ONLINE INQUIRY & INVESTIGATION

French Fries, Dialysis Tubing & Computer Models: Teaching Diffusion & Osmosis Through Inquiry & Modeling

PATRICIA MEIS FRIEDRICHSEN AMY PALLANT

Can eating large amounts of sugarless candy and gum lead to diarrhea? How does Milk of Magnesia® work? How does penicillin kill bacteria? These are some of the questions that students answer in this series of lessons on diffusion and osmosis. To begin the unit, students predict the results of a demonstration involving the relationship between soaking French fries in salt water and crispiness. To understand this relationship, students investigate the permeability of cell membranes by designing their own investigations using dialysis tubing. This investigation is a revised, inquiry version of the traditional lab found in many high school textbooks. To visualize the movement of molecules and the resulting effect on the cellular level, students use the innovative molecular modeling software, *Molecular Workbench* (available online at http://molo.concord.org at no cost). In the *Molecular Workbench* activities, students interact with dynamic computer models of diffusion, osmosis, and 3-D representations of molecules; observe the interactions and the net flow of molecules in air, in cells, and across a cell's semi-permeable membrane; and learn about the properties of the molecules. Students then apply their knowledge of osmosis to the novel situations listed above.

Student Difficulties

Osmosis is a critical process in living cells. However, many students struggle to understand diffusion and osmosis. Students tend to have a static view of molecules. They are often confused by which molecules are moving across the cell's semi-permeable membrane and the overall net movement of these molecules. It has been our experience that students tend to memorize rules, such as cells in hypotonic solutions take in water, but are unable to explain why. To develop a conceptual understanding of osmosis, students need to visualize the movement of individual molecules and to predict the effect at a cellular level.

PATRICIA MEIS FRIEDRICHSEN is Assistant Professor, Biological Sciences, and Learning, Teaching & Curriculum, University of Missouri-Columbia, Columbia, MO 65202; e-mail: FriedrichsenP@missouri.edu. AMY PALLANT is Senior Science Education Researcher, The Concord Consortium, Concord, MA 01742; e-mail: apallant@concord.org.

5E Instructional Model & Inquiry Standards

To support students in developing conceptual understandings of diffusion and osmosis, we sequenced these lessons using the 5E Instructional Model. In comparison to the traditional lecture followed by lab format, this model is based on current learning theory and more closely follows the natural inquiry of students as well as the formal processes of scientific inquiry (Bybee, 2002). The 5E Model consists of the following sequential phases: engagement, exploration, explanation, elaboration, and evaluation. The purpose of the engagement phase is multi-fold: (a) to elicit students' prior understandings and misconceptions, (b) to focus the students' attention on the new concept and (c) to provide motivation for the lessons that follow. In the exploration phase, students engage in hands-on activities in which they actively explore the phenomenon and collect data. In the explanation phase, students use their data to create evidence-based explanations of the phenomenon. Students apply their new understandings in the elaboration phase. Although informal, ongoing assessment occurs throughout the lessons, the evaluation phase involves a summative assessment of the students' understanding of the phenomenon. For a list of the specific content objectives for each phase in this unit, refer to Table 1.

This series of lessons align with the *National Science Education Standards* (National Research Council [NRC], 1996). The mini-unit incorporates the five essential elements of inquiry in which students:

1. engage in scientifically oriented questions
2. give priority to evidence
3. formulate explanations based on evidence
4. connect explanations to scientific knowledge
5. communicate and justify their explanations (NRC, 2000).

Table 1. Educational Objectives.

5E PHASE	EDUCATIONAL OBJECTIVES
Engage	• To generate interest in diffusion and osmosis. • Elicit prior knowledge by having students predict the outcome of the potato demonstration. Students create drawings to explain their predictions.
Explore	• Students design and carry out an investigation to determine the permeability of dialysis tubing to glucose, starch, and water. • Students collect data, organize data, and write evidence-based conclusions about the permeability of dialysis tubing. • Students communicate their results and conclusions to other class members.
Explain	• Using *Molecular Workbench*, students explore the diffusion of particles from an area of higher concentration to an area of lower concentration. • Using *Molecular Workbench*, students vary the solute concentrations inside the cell and/or its surrounding environment to explore the net movement of molecules and ions across a cell membrane, as well as the resulting effect on the cellular level. • Using the evidence from the dialysis tubing investigation and *Molecular Workbench*, students create evidence-based explanations for the potato demonstration. • Students engage in peer review of their evidence-based explanations.
Elaborate	• Students apply their understandings of osmosis to the following situations: effect of penicillin on bacteria, effect of Milk of Magnesia®, and effect of over-consumption of sugarfree gum and candies.
Evaluate	• Students apply their understandings of osmosis to explain the effect of 10% NaCl solution on red onion cells.

