The Young Scientist Program
Diffusion and Membrane Permeability Teaching Kit
Student Handout

Washington University Medical School

Funding provided by The Leon Lowenstein Foundation
http://ysp.wustl.edu

Today you will be learning about the process of diffusion and its relationship with equilibrium and selective permeability of cell membranes.

Helpful websites to visit and learn more about the topics covered in this lesson:

Diffusion and Osmosis: http://www.biologycorner.com/bio1/diffusion.html
Lesson Plan: http://en.wikiversity.org/wiki/High_School_Biology/Lessons/Lesson_3
Video of Diffusion and Temperature: http://www.youtube.com/watch?v=STLAJH7_zkY
Diffusion Overview: http://en.wikipedia.org/wiki/Diffusion

Equilibrium overview: http://en.wikipedia.org/wiki/Equilibrium
Equilibrium Video (goes a bit more detailed): http://www.youtube.com/watch?v=Gwn9oRvYiu4

Semipermeable membranes (a bit detailed): http://en.wikipedia.org/wiki/Selective_permeability
Semipermeable membrane slideshow lecture: http://www.youtube.com/watch?v=Ya9Vg46ZBPA
Video on iodine experiment: http://www.youtube.com/watch?v=DFhlqgMy1Dk&feature=fvwrel

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Tonicity (good description): http://en.wikipedia.org/wiki/Tonicity
Tonicity (another good overview) http://www.absoluteastronomy.com/topics/Tonicity
Tonicity slideshow lecture: http://www.youtube.com/watch?v=_slUL3kMZlU&feature=related

Goals: There are three main take home messages that you should comprehend by the end of the lesson. You should understand the meaning and importance of diffusion, equilibrium and selective permeability and how they affect living cells.

1. DIFFUSION is a passive (requires no energy), spontaneous (happens on its own) process involving the random movement of particles or molecules from an area of high concentration to an area of low concentration.

2. EQUILIBRIUM across a membrane occurs when the number of molecules is even on both sides, and there is no longer a concentration gradient.

3. SELECTIVE PERMEABILITY across a membrane can result in differing salt concentrations on either side of the membrane and will cause movement of water towards the side with higher salt concentration.

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Answer the following questions to the best of your ability. Your teacher may also record some responses on the board from the entire class. We will then return to these questions after the lesson and see if your answers have changed.

1. **What causes food coloring to spread out when dropped into a glass of water?**

2. **Why do you think Gatorade is better to drink than water when a person is dehydrated?**

3. **Why do plants wilt when dry and why do they become rigid when fully watered?**

4. **Why do your fingers prune after taking a bath?**

Class Experiments:

Materials (per group):
Two cups
Ziploc bag
Eppendorf tubing containing corn starch
Heated water (from preheated beaker)
Tap water
Two gummy bears
15ml tube containing iodine
Food coloring
Dropper
Gloves

Safety:
For the walking around the classroom experiment there is danger of injuring fellow students. WALK SLOWLY AND TAKE CARE NOT TO POKE OTHER STUDENTS WITH YOUR ARMS. Do not drink the iodine! Gummy bears should not be eaten if they have been used in an experiment, but if there are extra unused within the bag they can be consumed. Students should wear gloves for handling the iodine experiment.

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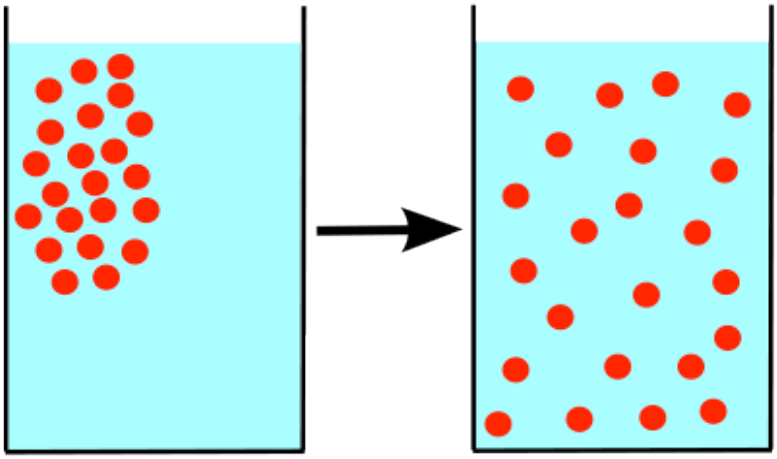
Diffusion

