass of the charcoal in the solution, the higher the initial mass of the solution, the faster the filtration rate. Another trend it showed is that the amount of fluid left in the funnel also affects the rate of filtration, the more solution in the funnel, the faster the filtration rate, and the less solution in the funnel, the slower the filtration rate.

2-F: Measurement of osmosis

Osmosis is concentration dependent as shown by 25% Sucrose went faster while the 50% went slower.

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

Permeability is size dependent, smaller molecules can easily pass through a permeable wall while bigger molecules do not.

2-H: The effects of tonicity on red blood cells-Demonstration

Hypertonic solutions will cause a red blood cell to shrivel up and die due to the presence of too much NaCl and not enough water, hypotonic cells will cause red blood cells to swell up due to excess water being present, and isotonic solutions will cause no net movement of water in or out of the cell leaving them relatively the same.