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Modeling Diffusion

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In order to increase students' awareness for and comfort with mathematical modeling of biological processes and increase their understanding of diffusion, the following lab was developed for use in 100-level, majors/non-majors Biology and Neuroscience courses. The activity begins with generation of a data set that uses coin-flips to replicates movement of dye molecules at an interface of a permeable gel. The class results then are collapsed into a single data set that is then used to predict the movement of real dye molecules over time, which are then measured by students in a "wet-lab" activity.

Keywords: diffusion, predictive, modeling, interdisciplinary

Introduction

In order to increase students' awareness for and comfort with mathematical modeling of biological processes and increase their understanding of diffusion, the following lab was developed for use in 100-level, majors/non-majors Biology and Neuroscience courses. Diffusion was selected for two reasons. First, we've found that students often have a difficult time discarding misconceptions about the way molecules move under this process. This observation is certainly not unique to our institution and was recently discussed by Meir et al. (2005) and Fisher et al. (2011). Second, the random motion that molecules undergo when they diffuse can be modeled easily with so-called "stochastic" models, i.e. those involving random number generators. Stochastic modeling can be implemented with little or no mathematical background, thus making it suitable for introductory classes with minimal mathematical pre-requisites.

Our approach was to build on the pedagogical principle that active-learning involves students more deeply in their own learning of physical processes (Meltzer and Thornton, 2012). Thus, we wanted our students learn how random motion drives diffusion by (1) being actively involved in generating trajectories of several molecules undergoing random motion and (2) by relating those trajectories to diffusion. To that end, the lab begins with a coin-flip activity designed to replicate the random movement of dye molecules as they diffuse across a permeable gel. The class results (240 data points per student) are collapsed into a single data set that is then used to predict the movement of real dye molecules over time. Students then complete a "wet-lab" in which they measure actual diffusion of food coloring into a set gelatin interface. As the model doesn't account for all variables, the students' predictions inevitably deviate from the observable diffusion. This allows for class discussions about the strengths and weaknesses of mathematical models and fundamental properties of diffusion.

In 2010, The American Association for the Advancement of Science (AAAS) released a document entitled Vision and Change in Undergraduate Biology Education: A Call to Action. http://visionandchange.org/files/2011/03/VC-Brochure-V6-3.pdf. This document outlines recommendations to change current biology pedagogy to include a greater understanding of the scientific process, the interdisciplinary nature of biology, and the integration of science within society. In particular the AAAS report stresses that students completing program in biology should have experience with modeling, simulations, computational and systems-level approaches to biological discovery. In trying to implement these concepts, our departments evaluated our current teaching practices and observed a real deficiency when it came to the mathematical modeling of biological concepts. Working with three different disciplines (biology, neuroscience, and biophysics) we developed this lab to deal with the misconceptions listed previously as well as introduce students to an interdisciplinary perspective through mathematical modeling.

Student Outline

Diffusion Lab, Part 1

DUE next class period **Name**_____

To help you understand how molecules move, we will develop a mathematical model that represents the diffusion of dye molecules into a gel. We will then use that model to try and predict actual experimental data from a lab we'll run next class period. Why would a scientist want to use a model rather than actually observe a system? In many cases it's not really practice (or even possible) to directly watch all components of a system. In our case, this would require tracking the millions of water and dye molecules as they randomly bump into each other over a 48 hour period. We simply can't do this. However, we can develop a model of the system that approximates this movement. Once we have this model, we can then compare it to the actual system. If our model is incorrect (the predicted values don't match those of our experiment) this tells us that we didn't account for all variables and need to refine the model.

The first step of this process requires that we collect some data to base our model on. **Your assignment** is to track the movement of 10 "molecules" over "120 minutes" of time. To do this, you will be flipping two coins 240 times. The outcome of each coin flip represents movement a molecule into (two heads, +0.5mm) or away (two tails, -0.5mm) from the gel. As the molecules could also move along the interface, an outcome of one head and one tail should be recorded as 0mm movement.

Molecule	1	2	3	4	5	6	7	8	9	10
Time 5m	*									
10	*									
15	*									
20	*									
25	*									
30 min	*									
35										
40										
45										
50										
55										
60 min										
65										
70										
75										
80										
85										
90										
95										
100										
105										
110										
115										
120 min										

Now complete the summary table on the top of the next page to determine the location of each molecule at the following three time points. Do this by adding up data in each column up to the relevant time point. For example the * box above should have the total of the six * boxes from the first table. Submit the results of the second table to your instructor. These data will be used to generate a class model.