Engagement Phase

Which fast food restaurant has the best French fries? We pose this initial question to stimulate student interest. French fries are a beloved food of high school and college students and this question brings a ready response. Next, we share with the students the results of an Internet search for French fries recipes. Many of the recipes suggest soaking the raw, cut potatoes in cold water before frying. Several recipes indicated that soaking removes excess starch in the potato, resulting in crispier fries. We ask the students to help us investigate this claim and pose the following question:

What happens to raw French fries when soaked in water?

Additionally, the recipes suggest salting the fries after cooking. We ask the students if it would be possible to combine these two steps by adding salt to the water, and if they think the amount of salt would make a difference.

To investigate these questions, we set up a demonstration using three 500-ml beakers and raw potatoes. Beaker A contains distilled water, Beaker B contains a 0.9% NaCl solution, and Beaker C contains a 10% NaCl solution. Next, we slice several potatoes into French fry size pieces. (Do not include any potato skin on the pieces.) We record the initial mass of each French fry before placing one French fry into each beaker. The students predict the results and make drawings to explain their predictions. We collect the students' work, using it to assess students' prior knowledge. Typically, students' drawings inaccurately predict the results and do not illustrate molecular-level representations. We begin the engagement phase, which takes approximately 20 minutes, at the end of the class period and allow the demonstration to set overnight. At the end of this short phase, we have generated interest in osmosis and gathered data on students' initial understandings of molecules, cells, and osmosis.

Exploration Phase

At the beginning of the next class, students observe the results of the potato demonstration. The potato in Beaker A (distilled water) increased in mass and turgidity. In Beaker B (0.9% NaCl), there is little change in the potato's mass or turgidity. The potato in Beaker C (10% NaCl) decreased in mass and turgidity. We pose the following question:

What's happening on the molecular and cellular levels in each of these potatoes?

We explain that scientists often use models to help them understand complex processes. We introduce the students to dialysis tubing as a model of a semi-permeable plasma membrane. We ask students to investigate if the dialysis tubing is permeable to glucose, starch, or water.

We modify the traditional cookbook lab found in many high school texts by making the investigation more inquiry-oriented. First, we pose the driving question:

Which of these substances will pass through the dialysis tubing: glucose, starch, or water?

We have students make predictions and provide them with a variety of lab materials to test their predictions, including 12 cm lengths of dialysis tubing soaked in water, small beakers, test tubes, test tube racks, funnels, microscope slides, eye droppers, dental floss, tape, rubber bands, and electronic balances. (We use 1 5/16" wide dialysis tubing.) We also provide a 10% glucose and a 10% cornstarch solution. For indicators, we demonstrate how to use a tincture of iodine (2%) solution as a starch indicator. To test for glucose, we use diabetic testing strips designed to determine glucose concentrations in urine. We suggest that the dialysis membrane could be used as a cell by tying one end closed or the tubing could be cut along its length, creating a larger, flat piece of membrane.

Students form small groups of two to four to plan their investigations. Some students use the dialysis tubing to create a "cell," tying one end of the tubing closed with the dental floss and filling the cell with one or more of the stock solutions. Some students use a flat section of the dialysis membrane to line the inside of a funnel, pouring various solutions through the funnel and testing the filtrate. If any particular group struggles with its experimental design, we suggest that it talk with other groups. Across the student groups, the experimental designs vary considerably.

A typical class is able to plan and set up its investigations within one 50-minute class period. As teachers, we do not give students feedback on their designs, allowing them to experiment and make mistakes. Depending on their experimental design, some students begin collecting data during the same class period, while the majority of the students let their investigation set overnight, collecting data at the beginning of the next class period.

Each small group is given a large sheet of paper and markers to design a data table and record its results. Each group is asked to formulate claims about the permeability of the dialysis tubing and to support its claims with evidence. We have each group show its experimental setup and present its claims and evidence (data table) to the entire class. See Table 2 for a typical example of claims and evidence.