Materials:
Two cups
Tap water
Heating block/microwave
Steaming water
Food coloring
Dropper
Timer

One important physics concept that our cells constantly use in order function properly is diffusion. This section in the lesson will focus on demonstrating the general concept of diffusion. Diffusion is the random movement of particles or molecules from an area of high concentration to an area of low concentration. No energy is required and the process occurs spontaneously. This is illustrated in the picture below. Particles will diffuse until they are evenly distributed, or in other words they will diffuse until equilibrium is reached.



Experimental Procedure and Discussion Questions
The concept of diffusion can be demonstrated with a simple drop of food coloring in a container of water.

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1. Your group should take 2 cups, a tube of food coloring and a dropper. Fill 1 cup with cool water from the tap up to the marked line, and have the other cup filled with heated water from the teacher.
2. Drop 2 drops of food coloring into the cups of water at the same time.
3. Observe what happens to the drop of food coloring. The drop should spread out from a single concentrated drop of color to eventually evenly color the entire container of water. This will happen rapidly at first and then slow down. One member of your group should time how long it takes for the water to become equally colored throughout by the food coloring, while other members should record the time for each. Be sure not to stir the water.

This is diffusion at work right before your eyes! The tiny colored molecules that make up the food coloring diffuse through the water from an area where they were highly concentrated to an area where there was previously no color.

Q: A: Describe what happened to the food coloring after being dropped into the water.

B: Does the speed of the color spreading out change over time? If so, why?

C: Can you think of something you could add to the cup to prevent the color from diffusing throughout the entire cup?

D: Does hot water make diffusion occur faster or slower? What about cold water?

Equilibrium

Materials:
6-20 students
Lab tape or arranged desks/chairs
Timer or classroom clock to count time

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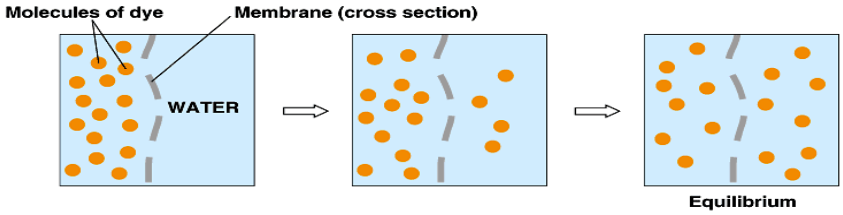
Two of the most fundamental concepts that drive cell function in your body are the ideas of **equilibrium** and **concentration gradient**. These concepts are the simplest ways in which cells in the body have the potential to do work that help them survive. A solution is when a type of molecule is mixed or dissolved in a solvent (like salt or sugar dissolved in water). To set up a concentration gradient you need at least 2 solutions and a sort of barrier (**semi-permeable membrane)** that allows molecules of a certain size to cross freely. For example, Solution A is a cup of water with 100 molecules of salt, while Solution B is a cup of water with 0 molecules of salt; Solution A is more concentrated than Solution B. Now imagine that both solutions are poured into separate halves of the same cup and are divided in the middle by a membrane, which only allows molecules of a certain size to pass across. This sets up a **concentration gradient,** where salt molecules are more packed together on one side of the membrane compared to the other. A chemical property called **equilibrium** dictates that the molecules will want to diffuse in a direction across the membrane such that over time, there is the same amount on both sides. So Solution A's salt molecules will want to move to the other side to Solution B's space, where it is less crowded with salt, and eventually there will be 50 molecules of salt in Solution A and 50 molecules in solution B. At this point, because there is no more difference in salt concentration between the two sides, the concentration gradient does not exist anymore. This situation is illustrated in the picture below.