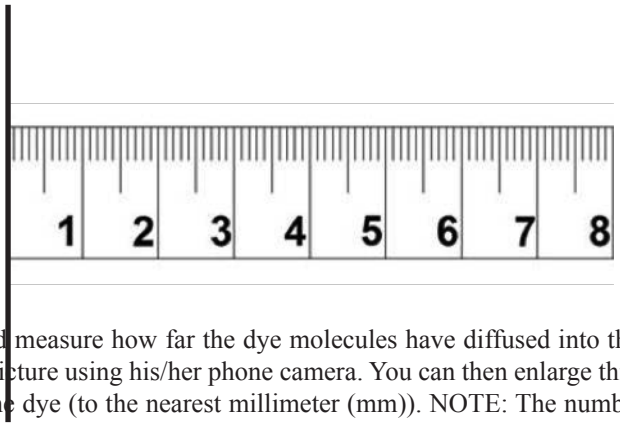
Molecule	1	2	3	4	5	6	7	8	9	10
30 min	*									
60 min										
120 min										

Part 2

At your table you have a petri dish half full of a semi-solid gel and a tube containing a dye solution. We will use this to test our model and determine the rate dye molecules diffuse into the gel. This is similar to the way diffusion occurs across a cell membrane or numerous other biological examples, but has the benefit of being visible without the need for complex imaging tools. Today we will record three measurements: the distance dye molecules move over 30, 60 and 120 minutes.

READ ALL INSTRUCTIONS BEFORE STARTING ANY PORTION OF THIS LAB.

Place this sheet of paper on one of the back tables. Then place the petri dish on the paper such that the edge of the gel is directly on top of the large black line. The empty side should be toward the left (not on top of the ruler). Carefully fill the empty side with dye solution and cover the dish with the lid. Try to avoid any spills. You should still be able to see the ruler markings clearly through the gel. At this point (time =0) there shouldn't be any dye within the gel – diffusion hasn't yet occurred. This is noted on the table at the bottom of this sheet.



At 30, 60 and 120 minutes remove the lid and measure how far the dye molecules have diffused into the gel. To make this easier, one member of the group should take a picture using his/her phone camera. You can then enlarge this image to help you determine the location of the leading edge of the dye (to the nearest millimeter (mm)). NOTE: The numbers on the ruler denote each centimeter (cm).

Time	Distance dye has moved into the gel
0	0mm
0 min	
60 min	
120 min	

Materials

Supplies & Equipment

For Each Lab Group:

- Three petri dishes filled with 2% gelatin solution (prepare night before, then remove ½ of gel just before start of lab; see Fig. 3)
- Fifty-ml colored water made with up to a 1:1 solution of food coloring and water (we used green, any dark color should work, however all groups should use the same color or the size of the dye molecules will influence the rate of diffusion producing more variability)
- Transfer pipette
- Calipers or printed ruler (see Fig. 3)

Time Frame

- Approximately 3-4 hours total time. This is scalable depending on length of diffusion demonstration and depth of coverage on modeling.
- The lab was designed to be presented over two lab periods. At Centenary College, where the activity was developed, the BIO 101 course is structured in a studio format, in which lecture and lab materials are presented in mixed sessions consisting of two, 165 min sessions each week (Leuck and Butcher, 2014 for details). The modeling component is introduced at the end of one lab period (~15min) and a homework assignment (see **Student Outline**) is given. The next lab period the wet-lab portion is run over a 3 hour period with additional lecture/lab material running in parallel. In our lab we use this time to finish the discussion on modeling, generate a model from the classes' data, and present additional lecture materials on relevant biological examples (i.e. ion diffusion).

Procedure

Following an introduction to diffusion, each student is given as a homework assignment the task of generating 240 data points by flipping two coins. Each outcome (illustrated in Fig. 1) represents positive movement into the gel, negative movement away from the gel, or no net movement (the dye moved along the interface). The handout we used is included in the **Student Outline** section.

The following lab, we ask students to set up the "wet lab" portion of the project at the start of the period. This provides 2-3 hours of time for the dye molecules to diffuse. The procedure for this set up is included in the **Student Outline** section and a representative image can be found in Figure 3. Once colored water has been added to each dish, we ask students to submit the summary data (i.e. the data summary at 30, 60, and 120 minutes found on the second table of the handout) to the instructor who then generates a graph of the locations of the modeled dye molecules. This graph serves as a prediction for where dye molecules will be located at a given time point. Note that the model described in Figure 2 only includes perpendicular movement into the gel. This simplifies the modeling activity as it doesn't track of dye molecules parallel to the interface. Sample models that were generated from approximately 200 data points are shown in Figure 2. Once the class graphs are generated, we ask students to discuss what the data is telling them. This can be accomplished either as a single group session, or in a think-pair-share format depending on the time allowed. Students are often confused by what the model shows them about the leading edge of the dye. For example, in the 30 min graph (Fig. 2), a small number of molecules are predicted to reach the 2mm point. Some students will claim that this means the leading edge will be clearly discernable at that point in the gel. In reality, such a small number of molecules will likely be invisible. The green bars on each graph are used to help