Occasionally, students share results that conflict with other groups' results. It's not unusual for one group to claim that the dialysis membrane is permeable to starch. We don't use our authority as instructors to say that these results are inaccurate; rather we welcome this opportunity to model the norms of science. The class is encouraged to ask questions about the group's experimental design. If the string is not tied tightly to close off the end of the dialysis tubing, starch can leak out of the end of the "cell," resulting in a false positive test for starch. Students are quick to pick up on this experimental error and suggest modifications to the group's design. Likewise, students might get a negative test for glucose if they place the glucose-filled cell in a large volume of water. Because of the extremely low concentration of glucose in the solution in the beaker, the test strips indicate a false negative result. Again, other students point out experimental design errors. We feel it is important for students to grapple with discrepant data because it stimulates important discussions about experimental design. Groups that obtained discrepant data (usually only one or two groups) are asked to make modifications to their experimental design and re-test the permeability of that particular substance. We also ask one to two groups whose data agreed with the class to repeat their procedure to see if their results are reproducible. It takes very little class time for these groups to work independently, re-testing the substance in question. However, we feel that it is critical to model the norms of science by having students make modifications to their experimental design and/or attempt to replicate their results.

Students usually collect data and share their results in one class period. Students are interested in seeing other groups' experimental designs and comparing their results. Because most (if not all the small groups) have the same results, the reporting of the results goes very quickly. After each group has presented its data from the dialysis investigation, we move to the explanation phase.

Explanation Phase

In this phase students make sense of the data they collected during the exploration phase. A common misinterpretation of this phase is for the teacher to explain the data. However, to validate the students' investigations and to foster student learning, it's important that students create and share their evidence-based explanations. Later, the teacher may need to add to or re-enforce concepts. To help students develop molecular-level explanations for their findings, we use the Web-based *Molecular Workbench* activity, "Cell Membranes – Diffusion and Osmosis," http://molo.concord.org/database/activities/72.html.

Recent advances in computational power and software have made it possible to create molecular dynamics models that are sufficiently powerful and interactive to simulate some of the complex phenomena in biology. The *Molecular Workbench* is a science-based learning environment built around a set of molecular dynamics engines. Because the underlying model is based on good approximations of physical laws, *Molecular Workbench* can produce emergent phenomena such as phase change, diffusion, osmosis, solubility, chemical equilibrium, self assembly, and biomolecule conformation. Using this software, students can visualize otherwise abstract phenomena, experiment with the model, and reason about the relationship of atomic-scale interactions and macroscopic phenomena in a structured environment. The *Molecular Workbench* software currently contains several hundred models and model-based activities.

The *Molecular Workbench* activity, "Cell Membranes—Diffusion and Osmosis" draws students into the molecular world of diffusion and osmosis. The activity begins by providing the students with a representation of a red blood cell's membrane. Students explore how water and dissolved materials move between a cell and its external environment. Students start with a simple model of a few molecules moving in a container in order to become familiar with molecular movement and this atomic-scale representation. Next they explore a model of diffusion to discover how the interactions of the molecules (collisions) result in the distribution of molecules over time. Students experiment with the dynamic model to investigate the role of pores in a cell. Through this investigation, we expect students to develop a sense of how a semi-permeable membrane allows some molecules to cross while others do not. Students explain why,

Table 2. Examples of Evidence-Based Claims.

CLAIM	EVIDENCE
Starch does not move through the dialysis membrane.	Negative starch test
Water moves through the dialysis membrane.	1. Increased mass of "cell" when placed in distilled water. 2. Decreased mass of "cell" when cell is left on lab table and water puddles on lab table.
Glucose moves through the dialysis membrane.	Positive glucose test

Figure 1. *Molecular Workbench* activity: Role of pore size.

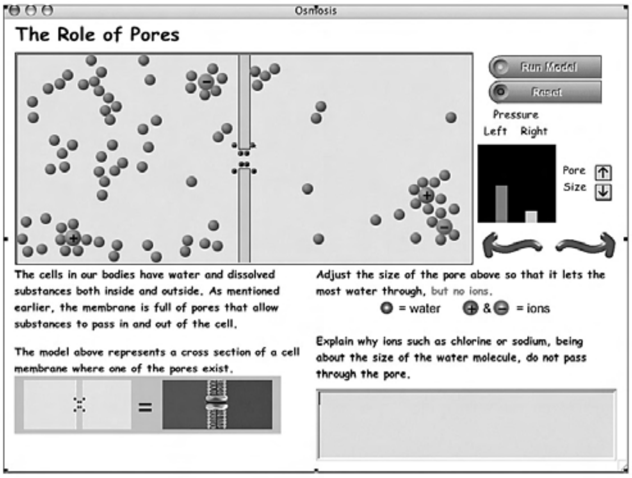


Figure 2. *Molecular Workbench* activity: Exploring osmotic pressure.