Experimental Procedure and Discussion Questions

For this experiment, you and your fellow students will demonstrate the concepts of concentration gradient and equilibrium.

1. The classroom will be divided in half with either desks/chairs or tape serving as the barrier. These will represent a cell membrane.

2. If you are selected, stand on one side of the room as directed by your teacher with other students. Extend out your arms and walk SLOWLY around your half of the room **without bumping into others** for 30 sec (your teacher will count). You cannot cross over to the other side. Try to walk as far apart from others as possible. This represents the concept of a concentration gradient because one side of the room is more concentrated with students than the other side.

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Q: A. Was it easy to move around without bumping into each other?

B. Would it be easier to walk around if you were able to use the entire room?

C. If given a choice, how many students should be crossing over to the other side, such that it is equally easy on both sides of the room to walk around, arms extended, without bumping into each other?

3. You will now repeat the experiment starting on the same side, but now you have the option of crossing over to either side as many times as you wish in order to spread out as much as possible. Again, try to maximize the distance between you and others, use the entire available area.

Q: D. What happened to the crowd initially when you were allowed to move around the entire room?

E. When time expired, were there a similar amount of students on both sides of the room? Was this expected?

F. Beginning from an equal number of students on each side, if 2 students moved to the opposite side of the room, would you still be at equilibrium? What would need to happen to restore equilibrium?

Molecules are constantly in motion and cross one barrier, come back, and cross over. Just because there is equilibrium doesn't mean the molecules have stopped moving, it only means that the relative number of molecules on either side at any given time does not change.

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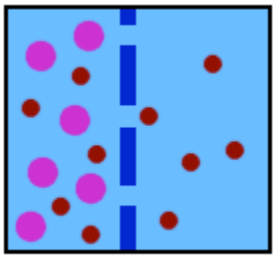
Selective Permeability/Tonicity – estimated time 15-20min (plus overnight)



Materials:
One cup
Steaming hot water
Heating block/microwave
Eppendorf tube containing corn starch
15ml tube containing iodine
Ziploc bag
Straw for stirring

Cells are surrounded by a membrane that keeps cell material in and unwanted material out. The cell membrane only allows movement of certain materials across, making it **selectively permeable.** Selective permeability allows our cells to maintain proper concentrations of needed molecules and ions by controlling what is allowed to enter or exit the cell. Cell membranes may restrict movement by requiring a specific electric charge, size, or interaction with proteins on the cell surface.

Experimental Procedure and Discussion Questions
In this exercise, we will explore examples of selective permeability and its effect on tonicity. In our experiment we will be demonstrating selective permeability based on size selection. A Ziploc bag containing iodine will be placed into a cup of water containing dissolved/suspended corn starch. Starch is a large molecule containing many atoms while iodine much smaller molecule. Starch turns purple when it becomes in contact with iodine, so we will be able to determine the presence of starch and iodine both outside and inside the tubing by color indication.

The Ziploc bag represents a selectively permeable membrane and will only allow very small molecules to move across.

1. Each group should have a Ziploc bag and a 15ml tube containing iodine. You should pour the iodine into the bag and close it tightly, ensuring that the bag is fully sealed before use. One member of your group should fill the cup with hot water from the teacher.
2. You should then add corn starch (in the small tube) to the water and mix until the starch is evenly distributed and dissolved.
3. Place the ziploc bag in the water and wait a few minutes.

Q: A. Do you think the liquid will turn purple? Where, inside or outside the bag?

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B. What possible factors may affect the speed of the reaction?