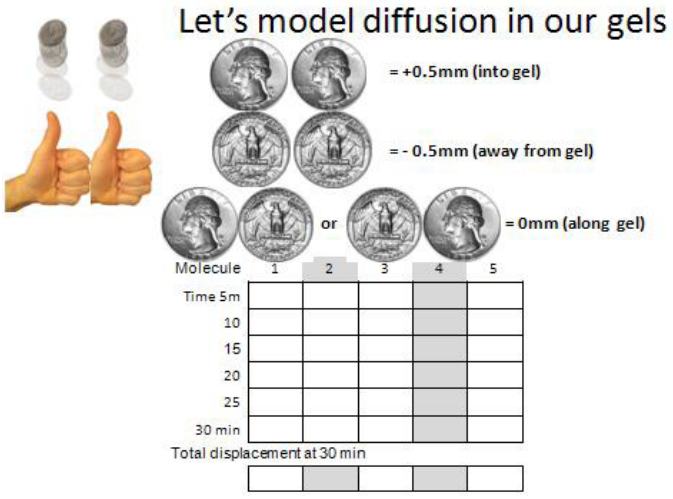


Figure 1. Graphic used to illustrate the rules of the coin flip model. Note in this examples, each student is only asked to generate 30 data points. In the actual homework assignment (see Student Outline) they are asked to generate 240.

students appreciate that a gradient of dye molecules will occur rather than a sharp edge. We ask each group to use the graphs to predict where the visible leading edge of the dye will be for each time point. We then discuss with the students how these predictions are really hypotheses that the model allows us to generate.

At the 30, 60 and 120 time points, we ask students to observe their dishes and note where the visible leading edge is actually located. This is often problematic (Fig. 4) for students and they should be repeatedly cautioned to avoid bumping the dish (or table). To help visualize the edge, we ask each group to capture an image using their cell phone cameras. While not every student will have such a device, at least one per group is virtually guaranteed. These images can then be enlarged on their cell phones to help with this determination. After all data points have been observed, we ask students to compare their predictions from the model and the actual observed measurements. Typically, this will produce some discrepancies which we use as discussion points. These may include discussions of limitations of the model or proposals to make it more accurate. More advance classes can then follow up the lab with lecture materials on the actual equations that are used to model diffusion. In our case, the latter was left for inclusion in a course in physics many of the students would take in subsequent years.

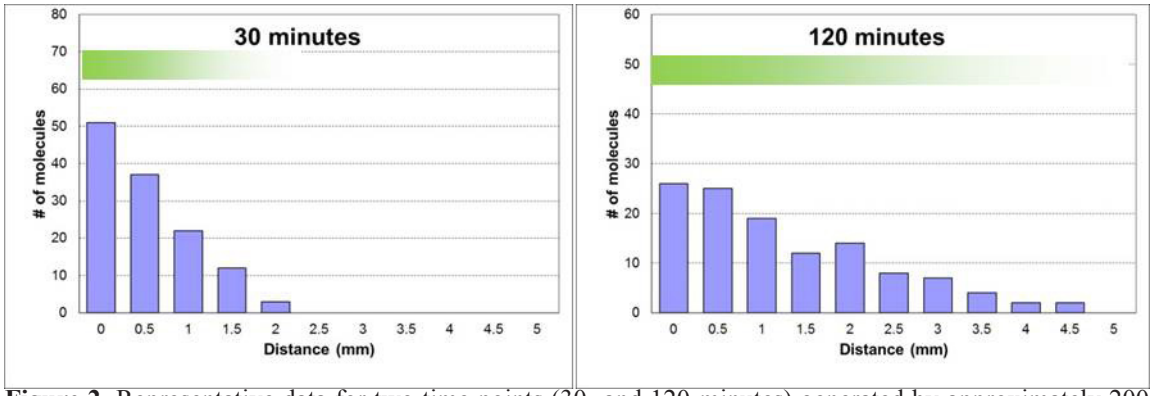


Figure 2. Representative data for two time points (30- and 120-minutes) generated by approximately 200 coin flips. As noted in the panel at left, by the 30 minute time point, most of the dye molecules are still located at or near the interface. By 120 minutes, they have diffused several millimeters into the gel. The green bars above each graph represent how the data might be visualized in terms of the color of the leading edge of the dye. Only positive movement (into the gel) is displayed in these graphs.