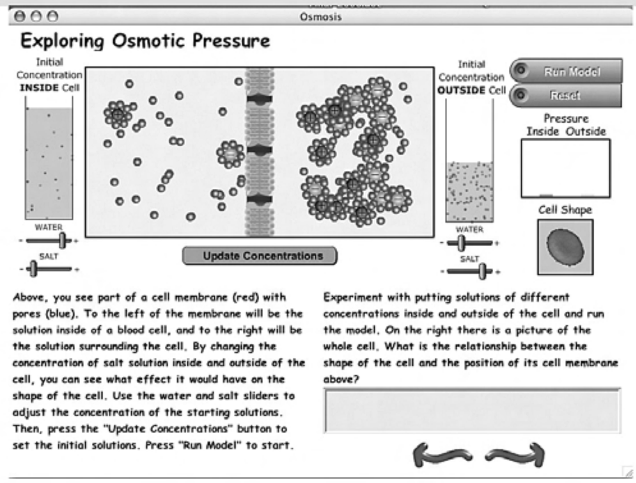
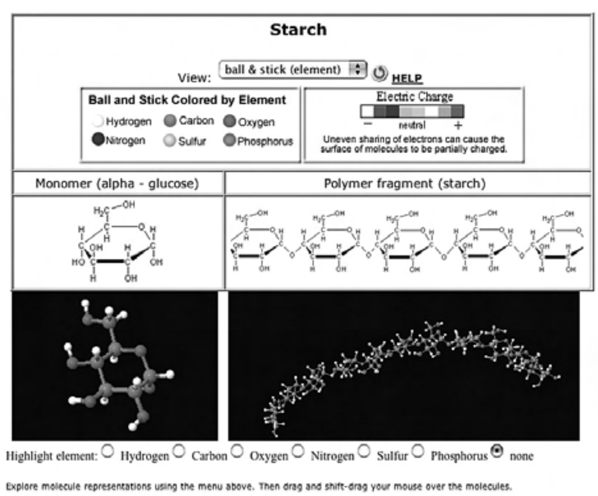


Figure 3. Tree of Life's Molecules activity: Comparison of glucose and starch molecules.



for example, chlorine or sodium ions (about the same size as water molecules) are unable to move passively through a pore. Students change the pore size and observe how water shells form around the ions, making the aggregate too big to pass through the pore, whereas single water molecules can move through the pore. (See Figure 1.) Finally, students experiment by varying the concentration of solutes inside and outside of the model cell membrane. They observe the movement of molecules and the distortion of a cell membrane, and relate it to the macroscopic properties of the shape of the cell. This is depicted as the red blood cell swells and shrinks as the solute concentrations in the model change. (See Figure 2.)

This activity requires approximately 20-30 minutes and works best when two students work together at a computer. However, the model can be run by individuals or as a demonstration. The teacher could project the activity and have the students make predictions regarding expected outcomes of the model, discuss ways to change variables, and, as a class, discuss responses to the assessments or have students record their own answers.

In typical biology textbooks, diffusion and osmosis are depicted by static images. When students rely on these images, they often fail to grasp many of the following concepts: all particles are in constant motion; diffusion results from random motion and/or collision of particles; diffusion is the net movement of particles from areas of high concentration to low concentration; and movement of particles continues even when the particles are uniformly distributed. These activities along with many other *Molecular Workbench* activities have been tested in classrooms across the country. Evaluations of the activities, as part of the NSF grant, have shown to increase student understanding of a variety of concepts significantly ($p < .01$) (http://workbench.concord.org/research/ http://molo.concord.org/research). In this case, after using the *Molecular Workbench* activities, students' explanations regarding cell turgor are based on the movement of particles and their interactions. This understanding is dynamic and students are able to easily transfer their new understandings to novel situations.

In addition to the "Cell Membrane–Diffusion and Osmosis" *Molecular Workbench* activity, students view 3-D representations of glucose and starch molecules by accessing the "Tree of Life's Macromolecules" activity: http://molo.concord.org/database/activities/226.html. (Students are instructed to "zoom" down from leaves on the Tree of Life to the molecular view of carbohydrates.) Students compare 3-D models of glucose and starch. By interacting with the 3-D representations of molecules, students become familiar with the properties of these molecules, including their structure, relative size, polarity, and electrostatic charges. They reason about the ways in which molecules interact with other molecules. Specifically for this lesson, the students focus on the relative sizes of glucose and starch molecules to help them understand why dialysis tubing is permeable to glucose, but impermeable to starch.