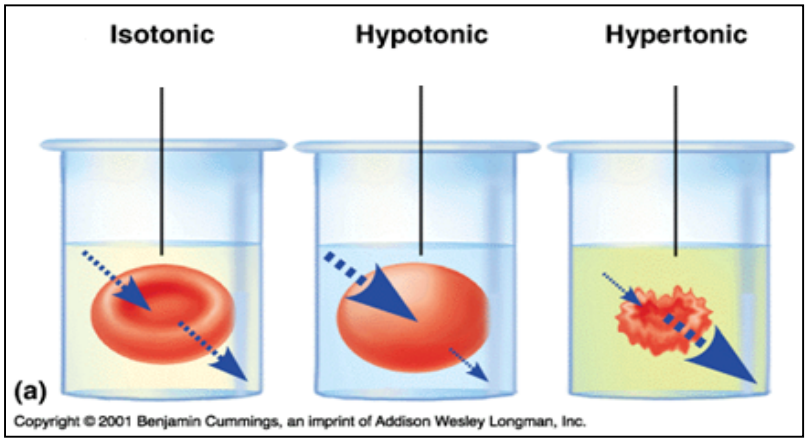
After the experiment:

Q: C. What did you observe inside the bag and outside the bag?

D. Of the starch and the iodine, which is able to pass through the membrane (plastic bag) and what is not allowed to pass through? Why?

E. Based on your knowledge of the selective permeability of starch and iodine, explain the reason for the differences in color inside and outside the bag.

Similar to other molecules such as proteins and salts, water also moves across a membrane down its concentration gradient. The movement of water across a cell membrane is known as **osmosis.** Selective permeability can result in an increased concentration of larger molecules such as proteins



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and sugars on one side of the membrane since they are too large to pass through. Because the concentration of large molecules is higher on one side of the membrane, the relative concentration of large molecules on the other side is less and the overall solution is not at equilibrium. This will cause the influx of water from the less concentrated side over to the more concentrated side in order to equilibrate the concentration of the salt on both sides of the membrane.

If the solution outside a cell has less solute (ions, salt particles, large molecules etc) than inside the cell the solution is known as a **hypotonic solution**, while a solution with more solute than the cell is a **hypertonic solution.** A solution of equal solute concentration to the inside of the cell is an **isotonic solution**. A hypotonic solution will cause the cell to swell as water enters to equilibrate the solute concentration while a hypertonic solution will cause the cell to shrivel as water leaves.

4. We will now use gummy bears to simulate the effect of a cell placed in a hypotonic solution. This will require overnight incubation in water to observe the full effect. Fill one cup with cool tap water and leave the other one empty. Place one gummy bear in each and leave overnight to compare size between the gummy bear with and without water.

Materials:
Two cups
Cool tap water
Two gummy bears

Q: D: What do you think will happen to the gummy bear in water?

The next class, slowly pour out the water from the cup without having the gummy bear fall out. Observe differences between the two gummy bears as you answer the questions below.

Q: E: What happened to the gummy bear? Did it grow or shrink, and did its appearance change? What caused this change in size?

F: Is the water hypotonic, hypertonic or isotonic relative to the inside of the gummy bear? How does this determine the direction of water flow?

G: What do you think would happen if the gummy bear was placed instead in very salty water? Why?

Conclusions

Return to the questions from the beginning of the lesson, have your answers changed?

1. **What causes food coloring to spread out when dropped into a glass of water?**

2. **Why do you think Gatorade is better to drink than water when a person is dehydrated?**

3. **Why do plants wilt when dry and why do they become rigid when fully watered?**

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Final Evaluation

Fill in the blank…. (use concentration gradient, diffuses, equilibrium, into, out of, increase, shrink, water, selectively permeable, hypertonic, isotonic, hypotonic, osmosis).

The passage below will utilize all vocabulary learned throughout the lesson to explain the final experiment.

When a gummy bear is placed in pure water, water flows _____ the gummy bear, causing it to _____ in size. The surface of the gummy bear acts as a _____ membrane, blocking movement of large molecules and only allowing movement of small molecules such as_____. The pure water solution is therefore a(n) _____ solution. If the gummy bear is placed in very salty water, water moves _____ the gummy bear down its _____, causing the bear to _____. The water _____ freely across the membrane through a process known as _____ until it has reached _____. The salt water is therefore a(n) _____ solution, while a solution of equal salt concentration as the inside of the gummy bear is a(n) _____ solution.

Please fill out the evaluations so that we can improve this lesson for the future. Hopefully this improves your understanding of diffusion and membrane permeability!

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