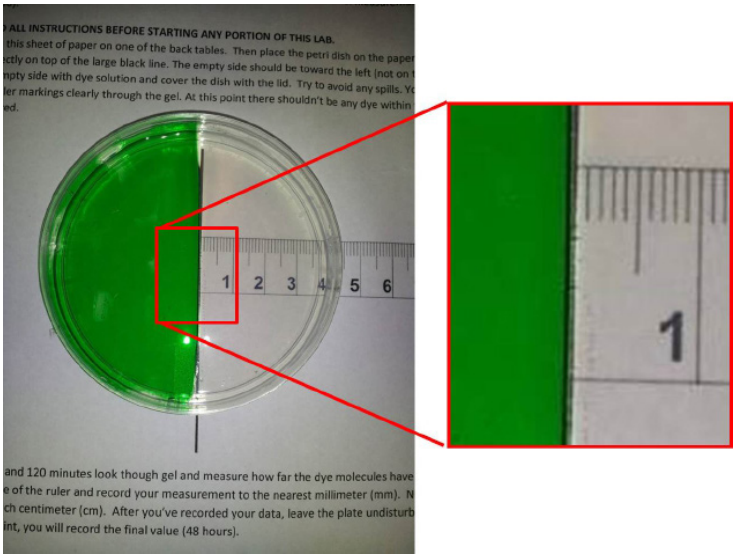


Figure 3. Representative image of gel set up. A single petri dish was filled with a 2% gelatin solution and allowed to set overnight. The following day, half of the gel was removed using a scalpel and flat spatula. The empty side of the dish was then filled with colored water (green side in above image) and the entire dish placed on the paper ruler (see Student Outline). To simplify viewing, students were asked to capture an image of the interface using their cell-phone camera. A representative enlargement of one such image is highlighted at right.

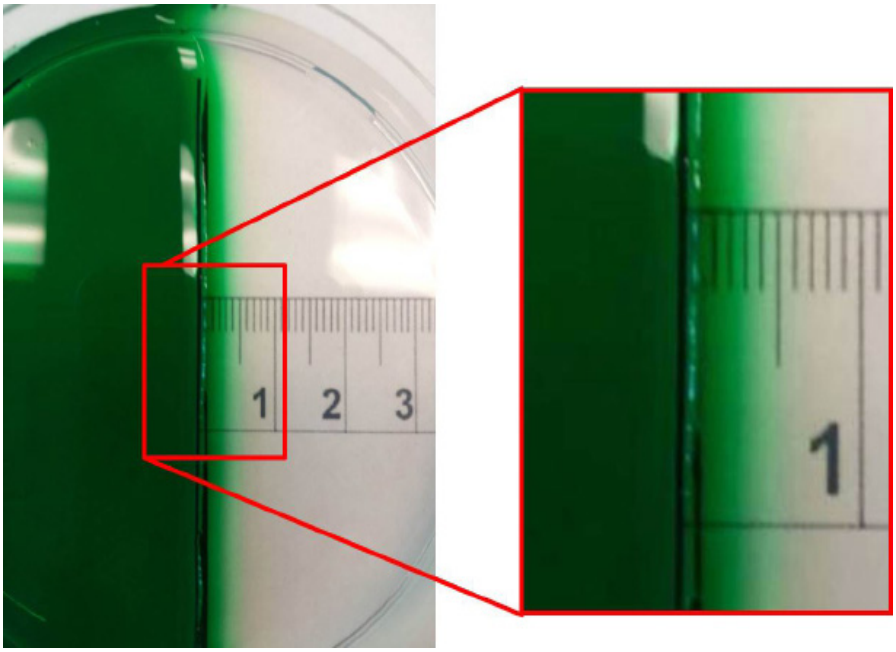


Figure 4. Representative image of gel after 120 minutes. By this point, the green dye has diffused into the gel several millimeters. The enlarged image at right illustrates how the leading edge of the dye is not sharply delineated. Each group of students is asked to discuss where they feel the leading edge is located and then reach a consensus measurement. This variability provides a point of discussion when compared to the model.

Notes for the Instructor

Figure 3 illustrates the typical set up for the "wet lab" portion of the procedure. Note that the gelatin is in contained in the half of the dish placed on top of the paper ruler. To save time, we chose to prepare the gels in advance. Once set, the gelatin can be easily removed from the petri dish using a scalpel to cut a straight interface, then carving out the gel using a flat spatula. This process works best if the gels are cooled in a refrigerator for at least several hours as the 2% solution is fairly soft at room temperature. We also found that students often have difficulty visually determining where the interface of the dye is located. As noted above, we ask students to capture an image using one of the numerous cameras most of them carry (cell phone, tablet, etc). This image can then be enlarged on the device providing greater clarity of the edge.

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