After students complete the two *Molecular Workbench* activities, we return to the original questions posed in the engagement phase.

What happens to raw French fries when they are soaked in water?

What happens to raw French fries when soaked in 0.9% NaCl and 10% NaCl solutions?

Table 3. Potato Demonstration Evidence-Based Explanations.

BEAKER #1. FRENCH FRY IN DISTILLED WATER	
Claim #1	Water molecules can pass through the potato cell membrane.
Evidence:	1. Water moved through the dialysis tubing in the lab.
	2. *Molecular Workbench* Activity with water and red blood cells.
Claim #2	The net movement of water molecules is **into** the potato cells because water is more concentrated in the beaker than in the potato cell.
Evidence:	Mass of French fry increased.
Claim #3	Starch molecules do not diffuse out of the potato cells.
Evidence:	1. 3-D molecules showed that starch is a very large molecule, too large to fit through cell pores.
	2. Negative starch test in Dialysis Tubing Investigation
Claim #4	Potato cell (and central vacuole) volume increases and pushes against cell wall, making the potato cell more turgid.
Evidence:	Class notes on plant cells from Cell Unit

BEAKER #2. FRENCH FRY IN 0.9% NaCl SOLUTION	
Claim #1	In Beaker B, some water molecules moved into the potato cells, while some water molecules moved out. The net movement of water molecules is zero.
Evidence:	1. *Molecular Workbench* Activity with water and red blood cell
	2. Mass of French Fry stayed the same.
Claim #2	The NaCl ions did not move into the potato cells
Evidence:	*Molecular Workbench* Activity with NaCL and red blood cell

BEAKER #3. FRENCH FRY IN 10% NaCl SOLUTION	
Claim #1	The NaCl ions do not move into the potato cells.
Evidence:	*Molecular Workbench* with NaCl ions and red blood cell
Claim #2	Water molecules were more concentrated inside the potato cell, so the net movement of water was out of the cell.
Evidence:	*Molecular Workbench* with water, NaCl and red blood cells
Claim #3	Potato cells (and central vacuole) lose water and become less turgid.
Evidence:	Potato demonstration Beaker 3, and notes from Cell Unit

We ask students to review their original predictions and revise their explanations to include molecular and cellular level representations. Within their small groups, students discuss how their understandings have changed based on the dialysis tubing investigation and the *Molecular Workbench* activities.

To demonstrate their new understandings of the potato demonstration, each small group is given a large sheet of paper to make new sketches of the potato demonstration. For each beaker, students are asked to draw one to two enlarged potato cells and include representations of water molecules, starch molecules, and NaCl ions. Students should indicate the net movement of particles across the potato cell membrane. The drawings should indicate that starch molecules stay inside the potato cells, while Na and Cl ions remain outside the cell in solution. In Beaker A (distilled water), the net movement of water molecules is into the potato cells. In Beaker B (0.9% NaCl), an equal amount of water molecules enter and leave the potato cell, and in Beaker C (10% NaCl), the net movement of water molecules is out of the potato cells into the environment.

If time allows, we ask students to write an evidence-based explanation for the potato demonstration results. In the explanation, students make claims about what is occurring at the molecular and cellular levels, supporting their claims with evidence from the dialysis tubing investigation and *Molecular Workbench* activities. For example, students might claim that starch molecules cannot move out of the potato cells. To support this claim, they use results from the dialysis tubing investigation and the 3-D models. (See Table 3 for an example of a written, evidence-based explanation.) We circulate around the room, asking probing questions when necessary. If class time is limited, we eliminate the written explanation and move to peer review of the posters.

We use the peer review process to model scientific norms and to give students an opportunity to clarify and revise their explanations based on peer feedback. We assign two small groups to work together. Each small group explains its poster while the other group offers feedback. The peer review process takes approximately 10-15 minutes. Next we select two to three small groups to explain their posters to the class, allowing us to facilitate a whole class discussion. Once we are certain that students understand that particles move from areas of high concentration to areas of low concentrations, we introduce the terms: hypertonic, isotonic, and hypotonic. These terms refer to the amount of solute in solution, either in the surrounding environment or in the cell's cytoplasm. If the cell's external environment is hypertonic (in comparison to its internal environment), then the net movement of water molecules will be out of the cell. Students add these new labels (hypertonic, isotonic and hypotonic) to their sketches. Students are given additional time if they want to make revisions to their posters, based on peer feedback and whole class discussion, prior to grading.

Elaboration Phase

In this phase, students apply their knowledge of osmosis to new contexts, thus strengthening their conceptual understandings. We ask students to research one of the following questions:

- How does penicillin kill bacteria?
- What happens when you chew too much sugarless gum (containing sorbitol) or sugarless candy?
- How does Milk of Magnesia® work?

We have students sign up to research one of these questions, so that there is an equal distribution of students across the three questions. As a homework assignment, each student writes a one-page explanation for his/her question, using Internet resources.

Penicillin inhibits an enzyme that produces chemical cross-links in gram-positive bacterial cell walls. The cell walls become weaker until they break, and the bacterium ruptures, due to osmotic lysis (Tortora et al., 2002). (For an online reference for students, see http://helios.bto.ed.ac.uk/bto/microbes/penicill.htm.) Sugarless gum and candies contain sorbitol, a polyalcohol sugar that is neither digested nor absorbed in the small intestine. Sorbitol passes into the colon where it drives an osmotic purge drawing fluids, mostly water, from the surrounding tissue (http://www.merck.com/mrkshared/mmanual/section3/chapter27/27b.jsp; http://www.foodintol.com/food_intolerance/hot_ibs.htm). Milk of Magnesia® is a saline osmotic laxative containing magnesium hydroxide which is poorly absorbed in the intestinal tract. The concentration of magnesium hydroxide draws water from the surrounding tissue by osmosis and retains fluids already within the intestine (http://www.gicare.com/pated/milk_of_magnesia.htm http://www.answers.com/topic/milk-of-magnesia).

The next day we use a jigsaw discussion technique. At the beginning of class, students who researched the same topic form small groups known as expert groups. Within expert groups, students compare explanations and reach a common understanding of how osmosis applies to their question. They also create drawings illustrating the osmotic effect on a cellular level. During the small group discussion, we circulate around the classroom, informally assessing the students' understanding and helping them work through any difficulties and/or misconceptions. For the second part of the jigsaw activity, new groups are formed consisting of an expert for each of the three questions. Each expert teaches the osmosis application he/she researched. If necessary, we follow up the jigsaw activity with a whole class discussion emphasizing the key concepts. The elaboration phase generally takes less than one 50-minute class period.

Evaluation Phase

The purpose of the evaluation phase is to formally assess students' understanding of the phenomenon, particularly when applied to a novel context. For this phase, we use the example of plasmolysis in red onion cells. The presence of the anthocyanin pigment in the epidermal cells contributes to a good visual illustration of osmosis. We show students a video clip of red onion cells in distilled water. (We capture the video clips before class, using a Boreal® microscope and Motic Images® software.) We orient the students to the image by pointing out the following plant cell structures: cell wall, plasma membrane, and large, central vacuole containing the red pigment. We point out that the central vacuole almost completely fills the interior of the cell, resulting in uniform, light pink color. Next we show students a video clip of the same red onion cells as the cells are being flooded with a 10% NaCl solution. We ask students to write an explanation of the cellular changes, and sketch of one to two onion cells indicating, with arrows, the net movement of molecules across the plasma membrane. (When the red onion cells are flooded with 10% NaCl, the net movement of water molecules is out of the cell. The central vacuole shrinks in size, concentrating the red pigment, resulting in patchy, dark red areas inside the cell.)

Conclusion

Although this series of lessons may take more time than what is typically allotted, we feel the amount of time is justified based on students' increased conceptual understanding of diffusion and osmosis. We assess student understanding by comparing their initial, written predictions and explanations in the engagement phase to their written and oral explanations in the explanation, elaboration, and evaluation phases. In the latter two phases, students are able to accurately predict and explain the net movement of molecules in novel contexts. Many of our students (prospective biology teachers) readily admit that in their previous high school and college biology courses they memorized the terminology (iso-, hypo-, and hypertonic) without having a conceptual understanding at the molecular and cellular level. Students find the *Molecular Workbench* models to be powerful tools that enhance their conceptual understanding of diffusion and osmosis. Additionally, the series of lessons model the work of scientists by incorporating the five essential features of inquiry (NRC, 2000). We have found that students develop a deep conceptual understanding of diffusion and osmosis that they continue to draw upon throughout the year.

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